# STUDIES ON THE OXYGEN REQUIREMENTS AND HATCHING MECHANISMS OF THE DOMESTIC FOWL

by

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The oxygen requirements of the chick embryo have been found to increase by at least 100% during hatching. This rise is probably a result of ventilating the newly functioning lungs and maintaining the body temperature at its pre-hatching level. Sustained homeothermic responses become evident only at the moment of escape from the shell membranes.

There are two essential phenomena in the termination of the embryonic existence. The first is the initiation of pulmonary respiration which appears to result from a high partial pressure of carbon dioxide in the blood stimulating the respiratory centres. Active hatching is the second essential phenomenon and is probably stimulated by an increase in the rate of thyroid hormone secretion.

The body temperature of the chick rises after hatching, and is probably a result of the progressive replacement of the yolk with actively metabolizing tissues. There is also a rise in the metabolic rate at this time.

Both the absolute oxygen requirements and the metabolic pattern of the growing fowl may be affected by the diet. There is a difference in the response to the diet according to the sex. A period of almost constant oxygen uptake was consistently noted and appeared to be virtually independent of the diet although high levels of dietary protein tended to reduce this depression in the metabolism.

At 4 weeks of age a large fall in the metabolic rate was found, especially in RIR x IS chickens. The significance of the fall is not known, but a fundamental change in the physiology of the fowl is indicated.

# PART I

## Introduction

## I.1 Introduction

Studies on the physiology of the avian embryo are greatly facilitated by its separate existence from the parent. In spite of this, however, comparatively little work has been carried out upon this most complex phase of ontogenesis. Only a few of the physiological changes that occur at hatching in the domestic fowl, Gallus gallus domesticus (L), have been determined. It is known that the metabolic rate doubles and that the newly-hatched chick, if not a complete homeotherm, is at least an advanced heterotherm. But the factors involved in the termination of the embryonic existence, the time at which thermoregulation appears and the cause for the greatly increased metabolic rate have not been fully investigated.

Similarly the body temperature of the chick has been shown to rise after hatching, but the cause of this rise is not known, nor has it received much attention. Little is known of the effect of temperature or diet on the gaseous metabolism of the fowl, especially for the period between hatching and sexual maturity.

In this thesis the problems outlined above have been investigated using oxygen uptake as the basis of these investigations. In the section dealing with the hatched fowl, work has been limited to the unstarved bird since there has been only one investigation in recent years into the oxygen requirements of the fowl in this nutritional state.

## I.2 Definitions

New or more rigorous definitions have been required in dealing with the hatching embryo. The termination of the embryonic existence, commonly termed hatching, may be satisfactorily divided into several clear cut periods, characterized by certain changes:

1. Full-term embryo

This term is used to describe the chick embryo which is ready to hatch but which has not begun to breathe. Since all embryological divisions have taken place the term foetus may be substituted for "embryo", although by convention the latter is most commonly used.

2. Parafoetal period

This is the period in the life history of the chick when it is dependent upon both the chorio-allantois and the lungs for gaseous exchange. During this period the chick may be termed the parafoetus. (This definition is an extension of that put forward by Romijn & Roos, 1938.)

3. Pipping

Movements of the head of the chick lead to the fracture of the shell at one point. If this fracture is followed by a quiescent period (see below) then the action may be termed pipping.

4. Quiescent period

The quiescent period is that period between pipping and the onset of active hatching when the chick exhibits little or no movement.

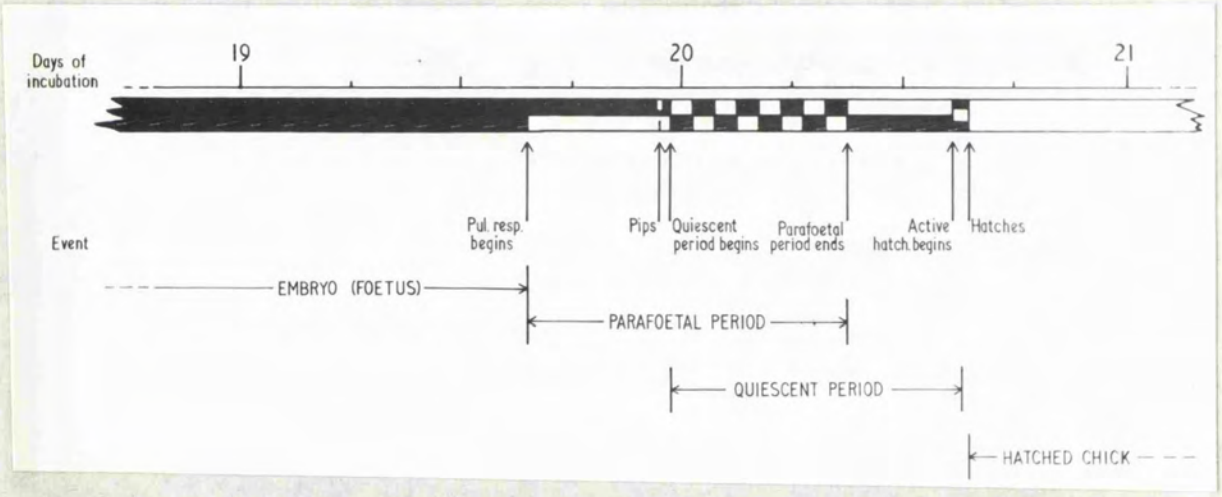


**Fig. 1.** A diagrammatic representation of the major physiological events that occur during the termination of the embryonic existence. The times are approximate.

5. Active hatching

The quiescent period is terminated when the chick commences to break down the shell in order to escape from it. This physical act is defined as active hatching.

The sequence of these defined events is shown diagrammatically in fig. 1.

In expressing the oxygen consumption of the fowl the term metabolic rate describes oxygen uptake in millilitres per gramme per hour (ml.$O_2$/gm/hr) whilst absolute oxygen requirements refers to oxygen uptake in millilitres per hour (ml.$O_2$/hr).

| Author | Breed | °C | Days of incubation | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | 5 | 10 | 12 | 17 | 19 | 21* |
| Pott & Preyer (1882) | ? | 37-39 | — | 3.2 | 7.0*** | 15.5 | 15.7 | — |
| Hasselbalch (1900) | ? | 38.0 | 0.64 | 3.7 | 9.0 | 15.9 | 14.7 | — |
| Bohr & Hasselbalch (1903) | ? | 38.0 | 0.48 | 2.4 | 4.9 | 15.7 | 19.7 | — |
| Romanoff (1941a) | WL | 37.5 | 0.52 | 3.2 | 6.1 | 17.3 | 18.2 | 40.0 |
| Giaja & Jovanoic (1950) | WL | 38.3 | — | 4.0 | 8.0 | 17.0 | 18.0 | 44.0 |
| Romijn (1951) | NHB | 39.5 | 0.57 | 3.9 | 10.0 | 23.5 | 22.4 | 59.8 |
| Beyer (1952) | BC | 38.0 | 0.44 | 5.2 | 8.9 | 18.4 | 20.6 | 43.8 |
| Romijn & Lokhorst (1960) | NHB WL | 37.7 | 0.92 | 4.8 | 11.6 | 23.8 | 22.5 | 55.1 |

*Hatched. ***13 days incubation. WL = White Leghorn; NHB = North Holland Blue; BC = Barred Columbian.

Table 1. Oxygen consumption during embryonic development: a comparison of published results. Figures in ml/hr at STP.

## I.3 Review of literature

### I.3.i Gaseous metabolism of the developing chick embryo

#### a) Oxygen consumption and carbon dioxide production during incubation

It is generally recognized that the single egg is the best experimental unit for studies on the gaseous metabolism of the hatching embryo, since if more than one embryo were used, fundamental changes in the metabolism of one might be masked by a different metabolism of another. However, at the beginning of incubation, when the absolute volumes of gases involved in respiration are very small, gross inaccuracies must be expected unless very sensitive techniques are used.

Baudrimont & Martin-Saint-Ange (1847) showed that the chick embryo required oxygen for development. They measured oxygen uptake, carbon dioxide and water production of the embryo at several stages of incubation, but their results, like those of Baumgartner (1861), proved subsequently to be quite inaccurate and are therefore, of historical interest only. Pott & Preyer (1882) conducted similar experiments, calculating the oxygen consumption indirectly and obtained figures of the same order as those of later workers (table 1). Their data show that the oxygen uptake and carbon dioxide production rose rapidly between the tenth and thirteenth day and then became fairly constant from the seventeenth to the twentieth day of incubation. Pott (1883), Bohr & Hasselbalch (1900) and Hasselbalch (1900) confirmed these findings, whilst the latter author showed that

| Author | Breed | °C | Full term | A* | P* | H* | H+1 day |
|---|---|---|---|---|---|---|---|
| Lussana (1906) | ? | 37.0 | 22.0 | 24.1 | — | — | 58.8 |
| Romanoff (1941a) | WL | 37.5 | 18.0 | — | — | 30.0 | 40.1 |
| Giaja & Jovancic (1950) | WL | 38.3 | 18.0 | — | 33.0 | 44.0 | 54.0 |
| Romijn & Lokhorst (1956) | ? | 38.0 | 36.1** | — | 36.8 | 45.1 | — |
| Visschedijk (1962a) | WL | 37.8 | 26.4 | 36.0 | 41.0 | 33.1 | — |
| | RIR | | 22.2 | 24.0 | 28.5 | 32.6 | — |
| | WLxRIR | | 23.5 | 24.1 | 43.4 | 51.6 | — |

*A = air breathing; P = pipped; H = hatched. **Assuming thermal equivalent of oxygen to be 4.686.

WL = White Leghorn; RIR = Rhode Island Red.

**Table 2.** The oxygen requirements of the hatching chick embryo: a comparison of published results.

Figures in ml/hr at STP.

the respiratory quotient during the greater part of the incubation period was indicative of a fat metabolism. He found that carbon dioxide production during the first three days of incubation fell progressively. Romijn (1962) has shown that this is due to the evolution of physically bound carbon dioxide from the shell itself.

The carbon dioxide production curve for the whole period of incubation was re-examined by Atwood & Weakley (1924). They found that there was no change in the rate of production up to the sixteenth day but confirmed the findings of earlier authors that the production of carbon dioxide was constant from that time to the twentieth day. The results of Murray (1925a) refuted those of Atwood & Weakley (1924) and confirmed those of Bohr & Hasselbalch (1900, 1903) by showing that there was a definite increase in the rate of carbon dioxide evolution from the eleventh day of incubation.

Lussana (1906) was the first author to note that there was a large increase in the oxygen consumption of the hatching embryo. He found that it trebled or even quadrupled between pipping and the first day after hatching and was accompanied by a rise in the respiratory quotient from 0.68 to 0.84. Romanoff (1941a) confirmed that the oxygen consumption of the chick doubled at hatching, whilst Giaja & Jovancic (1950) found that there were two distinct parts to this rise, one at pipping and the other at the time of the escape from the shell. According to Romijn & Lokhorst (1956) the rise at pipping - from 36.1 ml/hr to 36.8 ml/hr - was insignificant but Visschedijk

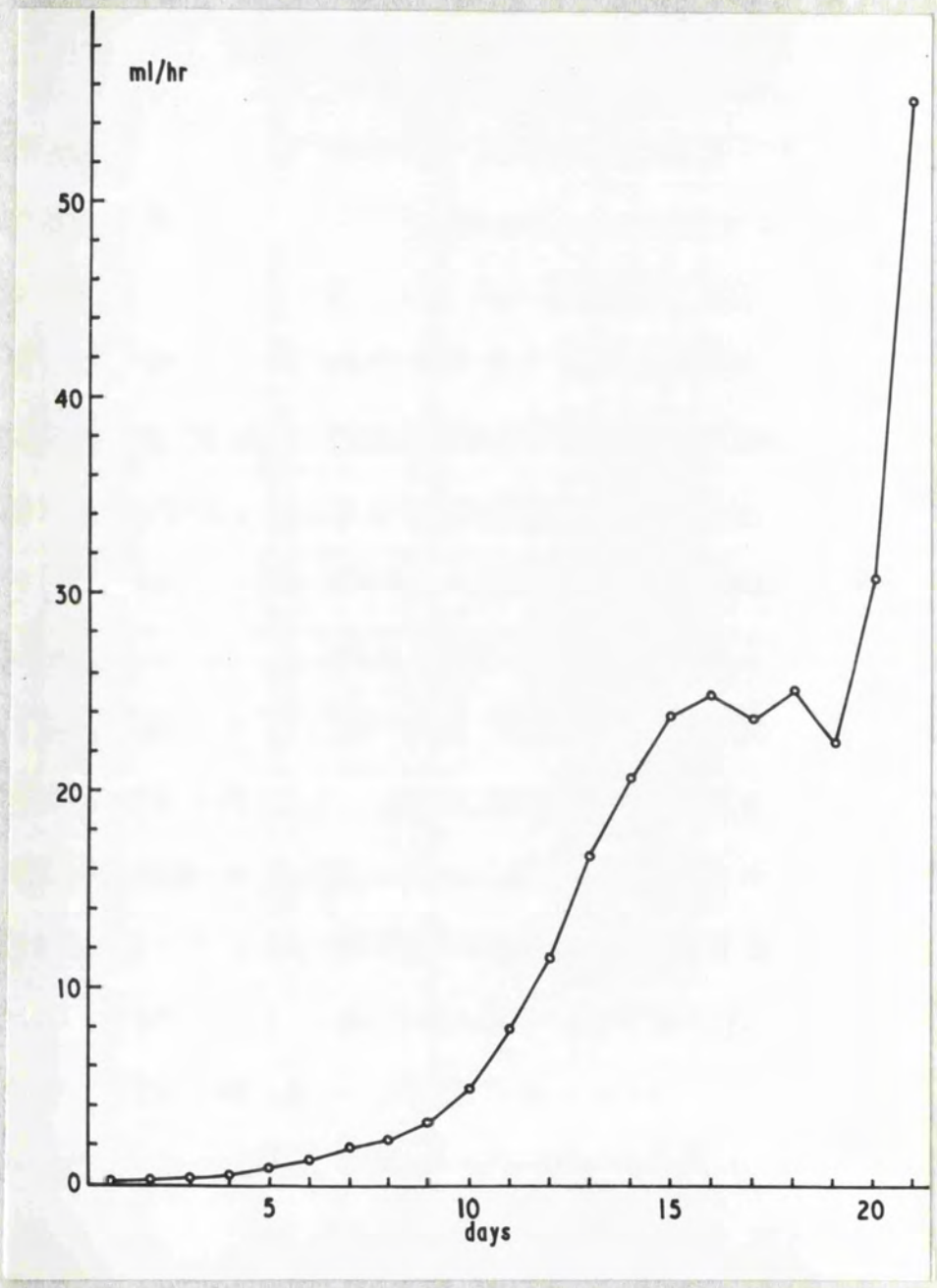


Fig. 2. The oxygen requirements of the developing chick embryo. Data from Romijn & Lokhorst (1960) and Romijn (1961).

(1962a) confirmed that there was a distinct rise in the oxygen requirements of the chick following pipping (table 2). Visschedijk further noted that there was a slight increase in uptake when pulmonary respiration was initiated.

The oxygen requirements of the developing embryo are, according to Romijn (1951) and Romijn & Lokhorst (1960), greater than other workers have found (see tables 1 and 2). It will be noted that the oxygen requirements are similar for the four breeds that have been examined and therefore this discrepancy cannot be of genetical origin but perhaps it is a result of employing more sensitive techniques. The oxygen uptake curve of the embryo is shown in fig. 2, the data for which have been taken from Romijn & Lokhorst (1960) and Romijn (1961). The essential features of this curve have certainly been known from the beginning of this century. It will be seen that the rate of oxygen uptake first increases at the tenth day of incubation. Oxygen uptake becomes relatively constant between the fifteenth and nineteenth day and then doubles within a day during which time the chick hatches. As the respiratory quotient is virtually constant throughout incubation, the curve for carbon dioxide production will be similar.

b) <u>The mechanisms controlling the changes in gaseous metabolism</u>

The increase in the rate of oxygen uptake by the embryo at about the tenth day of incubation has been shown to be due to the commencement of the secretion of thyroid hormones (Sun, 1932; Hansborough &

Khan, 1951; Romijn, Fung & Lokhorst, 1952; Stoll & Blanquet, 1953; Carpenter, Beattie & Chambers, 1954; Maraud, Stoll, Macario & Blanquet, 1954).

Changes in the metabolism after the fifteenth day of incubation are very imperfectly understood. In spite of a constant oxygen uptake the embryo continues to grow during this period (Murray, 1925b; Romijn & Lokhorst, 1951) although the growth rate is reduced (Romanoff, 1929; 1941a; 1960). The observations of Romijn et al. (1952) suggest that oxygen uptake at this time may also be influenced by the thyroid gland, since embryos treated with thyroxine were not found to exhibit this characteristic levelling out of oxygen consumption. This is supported by the work of Beyer (1952), although he found that there was a tendency for the oxygen consumption of embryos treated with thyroxine to become approximately constant from the seventeenth to nineteenth day.

Another explanation for the constant oxygen uptake may be that the oxygen requirements of the embryo are equal to, or even exceed, the volume of oxygen that can diffuse across the shell. Although Hufner (1892) made some measurements of the diffusion rates, the only accurate work has been carried out by Romijn. He found, (Romijn, 1950a), that the permeability of the shell increased during incubation. Calculations from his data show that at the beginning of incubation, at a temperature of 30°C and at a pressure gradient of 100 mm water, about 0.12 ml oxygen can diffuse across the membranes of the whole egg each hour, whilst on the twentieth day,

under the same conditions, this increases to 126 ml/hr. He suggested that about three-quarters of the oxygen required by the embryo on the twentieth day could be supplied through the shell covering the air space, and then concluded, erroneously, that "at least 80 per cent of the total respiratory metabolism is established by the small part of the shell, viz. that area covering the air space". Further work, (Romijn, 1954a), showed that the permeability of the shell over the air space was greater than that of the rest of the shell throughout the incubation period. It is not clear whether this difference is real, for Romijn did not state whether he had removed the inner membrane from the calcareous shell. This membrane is not in contact with the shell over the air space and failure to remove it from the rest of the shell would make the two parts incomparable. During the first seventeen days of incubation he found that the volume of oxygen that could diffuse across the air space increased from 0.71 ml/$cm^2$/hr to 50.08 ml/$cm^2$/hr at a temperature of 30°C and at a pressure gradient of 100 mm water, whilst over the rest of the shell, under the same conditions, the volume increased from 0.08 ml/$cm^2$/hr to 32.24 ml/$cm^2$/hr. Thus on the seventeenth day approximately 2400 ml.$O_2$/hr could diffuse across the shell. Since the pressure gradient is much greater in the living system (probably up to 64 mm of mercury) the volume of oxygen that could reach the embryo would be increased by a factor of nine. The maximum requirements of the full-term embryo are unlikely to exceed 40 ml.$O_2$/hr, therefore the

permeability of the shell is no barrier to the supply of oxygen to the embryo at any time during the incubation period.

The causes of the final rise in the oxygen consumption are not precisely understood. The rise which occurs at the onset of pulmonary respiration (table 2) may be partly due to the increased muscular activity. The same explanation cannot be advanced for the second rise which occurs during active hatching since the metabolic rate (ml.$O_2$/gm/hr) is maintained even when the bird rests or sleeps (Giaja & Jovancic, 1950). These authors have suggested that the emergence of thermoregulation, which occurs at this time (Pembrey, Gordon & Warren, 1895; Giaja, 1925; Romijn, 1954b; Romijn & Lokhorst, 1955) may be a cause of this rise. Sun (1932) has shown that the iodine content of the thyroid gland increases dramatically during the first six hours after hatching, suggesting that the thyroid might be involved.

I.3.ii The termination of the embryonic existence

a) The initiation of pulmonary respiration

The change from chorio-allantoic respiration to pulmonary respiration is complex, involving changes in the vascular system and the removal of the fluids filling the respiratory tract. The nature of the pulmonary stimulus is not known, and the work that has been carried out is somewhat contradictory.

Under natural conditions pulmonary respiration is not initiated until the nineteenth or twentieth day of incubation. However it has been established that embryos are able to exhibit respiratory movements

from the thirteenth day (Kuo & Shen, 1937; Windle & Barcroft, 1938). Byerly & Olsen (1931) suggested that "air hunger", i.e. anoxaemia, was the main stimulus, but pointed out that pulmonary respiration could not begin until the fluid in the amniotic sac and the respiratory passage had been absorbed.

The removal of the amniotic fluid begins on the sixteenth day of incubation (Romanoff & Hayward, 1943; Kugler, 1945) as a result of active absorption by the embryo (Wislocki, 1921; Vrbitch, 1924; Taylor & Saenz, 1949). Kuo & Shen (1937) found that true respiratory movements did not occur until the fluid had been removed and that injections of isotonic saline into the amniotic sac depressed the respiratory rate. They concluded that the removal of the fluid, resulting in the drying of the skin, was the main stimulus, but suggested that carbon dioxide might also be involved, for they found that the blood flowing through the chorio-allantois progressively darkened in colour from the fifteenth or sixteenth day of incubation. It may be concluded therefore, that this gas might act through the respiratory centres to initiate pulmonary respiration.

Carbon dioxide, in moderate concentrations, (0.8 - 1.6%), was found to initiate respiratory movements in the full-term chick embryo by Windle & Barcroft (1938). Windle, Scharpenburg & Steele (1938) suggested that hypoxia and hypercapnia were responsible for the initiation of pulmonary respiration because respiratory movements were increased when the diffusion rates across the shell of the air space were reduced by waxing. They concluded that carbon dioxide exerted

its effect through the respiratory centres. However, Romijn (1948) presented evidence that the respiratory centres of the full-term embryo and the parafoetus were insensitive to gaseous changes. He found that the respiratory rate of the parafoetus was unaffected even when it was breathing an atmosphere containing 4-10% carbon dioxide. Furthermore, the respiratory rate of the chick immediately after hatching, when placed in an atmosphere containing 7% carbon dioxide, 12% oxygen and 81% nitrogen, was similarly unaffected. A chick one day old was found to rapidly develop dyspnoea when placed in the same atmosphere. Recently, Visschedijk (1962a) threw doubt on Romijn's results by showing that the parafoetus is sensitive to carbon dioxide and responds by activity of the cervical muscles which resulted in pipping.

If carbon dioxide is involved in the initiation of pulmonary respiration, it seems that the air space may be involved in producing the necessary partial pressure in the blood. The changing concentrations of carbon dioxide and oxygen within the air space have been measured by Aggazzotti (1914), Romijn & Roos (1938), Roos & Romijn (1941) and Romijn (1954a). They found that the concentration of carbon dioxide increased progressively from the third day of incubation to reach about 5% on the nineteenth day. On the twentieth there was a sharp rise, the concentration often reaching 9%. In contrast, the oxygen concentration fell during incubation from 21% to 13-15% on the nineteenth day, and then to 9-13% during the twentieth. The relatively high concentration of carbon dioxide would be more than

sufficient, in Windle and Barcroft's view, to initiate pulmonary respiration. Indeed, if a maximum of 1.6% carbon dioxide is required, then the embryo ought to begin air breathing on the thirteenth day of incubation at the latest.

Both Noyons & de Hesselle (1939) and Romijn (1948) found that embryos with perforated air spaces still hatched normally. This shows that the intact air space is not at all essential for the completion of development and therefore, if carbon dioxide is involved in the initiation of pulmonary respiration, any necessary partial pressure in the blood must be realised by other methods.

A state of confusion exists over the nature of the pulmonary stimulus. At present two theories emerge, although they may be complementary:

1. Hypercapnia and possibly hypoxia stimulate the respiratory centres which in turn initiate pulmonary respiration.
2. The removal of the amniotic fluid from the respiratory tract stimulates pulmonary respiration, possibly through the drying of the skin.

b) <u>The pipping stimulus</u>

A stimulus for pipping has recently been demonstrated and may be distinct from the pulmonary stimulus. Visschedijk (1962a) showed that the time of pipping could be altered by varying the composition of the gas mixture in the air space. This treatment was without effect upon the time of the onset of pulmonary respiration. He found that a high concentration of carbon dioxide, and to a lesser extent,

| Age at injection | Dose µg | Hatch (days) | Growth | Mortality | Metabolism | Reference |
|---|---|---|---|---|---|---|
| 0 | 6-12 | -- | ? | ? | ? | 2 |
| 0 | 0.025 | 1.0-1.5 | ++ | +- | ++ | 4 |
| 0 | 2-16[x] | +- | ? | +- | ? | 5 |
| 0 | 0.1 | 1.0 | ++ | ? | ++ | 6 |
| 0+17 | 2 + 2-4[x] | + | ? | ? | ? | 5 |
| 5 | 0.025[x] | ? | +- | ++ | ++ | 1 |
| 8 | 0.013-0.1 | ? | +- | ? | ++ | 3 |
| 15 | 1.0 | +- | ? | ? | +- | 6 |

++ definite acceleration  +- no effect

\+ slight acceleration  ? not measured

-- definite retardation

1. Hanan (1928). 2. McCartney & Shaffner (1949). 3. Romijn, Fung & Lokhorst (1952). 4. Beyer (1952). 5. Rogler et al. (1959b). 6. Portet (1960).

[x] d,l-thyroxine.

Table 3. The effects of exogenous thyroxine upon the chick embryo. It has been assumed that the l-isomer was used.

a low concentration of oxygen, stimulated pipping. The chick in the parafoetal period must therefore be sensitive to carbon dioxide (cf. Romijn, 1948). Furthermore, the necessary concentration of gases must, in part, be realized through diffusion insufficiencies of the shell of the air space.

c) The hatching stimulus

No direct work has been carried out on the nature of the hatching stimulus. It seems likely that the thyroid is involved for several authors have demonstrated that this endocrine gland is particularly active at hatching (Sun, 1932; Blanquet, Stoll, Maraud & Capot, 1953; Rogler, Parker, Andrews & Carrick, 1959a).

The effects of exogenous thyroxine on the growth and metabolism of the embryo have been investigated by several workers (see table 3) but few have specifically noted any effects on the time of hatching, and those results available are somewhat contradictory, moreover. Beyer (1952), in contrast to Romijn *et al.* (1952), found that development was accelerated and that hatching was advanced by up to one and a half days even though both used similar quantities of thyroxine. Rogler *et al.* (1959b) found that hatching could be advanced only by giving a second injection of thyroxine on or about the seventeenth day in addition to one on the first day.

Portet (1960) found that the injection of 0.1 µg of thyroxine before incubation led to an increased rate of metabolism, which was accompanied by accelerated development and earlier hatching. If, however, 1 µg of thyroxine was injected at fifteen days, none of these

| Age at injection | Dose (mg) | Delay in hatch (days) | Growth | Mortality % | Metabolism | Reference |
|---|---|---|---|---|---|---|
| 0 | 2.0 | 2.5 | -- | ++ | ? | 6 |
| 8 | 0.5-1.0 | ++ | -- | ? | ? | 2 |
| 8 | 0.25 | ++ | +- | ? | -- | 4 |
| 8 | 2.5 | 5.0 | ? | ? | ? | 4 |
| 10 | 1.0 | 3.0 | ? | ++ | ? | 1 |
| 11 | 2.0 | 3-7 | ? | ++ | ? | 5 |
| 11 | 3.0 | | ? | 100 | ? | 5 |
| 11 | 5.0 | | ? | 100 | ? | 5 |
| 11 | 10.0 | | ? | 100 | ? | 5 |
| 7-17 | 1.0 | 10 | ? | ++ | ? | 1 |
| 14 | 0.5-2.0 | ++ | +- | ? | ? | 3 |
| 17 | 1.0 | 1.0 | ? | ? | ? | 1 |

++ definite effect  +- no effect

-- definite retardation  ? not measured

1. Grossowicz (1946). 2. Adams & Bull (1949). 3. Adams & Buss (1952). 4. Romijn et al. (1952). 5. Romanoff & Laufer (1956). Rogler et al. (1959b).

Table 4. The effects of thiouracil and thiourea upon the developing chick embryo.

effects could be detected. The published results have been summarized in table 3.

Goitrogenic substances have given more consistent and conclusive results. Grossowicz (1946) found that if thiourea were injected into the unincubated egg hatching was delayed by up to ten days. One milligramme of this substance delayed hatching by one day if injected on the seventeenth day of incubation, whereas hatching could be delayed by three days if the same amount were injected on the tenth day. He further noted that 10 µg of thyroxine completely neutralized the action of 2 mg of thiourea when both were injected on the seventeenth day. Adams & Bull (1949) found that thiouracil delayed hatching, whilst Adams & Buss (1952) noted that propyl thiouracil had a much greater delaying action upon hatching than methyl thiouracil. Romijn et al. (1952) found that thiouracil reduced the metabolic rate markedly between the tenth and the sixteenth day of incubation, that it had no significant effect upon the growth rate and delayed hatching by up to five days. Rogler et al. (1959b) found 2 mg of thiouracil delayed hatching by 2 days when injected before incubation, and Romanoff & Laufer (1956), also using 2 mg thiouracil, found a delay of 4 days if it were injected on the eleventh day (see table 4).

It is important to note that thyroid activity is regulated by the pituitary gland. Little work has been carried out on the pituitary-thyroid interrelationship in birds. Woodside (1937) and Aron (1939) showed that administration of thyrotrophic hormone to the developing embryo resulted in an intense activity in the thyroid gland. Woodside

(1937) did not state whether the time of hatching was affected when 20 day old embryos were treated. Fugo (1940) however, was able to show that the final development of the thyroid did not occur when the embryo was hypophysectomized.

## I.3.iii The development of homeothermy and a constant body temperature

### a) Homeothermy

Animals that are able to regulate their heat production over a wide range of environmental conditions thereby maintaining a constant body temperature are said to be homeothermic. Romanoff (1941b) incorrectly defined the phenomenon as "the increase in body temperature above that of the environment". Homeotherms may be classified as precocial or altricial, depending upon the time that the homeothermic response emerges. In the former group, of which the domestic fowl is an example, some degree of homeothermy is developed at hatching; in the latter group, homeothermic responses are lacking at hatching but gradually develop later, e.g. the wren, Troglodytes troglodytes troglodytes, (Kendeigh, 1939).

Pembrey et al. (1895) found that the chick embryo was poikilothermic until the twentieth day of incubation. After this time they found that lowering the environmental temperature did not affect the metabolic rate of the embryo and therefore termed this phase a "neutral condition". As soon as hatching had taken place the homeothermic response became evident, and was well developed by the first day after hatching. Giaja (1925) confirmed the existence of the neutral condition in the pipped egg. Henderson & Brody (1927), Henderson

(1930) and Romanoff (1941b) suggested that homeothermy developed gradually from the mid-point in the incubation period. Both Henderson & Brody (1927) and Henderson (1930) found that after the fifteenth day, changes in the environmental temperature had little effect upon the growth rate, and therefore concluded that the homeothermic response appeared on the fifteenth day. Romanoff's work has been invalidated by an incorrect definition (see above) and the techniques of Henderson and Brody cannot be accepted as valid. Moreng & Philips (1950), with no justification whatever, stated that " ... the baby chick behaves like a cold-blooded animal".

Romijn (1954b) and Romijn & Lokhorst (1955) showed that drastic cooling of the nineteen day old embryo (from 38°C to 26°C) resulted in a rise in the respiratory quotient, although the heat production still fell. This they termed "chemical shivering". They were unable to demonstrate the neutral condition, but found that immediately after hatching, although the chick was still wet, the homeothermic response was well developed.

Hoffmann & Shaffner (1950) noted that the thyroids of chicks incubated at 36°C were larger than those of chicks incubated at 39°C, suggesting that there was some response to cold. Tixier-Vidal (1957) carried out a more detailed examination of the thyroids of full-term embryos which had been subjected to cold (27°-28°C) for two or three days and found that the thyroid responded in the normal way by an increase in the epithelial cell height and a decrease in the amount of colloid. Although this physiological response was not sufficient to

increase the metabolic rate, it may be consistent with the elevated respiratory quotient observed by Romijn & Lokhorst (1955). That the thyroid is active in chemical thermoregulation is contrary to the findings of Stahl, Pipes & Turner (1961). These authors reported that the hatched bird had to be subjected to at least 10 days of continuous cold stimulation before any changes in the thyroid secretion rate could be detected.

Complete homeothermy is certainly developed in the domestic fowl by the end of the first week after hatching (Kleiber & Winchester, 1933; Romanoff, 1941b; Randall, 1943; Romijn, 1954b; Romijn & Lokhorst, 1955).

The consensus of opinion, therefore, is that the homeothermic response emerges in the fowl at some time during hatching and is completely developed by the end of the first week of post-embryonic life.

b) Body temperature

From about the tenth day of incubation the body temperature of the embryo exceeds that of the environment (Penjonschkewitsch & Retanow, 1934; Romanoff, 1941b; Romijn, 1954b; Romijn & Lokhorst, 1955, 1956). At hatching the body temperature is about 40°C, as compared with a normal incubator temperature of 38°C. During the days following hatching the body temperature rises progressively and finally becomes constant, at about 41°C, on the fifth day according to Lamoreux & Hutt (1939) and Hutt & Crawford (1960) but on the tenth day according to King (1956). Lamoreux & Hutt (1939) found that the

temperatures of White Leghorns were significantly higher than those of Rhode Island Reds at the end of the first fortnight. There is no sexual difference in the body temperature during the first fortnight of post-embryonic life (Card, 1921; Lamoreux & Hutt, 1939).

The rise in body temperature may not be progressive, for the data of Card (1921), King (1956) and Hutt & Crawford (1960) show that there is a transient fall between the fourth and fifth days after hatching. The cause of the rise has received little attention, but it has been suggested that it is a result of the mass of the bird increasing at a greater rate than its surface area (Kendeigh & Baldwin, 1928; Baldwin & Kendeigh, 1932; Randall, 1943).

A diurnal rhythm in body temperature was first reported by Hildén & Stenbäck (1916). They found that the variation was 0.95°C/day. Heywang (1938) confirmed this and suggested that the diurnal rhythm of the environmental temperature might be the cause of this variation. By maintaining a constant environmental temperature, Wilson (1948) was able to reduce the rhythm to 0.17°C, although he made no mention of the photoperiod. Hildén & Stenbäck (1916) found that the rhythm could be reversed simply by reversing the photoperiod. More recently Bajpai (1962) showed that the diurnal rhythm was unaffected by daylengths of either 16 hours or 8 hours, but that the group receiving 16 hours' daylight had a significantly higher body temperature. It seems more likely, therefore, that it is the degree of physical activity which affects the body temperature.

## I.3.iv Gaseous metabolism of the hatched chick

The confusion arising from the application of indirect calorimetry to avian material is discussed more fully in section I.3.v (page 25). Much of the early work was of this type and the failure to publish the formulae used to transform gas volumes into units of heat makes the work less valuable. Where the formulae were given the data have been reconverted to their original state.

### a) Factors affecting gaseous metabolism

According to Mitchell & Kelley (1933) there was a "dearth of data" available on oxygen consumption, carbon dioxide production and water metabolism of the domestic fowl. Since that time more data have become available but they are often of limited value through the failure of the authors to record the precise conditions of their experiments.

Both the nutritional state of the animal and the environmental temperature have a great influence on gaseous metabolism. Mitchell & Haines (1927b) and Dukes (1937) showed that the basal metabolic rate is realized only after some forty eight hours' starvation. The birds said by Barott & Pringle (1946) to be in a basal state had been starved for only twelve hours and had therefore only reached the post-absorptive state (Hillman, Kratzer & Wilson, 1953). Precise control of the temperature is necessary if the data on gaseous metabolism are to be of any value. In the work of Mitchell & Haines (1927a) the environmental temperature was usually about 21°C, but occasionally it rose to 34°C. Again if the times of temperature changes during the experimental period are noted valid comparisons of the published figures are then

| Author | 0-5days | 2 weeks | 5 | 8 | 12 | Adult |
|---|---|---|---|---|---|---|
| Mitchell & Haines (1927a) | — | — | — | — | — | 16.5* |
| Benedict et al. (1932) | — | — | — | — | — | 15.0-28.0 |
| Kleiber & Dougherty (1933) | 38.0 | 38.0 | — | — | — | — |
| Barott et al. (1936) | 35.5 | — | — | — | — | — |
| Barott & Pringle (1941) | — | — | — | — | — | 22.8-27.7 |
| Barott & Pringle (1946) | 35.0 | 35.0 | 32.2-35.0 | 29.4-35.0 | 26.7-35.0 | 15.5-23.9 |
| Romijn (1950b) | 35.0 | 35.0 | 35.0 | 35.0 | — | 27.0-32.0 |
| Romijn & Lokhorst (1961a) | 32.0 | — | — | — | — | 28.0-32.0 |

* This was the lower critical temperature; Barott & Pringle (1941) recalculated the zone to be 23.9-26.6°C.

Table 5. The zones of thermal neutrality of the starving fowl: a comparison of results.

Figures in °C.

possible.

In view of the possibility that the diet may affect the gaseous metabolism of the bird (see page 23 et seq) it would also be advantageous if the formula of the diet were given.

b) The zones of thermal neutrality

In work upon respiratory metabolism knowledge of the zones of thermal neutrality is of paramount importance. Mitchell & Haines (1927a) determined the lower critical temperature of the adult fowl, but Barott & Pringle (1941), in a re-examination of the data, were able to show the extent of the whole zone. The findings of workers in this field have been summarized in table 5. It is probably important to note that all the determinations were made on birds that had been starved for at least twelve hours.

Benedict, Landauer & Fox (1932), Romijn (1950b) and Romijn & Lokhorst (1961a) have pointed out that feathering is an important factor in determining the zones of thermal neutrality: the zone for the poorly feathered bird is narrower and shifts into higher temperature ranges.

c) The basal metabolism of the fowl

Mitchell & Haines (1927b) found that forty eight hours' starvation was necessary to bring the fowl to its basal condition. This was confirmed by Mitchell & Kelley (1933), Henry, Magee & Reid (1934) and Dukes (1937).

Mitchell, Card & Haines (1926) investigated the basal metabolism of the fowl during the first month of life and found that there was

| Author | Breed | 1day ♂&♀ | 8 ♂&♀ | 15 ♂&♀ | 56 ♂ | 56 ♀ | 84 ♂ | 84 ♀ | Adult ♂ | Adult ♀ |
|---|---|---|---|---|---|---|---|---|---|---|
| Mitchell et al. (1926) | WPR & WL | 1.17 | 1.79 | 1.72 | — | — | — | — | — | — |
| Mitchell et al. (1927) | WL | — | 1.78 | — | — | — | — | — | 0.68 | 0.67 |
| Mitchell & Kelley (1933)* | WPR & WL | 2.39 | — | — | — | — | — | — | 1.01 | 1.08 |
| Nichita & Mircea (1933) | Dwarf | — | — | — | — | — | — | — | — | 0.58 |
| | Transylvan | — | — | — | — | — | — | — | — | 0.80 |
| Barott et al. (1936) | ? | 1.25 | — | — | — | — | — | — | — | — |
| Dukes (1937) | PR | — | — | — | — | — | — | — | — | 0.70 |
| Barott et al. (1938) | RIR | — | 1.30 | 1.39 | 0.93 | — | 0.80 | — | 0.78 | — |
| Barott & Pringle (1941) | RIR | — | — | — | — | — | — | — | — | 0.65 |
| Perek & Sulman (1945) | WL | — | — | — | — | — | — | — | — | 0.45 |
| Barott & Pringle (1946)** | RIR | 1.25 | — | 1.40 | — | 1.10 | — | 0.89 | — | 0.70 |
| Romijn (1950b)*** | NHB | — | 2.36 | — | 1.95 | 1.50 | — | 1.11 | 0.50 | 0.50 |
| Romijn (1950d) | NHB | — | — | — | — | — | — | — | 0.53 | 0.53 |
| Washburn & Siegel (1963)** | WR | — | — | — | 0.89 | 0.89 | — | — | — | — |

* Only $CO_2$ was measured by the authors; $O_2$ was calculated by assuming RQ to be 1.00.
** Chickens starved for 12 hr only. *** Assuming thermal equivalent of $O_2$ to be 4.686 (Romijn, 1950d)

Table 6. A summary of published findings on basal metabolic rates of fowls (ml.$O_2$/gm/hr).

little difference in the oxygen requirements of White Leghorns and White Plymouth Rocks. This was confirmed by Mitchell & Kelley (1933). Breed differences were first noted by Nichita & Mircea (1933) (see table 6). In a more complete study of the metabolic pattern, Mitchell, Card & Haines (1927) found that the basal metabolic rate (ml.$O_2$/gm/hr) of White Leghorns reached a maximum on the eighth day after hatching although the oxygen consumption in millilitres per square metre of the body surface continued to rise until five weeks after hatching. Thereafter the metabolic rate fell and became constant from the tenth week. They also found that the adult male had a higher metabolic rate than the adult female, and that castration led to a reduction in the rate, although not always immediately. Mitchell & Haines (1927b) and Romijn (1950b, d), however, were unable to confirm that males had a higher metabolic rate than non-laying females. Gerhartz (1914), Mitchell & Haines (1927b) and Dukes (1937) found that the laying hen had a higher rate than the non-laying hen.

Studies on the gaseous metabolism of the fowl under accurately controlled environmental conditions were initiated by Barott and his associates. Barott, Byerly & Pringle (1936) determined the oxygen consumption, carbon dioxide and heat production of unfed chicks during the first five days after hatching. At the point of thermal neutrality (35°C) the metabolic rates of the chicks were constant throughout the period, whilst those whose yolk sacs had been surgically removed showed a gradual decline in their oxygen requirements. Barott, Fritz, Pringle & Titus (1938) determined the basal metabolic rate of the

growing fowl during the nineteen weeks following hatching, and found that the rate rose progressively to the fifteenth day but then slowly declined, reaching a constant level by about the hundredth day.

Perek & Sulman (1945), in an examination of the thyrogenic theory of moulting, found that whereas there was little seasonal variation in the metabolic rate, it was elevated in the moulting hen. It must be pointed out, however, that a higher respiratory intensity in the moulting hen is no proof that there is an increase in thyroid activity. Benedict et al. (1932), Romijn (1950b) and Romijn & Lokhorst (1961a) have shown that the higher metabolic rate of the poorly feathered bird is a result of the reduced insulation.

The above findings are summarized in table 6. It will be noted that the results are extremely variable.

d) The resting metabolism of the fowl

Little attention has been paid to the metabolism of the unstarved fowl. Brody (1930) measured the oxygen uptake of two breeds during the four weeks after hatching and showed that the metabolic rate rose until the body weight reached 60 gm (10 days old) and then remained constant for the rest of the period of observation. Kibler & Brody (1944) extended this study to include the adult fowl employing more precisely controlled environmental conditions. They found that the metabolic rate during the first month after hatching was virtually constant, but when the birds had attained a body weight of between 300 gm and 500 gm there was a marked, progressive fall. Thereafter it fell less rapidly. They suggested that there was no significant

| Author | Breed | 1 day ♂♀ | 8 ♂♀ | 15 ♂♀ | 56 ♂ | 56 ♀ | 84 ♂ | 84 ♀ | Adult ♂ | Adult ♀ |
|---|---|---|---|---|---|---|---|---|---|---|
| Brody (1930) | WR | — | — | 1.49 | — | — | — | — | — | — |
| | RIR | — | 1.82 | 1.24 | — | — | — | — | — | — |
| Kibler & Brody (1944) | RIR | 1.76 | 1.72 | 1.70 | 1.31 | 1.56 | 0.90 | 0.87 | 0.75 | 0.69 |
| Romijn (1950c) | NHB | — | — | — | — | — | — | — | 0.74 | 0.59 |
| Beattie & Freeman (1962) | Br.A* | 1.38 | 1.60 | — | — | — | — | — | — | — |
| | Br.B* | 1.80 | 2.70 | — | — | — | — | — | — | — |

* Commercial broiler strains.

Table 7. The resting metabolic rate (ml.$O_2$/gm/hr) of the fowl at various ages.

For this table and for table 6 it has been assumed that the measurements were made at temperatures within the zones of thermal neutrality.

sex difference in oxygen requirements. Brody, Funk & Kempster (1932) had previously reported that adult males consumed 25% more oxygen per unit time than adult females, and this was confirmed by Romijn (1950c).

In a recent study, Beattie & Freeman (1962) compared the oxygen requirements of two broiler strains, the first (strain A) had a metabolic rate not unlike that of the White Leghorns studied by Brody (1930) but the second (strain B) had a much higher metabolic rate throughout the period of observation. They found that the metabolic rate increased progressively until the birds weighed 60 gm (strain A) or 70 gm (strain B) and then became virtually constant for both strains during the rest of the observational period of a fortnight. This confirmed the findings of Brody (1930), Brody et al. (1932) and Barott et al. (1938).

Examination of table 7 shows that the only complete study is that of Kibler & Brody (1944).

e) The effect of the diet on metabolism

Much work has been carried out on the effect of different diets on the growth rate (e.g. Davidson, 1956; Baldini & Rosenberg, 1957; Combs, Romoser & Supplee, 1957; Combs, Supplee, Quillin, Blamberg, Donaldson, Romoser & Helbacka, 1958; Rand, Scott & Kummerow, 1958; Mraz, Boucher & McCartney, 1958; Norris, Dam, Nelson & Hopkins, 1959; Beilharz & McDonald, 1959, and Marion & Edwards, 1963) but more fundamental work on the effect of the diet upon the metabolism is limited.

Brody et al.(1932) examined the effects of varying protein levels on the heat production of the growing fowl and found that whilst a diet containing 30% dried skimmed milk (total crude protein 18.9%) gave the best growth rate and the greatest overall heat production per unit time, a diet with 5% dried skimmed milk (total crude protein 12.6%) led to the greatest heat production per unit weight during the first 45 days after hatching but then assumed the lowest rate by 65 days.

Singh & Shaffner (1950) found that birds fed a high energy diet had slightly higher oxygen requirements compared with those of birds fed a low energy diet. Mellen, Hill & Dukes (1954) confirmed this for male chickens and found that the increase in the metabolic rate was statistically significant. They further observed that the diets were without effect upon the females. Mellen and his co-workers were unable to detect any changes in the thyroid weights of either sex. However, the work of Singh & Shaffner (1950) and that of Mellen et al. (1954) has been criticised by March & Biely (1957) on the grounds that the low energy diets were sub-optimal and offered a low plane of nutrition.

March & Biely (1957) found that supplementation of a basal diet with either 12% tallow or 12% herring oil led to different responses by the fowl. Tallow produced a depression in the thyroid weight, but had little effect on the growth rate and no effect on oxygen uptake, whereas herring oil depressed the growth rate slightly, but had little effect on the thyroid weight or oxygen uptake.

Treat, Ferguson, Davies & Cough (1960) found that a diet supplemented with 15% rice oil led to a reduction in the weight of both the thyroid and adrenal glands within 8 weeks.

A difference in metabolic response according to sex has been noted by Mellen et al. (1954) and similar differences can be seen in the work of Kennelly & Maynard (1955). The latter authors found that the males had a higher growth rate if fed a diet containing a high level of fat, whereas the females showed no such increase.

### I.3.v The application of indirect calorimetric techniques to the fowl

The use of indirect calorimetry with avian material has been questioned by several authors (Richardson, 1929; Mellen & Hill, 1955 and King, 1957) mainly on the grounds that theoretically impossible respiratory quotients are characteristic of the developing embryo (Hasselbalch, 1900; Bohr & Hasselbalch, 1903; Lussana, 1906; Romijn & Lokhorst, 1951, 1960) and of the starving fowl (Mitchell, 1927; Nishita & Mircea, 1933; Henry et al., 1934; Benedict & Lee, 1937; Dukes, 1937; Romijn, 1950b, d; Mellen & Hill, 1955; Romijn & Lokhorst, 1961a). Calculations of the heat production by the standard equations are therefore made difficult.

It has been suggested that these low respiratory quotients are a result of either protein catabolism (Henry et al., 1934) or certain biochemical reactions whereby endogenous oxygen is formed, or oxygen is absorbed without production of any carbon dioxide (Adams & Poulton, 1932, 1935; Poulton, 1938a, b; Romijn & Lokhorst,

1961b):

e.g. $3\ C_6H_{12}O_6 \rightleftharpoons C_{18}H_{36}O_2 + 8O_2$

King (1957) showed that the respiratory quotient in the production of chicken urine was 0.735 (based on data from Coulson & Hughes, 1930) but the latter authors' data were criticised by Sturkie (1958) on the grounds that the degree of diuresis had not been assessed. Henry et al. (1934) found that the respiratory quotient of a starving fowl fed albumen fell from 0.69 to 0.65 - 0.66 when catabolism was at its most intense.

In spite of the problem of respiratory quotients below the theoretical minimum, some workers have used direct and indirect methods of calorimetry simultaneously in experimental studies. The embryo has been studied by Bohr & Hasselbalch (1903), Barott (1937) and Romijn & Lokhorst (1960: 1962). Bohr and Hasselbalch obtained a good correlation between the tenth and sixteenth day of incubation but Romijn & Lokhorst (1960) found a good correlation only to the twelfth day. Thereafter both authors reported large discrepancies - actual heat production exceeding calculated heat production (Bohr and Hasselbalch), the calculated figure exceeding the actual figure (Romijn & Lokhorst, 1960). However, Romijn & Lokhorst (1962) found that a good correlation for the whole period of incubation could be obtained by maintaining the relative humidity of the environment at 80%. An explanation of this finding has yet to be offered. The data obtained by Barott (1937) showed a very poor correlation

| Age days | Heat cal/gm/hr Obs. | Cal. | % diff. | Author |
|---|---|---|---|---|
| 1-5 | 6.00 | 5.84 | -2.7 | Barott *et al.*(1936) |
| 1-7 | 5.50 | 5.88 | +6.9 | Barott & Pringle (1946) |
| 14 | 6.25 | 6.52 | +4.3 | Barott & Pringle (1946) |
| 15 | 6.20 | 6.50 | +4.8 | Barott *et al.* (1938) |
| 35 | 5.95 | 6.29 | +5.7 | Barott & Pringle (1946) |
| 56 | 4.97 | 5.37 | +8.0 | Barott & Pringle (1946) |
| 84 | 3.90 | 4.18 | +7.2 | Barott & Pringle (1946) |
| 100 | 3.30 | 3.50 | +6.7 | Barott *et al.* (1938) |
| 365 | 2.75 | 2.87 | +4.4 | Barott & Pringle (1946) |
| Adult | 3.00 | 3.05 | +0.2 | Barott & Pringle (1941) |

**Table 8.** A comparison of the observed and calculated heat production of the starving fowl. The respiratory quotient was above 0.707 in all experiments.

throughout. This appears to have been due to experimental insufficiencies. The results from the hatched bird are compared in table 8, and it will be noted that the correlation is only fair, in spite of the fact that the respiratory quotients were always above the theoretical minimum.

It seems that where information on the heat production of the fowl is required it should be obtained directly, but where the two techniques are used simultaneously then special features of the metabolism may come to light (e.g. the suggested conversion of fat to carbohydrate (Romijn & Lokhorst, 1960, 1961b)). Indirect calorimetry appears to be unsatisfactory at other times and the conclusion of Deighton & Hutchinson (1940) is still valid that "there is clearly much to be discovered about avian metabolism before indirect methods can be considered fully satisfactory for calorimetric work with birds".

## I.4 Objectives of Thesis

The work has fallen into four sections; two concerned mainly with the embryo and two with the hatched chicken.

a) Embryonic respiration

The oxygen consumption and carbon dioxide production of the developing embryo have been very well investigated, but little is known of the metabolic changes that occur at hatching. Giaja & Jovancic (1950) have indicated that research into the changing oxygen requirements of the hatching embryo is necessary. Their paper has been taken as a starting point, and an attempt has been made to provide more detailed knowledge of the oxygen requirements of the hatching embryo.

b) Factors concerned with the termination of the embryonic existence

The factors concerned with initiating hatching are substantially unknown. There appears to be a pulmonary stimulus, the nature of which is a subject of some controversy. It is not known whether this stimulus is also responsible for the escape from the shell membranes. An attempt to determine the nature of the stimulus or stimuli which lead to hatching has been made.

Whilst work was being carried out on this topic Visschedijk (1962a) published a very full account of a "pipping stimulus". His work had been conducted along very similar lines to the present author's, and although the latter's investigations were not so well advanced, similar conclusions had been drawn. In view of this publication researches into the nature of the pipping stimulus *per se*

were discontinued.

c) Development of the homoeothermic response and the constant body temperature

It is well known that the domestic fowl is precocious in its development of homoeothermy, but the exact time at which the response appears has not been determined. This has therefore been investigated.

In view of the disparity between the results of Lamoreux & Hutt (1939), Hutt & Crawford (1960) and King (1956) as to the age at which the body temperature of the hatched chick becomes constant, the whole matter has been reinvestigated, and attempts have been made to correlate the rising body temperature with oxygen consumption.

d) Oxygen metabolism of the unstarved chicken

The gaseous metabolism of the fully fed chicken has received very little attention and in those studies that have been published rigorous control of the experimental conditions has been generally lacking.

Here the effects of the diet on oxygen metabolism of the unstarved fowl during its period of rapid growth have been investigated. All the diets used in these studies were formulated to ensure that they were adequate in all nutritional requirements. At the same time the growth rate, food consumption, thyroid and adrenal weights were determined in an attempt to elucidate how the diets might be affecting the oxygen uptake.

The environmental temperatures at which the oxygen consumption were determined were always controlled to within $\pm$ 0.1°C. However,

the zones of thermal neutrality for the young unstarved chicken are unknown and therefore had to be determined in order that the adverse effects of temperature on the metabolism could be eliminated.

## I.5 Materials and Methods

### I.5.i Animals and their management

a) Birds

A standardized and representative source of animals is necessary for all physiological work. Therefore for each breed examined, a small group of hens (a minimum of 15 but up to 60 hens in later experiments) was selected at random from a larger flock. Two cocks were similarly selected. Work was carried out on a commercial laying strain, a broiler strain, White Leghorns (WL) and a Rhode Island Red - Light Sussex cross (RIR x LS).

b) Management

Incubation techniques and embryos

Eggs were collected daily from the groups and stored for periods of up to one week at 10° - 12°C. They were incubated in a forced draught incubator at an environmental temperature of 37.7° ± 0.2°C and a relative humidity of 47 ± 2% until 70% of the embryos had pipped. The relative humidity was then raised to about 80% for 24 hours and finally reduced to the original level about 6 hours before the chicks were removed from the incubator. The eggs were "turned" hourly during the first 18 days.

Embryos required for the experimental work were usually taken on the eighteenth or nineteenth day. Those required for investigations into oxygen metabolism were placed in the respirometers and maintained at 37.7° ± 0.1°C and a relative humidity of about 60% whilst those used for experiments on the termination of the embryonic existence were

| Ingredient | lb. |
|---|---|
| Ground wheat | 35 |
| Ground barley | 10 |
| Ground oats | 10 |
| Middlings | 15 |
| Fishmeal (66%) | 10 |
| Soyabean meal (44%) | 10 |
| Dried grass meal | 5 |
| Unextracted dried yeast | 2.5 |
| Skimmilk powder | 3 |
| Limestone flour | 1 |
| Manganised flour | 0.5 |
| Choline chloride | 0.1 |
| | 112.1 |
| Vitamin A | 4m i.u./ton |
| Vitamin $B_2$ | 1 mg/lb |
| Vitamin $D_3$ | 1m i.u./ton |
| Estimated crude protein content | 21.0% |
| Estimated metabolizable energy | 1163 kcals/lb |

Table 9. Formula of the standard diet.

kept in a still air incubator at 37.7° ± 0.2°C at a relative humidity of 62 ± 5%.

Hatched birds

All chickens were reared in specially designed wire-floored cages. The room temperature was maintained at 17° ± 1.0°C and supplementary heating was supplied to the chicks during the first three weeks by infra-red heaters (150 watts). Food and water were available ad libitum. The daylength was 14 hours.

The formula for the standard ration is given in table 9. It was a ration with an estimated crude protein content of 21%, and with a metabolizable energy content of 1163 kcals/lb. Where special diets were used, the formulae are given in the relevant section.

I.5.ii Experimental techniques

The determination of oxygen consumption

The classical gravimetric method of determining the oxygen consumption of an animal is that of Haldane (1892). The method, however, is time consuming without any compensatory increase in sensitivity. Carpenter (1928) showed that the minimum period of observation for the adult fowl was two hours, and the period would therefore have to be proportionately longer for younger birds. Recently developed techniques have aimed at speed and simplicity (e.g. Strite & Yacowitz, 1956; Charkey & Thornton, 1959) but these do not incorporate accurate control of the environmental temperature, and are therefore of limited value. Perhaps the best method available is that developed by Bargeton & Krumm-Heller (1949) which is

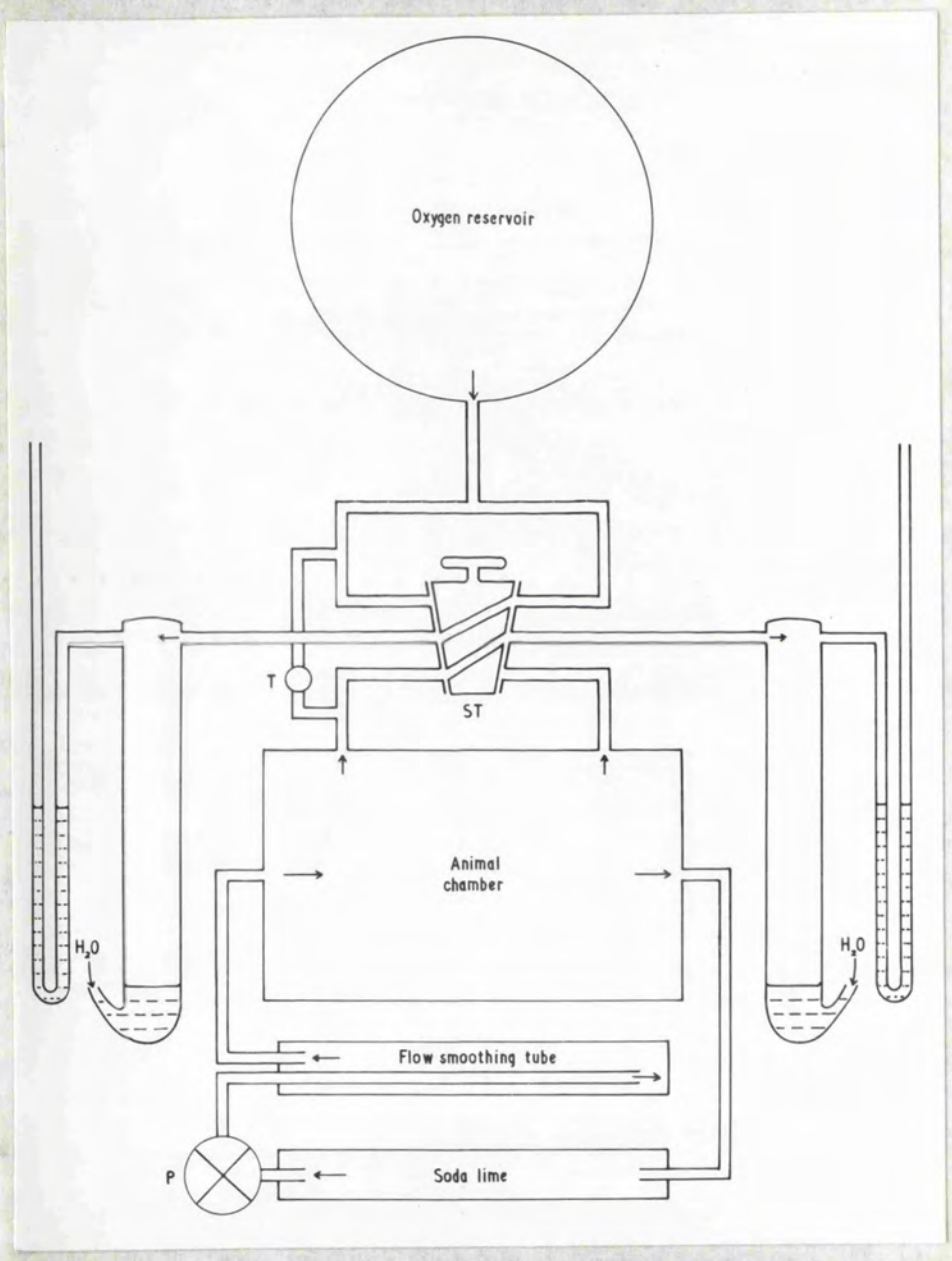


Fig. 3. Diagram of the Bargeton - Krumm-Heller respirometer.

ST = Selector tap; T = Circuit bypass.

both reliable and accurate.

A submersible diaphragm pump (P) activated by an auxiliary pump, moves air round the closed circuit apparatus (see fig. 3). The rate of flow is controlled by varying the speed of the auxiliary pump. The resulting pulses of air are converted into a continuous flow by directing the air through a column of glass wool.

The air passes into the animal chamber and then through a carbon dioxide absorbent (soda-lime) and back to the pump. Two secondary circuits are derived from the animal chamber. Each is connected to an oxygen storage tank and a manometer, the other arm of which is open to the atmosphere. Only one oxygen tank is connected to the main circuit at any time, the other is in communication with an external source of pure oxygen. The tanks are alternately brought into the main circuit by turning the selector tap (ST) through 180°.

The carbon dioxide produced by the animal is absorbed by the soda-lime. As oxygen is consumed there is a gradual and progressive fall in the pressure of the system, and this is registered on the manometer which is in circuit. Oxygen is displaced from the oxygen storage tank into the main circuit by forcing water into the bottom of that tank. When the pressure of the system returns to normal (i.e. atmospheric) the flow of water is stopped. The volume of water forced into the storage tank is measured by a burette and is equal to the volume of oxygen absorbed by the animal. This volume is corrected to standard temperature and pressure (STP).

The second oxygen tank is in connection with an external source

of pure oxygen. This tank can be refilled with oxygen simply by withdrawing the water from the tank into the burette. Since there is a temperature gradient between the room and the experimental environment a certain time must be allowed to elapse before using that tank.

The duplication of the oxygen tanks makes it possible to determine oxygen consumption continuously. When one tank is depleted, the other is brought into circuit and the depleted one is refilled with oxygen. If determinations are to be made intermittently whilst it is desirous to keep the material in the respirometer, then the external source of oxygen can be brought into direct connection with the main circuit by turning tap (T).

The environmental temperature is controlled during the determinations by submerging the whole apparatus in a constant temperature water bath. This is controlled to within $\pm$ 0.1°C.

Two models of this respirometer were used. The smaller model was designed to take animals up to a weight of 300 gm live weight, whilst the larger apparatus accommodated animals up to 3 kg. Here a centrifugal air pump replaced the diaphragm pump and obviated the need for a "flow smoother" tube.

The technique for determining oxygen consumption

The whole apparatus was brought into approximate equilibrium before the animal was placed in the chamber. Once this was done, the chamber was sealed and the apparatus returned to the constant temperature water bath. Equilibration took from 5 to 30 minutes

depending upon the material used and the temperature gradient between the respirometer and the atmosphere. During this time the oxygen requirements of the bird were met by direct diffusion from the external source of oxygen. When the equilibration was complete, an oxygen tank was brought into connection with the chamber by turning tap T. Each oxygen tank was in circuit for a fixed time - from 3 to 30 minutes depending upon the material used. The figures for the oxygen consumption of the bird were averaged and the hourly reading determined. The barometric pressure, environmental temperature, age and weight of the bird were noted.

All determinations were carried out in daylight.

The embryo

The times of pipping and hatching were recorded for each embryo. After hatching had taken place observations were continued for another hour before the weight of the chick was determined. This weight was taken to be that of the full-term embryo; in this way the metabolic rate of the embryo could be calculated. No allowance for the weight of the yolk sac and its contents was made, in spite of Needham's (1932) finding that it is virtually "dead weight".

The hatched bird

Birds were observed singly. After being weighed to the nearest 0.1 or 1.0 gm, depending upon size, the bird was placed in the chamber. No attempt was made to restrain the bird, and it was free to adopt any postural position, since Bartlett (1959) has shown that restrained guinea pigs have a higher metabolic rate than unrestrained, and

Bartlett, Helmendach & Inman (1954), Bartlett, Bohr & Inman (1955), Bartlett & Quimby (1958) and Hahn & Koldovsky (1958) have shown that restraint leads to a fall in body temperature in the rat and cat. During the equilibration period the bird usually settled down and often slept, especially if the environmental temperature was within the zone of thermal neutrality. No food or water was available during the period of observation: thus the resting metabolism was measured.

In experiments on the development of homeothermy, no chick was used more than once since exposure to a low environmental temperature might affect the subsequent metabolic behaviour of the chick (cf. Hahn, 1956).

Calculation of oxygen consumption

Environmental temperature = T°C.

Barometric pressure = P mm of mercury.

Let each oxygen storage tank be used for n minutes.

Then:

Average vol. of $H_2O$ forced into the tanks = average vol. $O_2$ absorbed

= a ml/n minutes

$O_2$ consumption = $\frac{60a}{n}$ ml/hr

$O_2$ consumption at STP = $\frac{60a}{n} \times \frac{273}{273 + T} \times \frac{(760 + P)}{760}$ ml/hr

---

Histological examination of the thyroid glands

The bird was killed by cervical dislocation. Embryos were killed similarly after they had been removed from their shell. The

thyroids, lying at the base of the neck close to the carotid arteries, were dissected out and cleared of parathyroid and fatty tissue. Formol sublimate was used as the fixative. Sections were cut from paraffin blocks at 5 μ, and stained in haemotoxylin and eosin.

Organ weights

In some experiments the weights of the thyroid and adrenal glands were determined. The chickens were killed by cervical dislocation. The thyroids were prepared as for histological examination. The adrenal glands were removed and cleared of any connective tissue. Glands were then blotted on filter paper, and weights determined on a constant load balance (Mettler).

The determination of surface area

The surface area of the egg was measured directly by tracing centimetre squares on to the shell.

Hatched birds were killed with ether and the skin cut along the mid-ventral line from the vent to the base of the skull. The two flaps of skin were opened out, care being taken to prevent stretching, and the outline traced on to squared paper.

Waxing, perforating and aerating the air space

The eggs were candled and the limits of the air space marked. Waxing was carried out by quickly dipping the shell into molten paraffin wax (MP 56°C), care being taken not to cover any part of the shell over the chorio-allantois.

Perforation of the air space was carried out with a dental drill

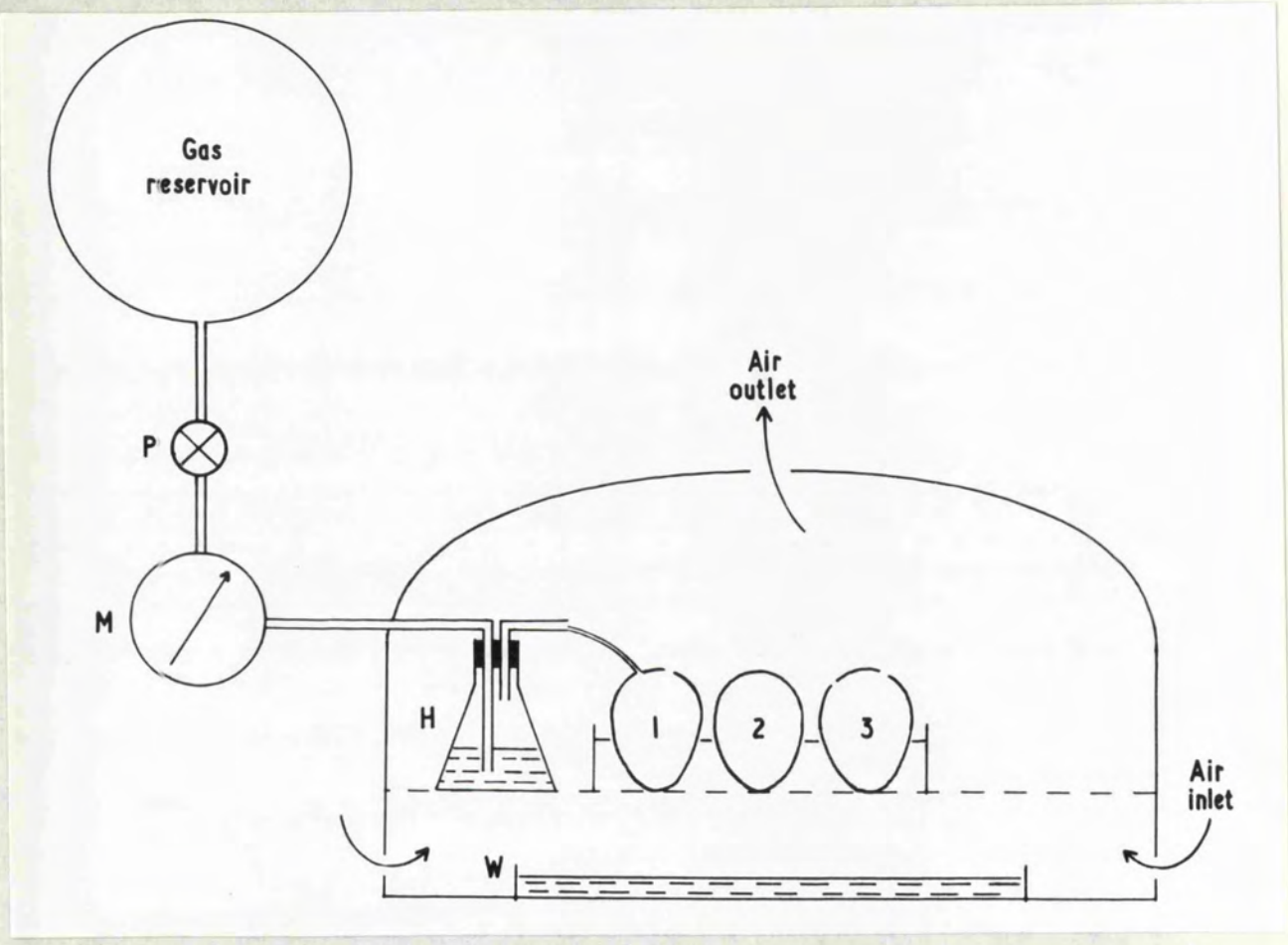


Fig. 4. Apparatus for the ventilation of the air space of the embryo. The gas from the reservoir passes through the pump P and meter M into the humidifier H and thence to the egg. The water tray W maintains the environmental humidity at approximately 62%. Egg 1 = ventilated air space; 2 = normal control; 3 = perforated control.

at the apex of the egg.

The following apparatus was devised for the aeration of the air space. A small pump P, delivering approximately 10 l/min was connected through a wet gas meter (M) to a humidifier unit (H) maintained at a temperature of 38°C. From this humidifier six separate tubes were derived, each passing into the air space of an embryo, and sealed there by paraffin wax. The apparatus is illustrated in fig. 4.

A second hole in the air space of the embryo allowed the gases to escape into the incubator. Assuming the volume of the air space to be 10 ml, the ventilation rate was 160 changes of air/embryo/hr.

The injection of drugs into the air space of the full-term embryo

The volume of fluid injected into the air space was standardized to 0.2 ml. No attempt was made to maintain aseptic conditions, since it was found in preliminary trials that the percentage hatch and the course of the termination of the embryonic existence were unaffected. All solutions were brought to approximately 38°C before injection. Injection was through a small hole made with a dental drill, which was then sealed with paraffin wax.

Triiodothyronine and thyroxine: Sodium-l-triiodothyronine[1] or sodium-l-thyroxine[1] was dissolved in a small volume of N sodium hydroxide, and then diluted to the required concentration with physiological saline.

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1 Glaxo Laboratories Ltd., Greenford, Middlesex.

Thiouracil: 2-thiouracil[1] was dissolved in N sodium hydroxide and saline added to give the required dilution. Control embryos were injected with the same solvent.

Thyrotrophic hormone: Thyrotrophic hormone, of bovine origin[2], was dissolved in saline.

Determination of the onset of pulmonary respiration, pipping and hatching

The time at which the chick (parafoetus) responded by cheeping to a slight mechanical shock was taken to indicate that pulmonary respiration had been established.

The single, point fracture of the shell is characteristic of pipping and was readily observed.

Hatching was judged complete when the chick had escaped from the shell membranes.

The removal of the yolk sac from the chick (lecithectomy)

This technique was originally described by the term "deutectomy" by Adamstone (cited by Sloane, 1936). This term is not accepted, the term lecithectomy being preferred (lekithos = yolk, ek = out, tomeo = cut).

No standard technique for the removal of the yolk sac has yet been devised. Parker (1929), Sloane (1936) and Harvey, Parrish & Sanford (1955) operated through the right side of the chick, whilst

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1 Light & Co. Ltd., Colnbrook.

2 "Thytropar". Armour Pharmaceutical Co., Eastbourne, Sussex.

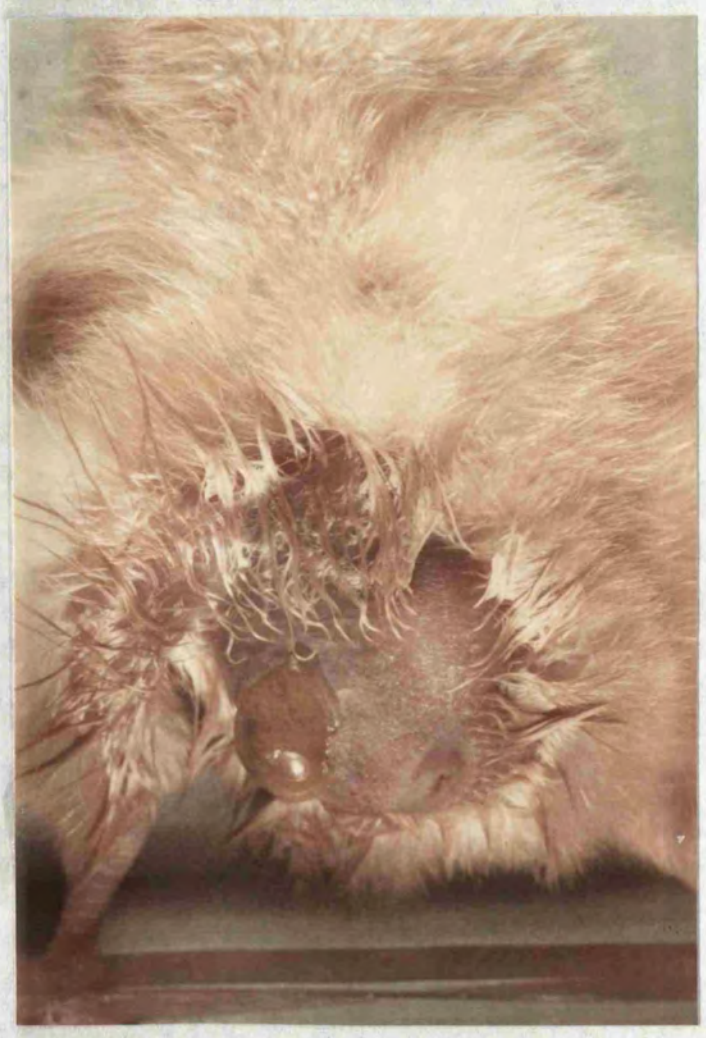


Plate 1. Lecithectomy of the chick - exposure of the yolk sac.

Menge, Moreng & Combs (1951) operated through an enlarged umbilicus.

The technique described here is broadly that of Harvey et al. (1955), but several modifications have been found to be advantageous.

The chick was lightly anaesthetized with ether and the down feathers on the right side of the abdomen up to the umbilicus were removed by plucking. The chick was placed on the operating board on its back, with its legs fastened down to its side in order to keep the body wall taut. Deep anaesthesia was then induced and the operative area swabbed with 70% alcohol. An incision about one half inch long and one half inch to the right of the mid line was made. In this region the peritoneum is thicker and therefore makes subsequent suturing easier. The incision was not made on the left side as this is almost completely filled by the gizzard. Slight haemorrhage may occur, for there is a small blood vessel running transversely across the area, but this could normally be avoided as it is visible through the skin.

When the peritoneum was cut - this wound was less than one half inch long - the yolk sac could be recognized by its yellow or greenish-yellow colour. Often it protruded slightly (plate 1). The sac was seized with blunt forceps and pulled through the wound, care being taken to free the sac from the umbilicus. When the yolk stalk was exposed, a ligature was applied with surgical thread. The stalk was then severed (plate 2). The wound was closed with two or more sutures. Care was taken to ensure that the edges of the peritoneum were drawn together.

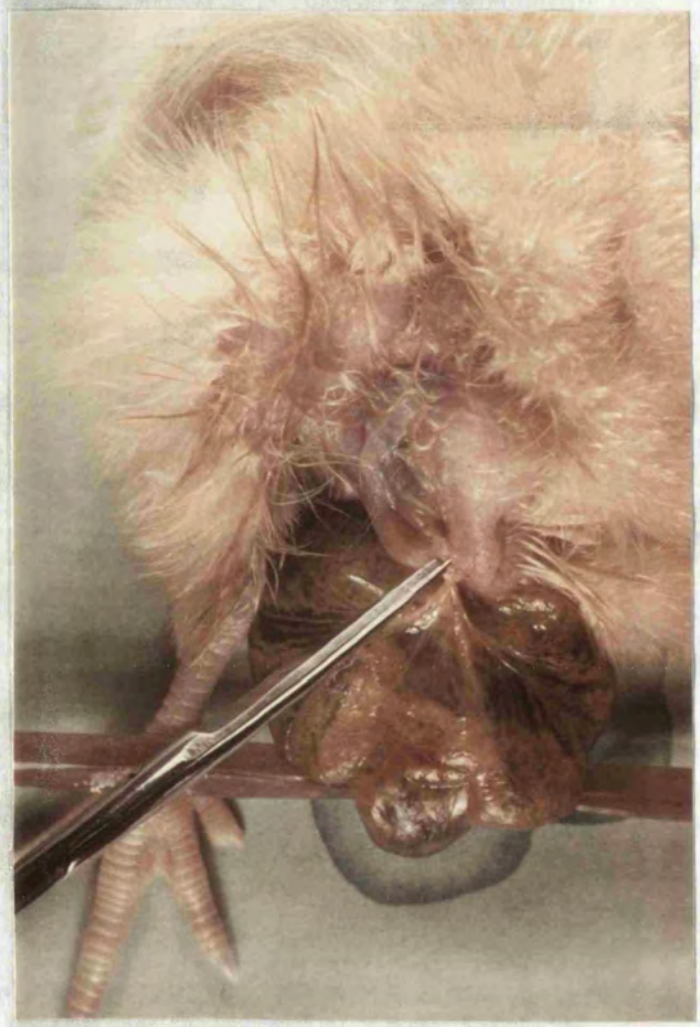


Plate 2. Lecithectomy of the chick - ligation and severance of the yolk sac stalk.

The chick was placed under an infra-red heater which helped to reduce post-operative shock. Later 10,000 units of streptomycin were injected into the thigh muscle to help combat infection. The bird usually recovered within two hours of the operation. The percentage mortality was low, not greater than 3%.

Determination of body temperature

The problems of determining the body temperature accurately, with good repeatibility, have been fully discussed by Lamoreux & Hutt (1939). The technique used here satisfied the requirements they listed.

All temperatures were determined between the hours of 11 a.m. and 11.45 a.m. A gradient in the environmental temperature was maintained in the cages thereby allowing the chick to maintain its optimal body temperature. The room temperature was maintained at 17° ± 1°C.

A clinical half-minute thermometer was used; the depth of insertion was standardized to 15 mm by a tape stop. Insertion into the cloaca and rectum was facilitated by lubricating the bulb of the thermometer with liquid paraffin. The thermometer was regularly sterilized to prevent the transmission of infection to the cloaca.

The birds were selected at random each day. Birds less than one week old were held in the hand in a crouching position and then inverted to allow the thermometer to be inserted. The legs of the birds more than one week old were allowed to hang freely in order to prevent respiratory distress. The body temperature was determined

to within 0.1°C and where necessary the sex was recorded.

Analysis of food

Analyses of food samples to give the crude protein, fibre calcium, phosphorus and moisture content were carried out professionally.

Oil content: 3 gm food samples were weighed out in duplicate. Each was placed in a thimble and subjected to the Soxhlet method of extraction with petroleum ether (40/60).

Extraction was carried out for a minimum of 3 hours. The solvent and the dissolved fatty material were transferred to a weighed flask and the solvent was evaporated off and the oil weighed.

Pentosans: The official method of analysis of the Association of Official Agricultural Chemists (1960) was used.

Reagents: Dilute (1:2 v/v) HCl.

0.7% (w/v) phloroglucinol in dil. HCl.

3 gm of the food sample were distilled with 100 ml dilute hydrochloric acid. When about 30 ml of distillate had been collected, 30 ml of dilute acid were added to the food and distillation continued in this way until 360 ml of distillate had been collected. A solution of phloroglucinol was added to give a final volume of 400 ml. The solution is allowed to stand overnight to ensure complete precipitation of the phloroglucide. The precipitate was collected in a weighed Gooch crucible and washed with distilled water. After drying at 100°C for 24 hours the weight of the precipitate was determined.

Calculation

Weight of phloroglucide = $\alpha$ gm

Pentosans = $(\alpha + 0.0052)0.8866$ gm

Indigestible organic matter

From the data on protein, oil pentosan and fibre contents of the diet the indigestible organic matter could then be determined by the following formula:-

$$IOM = \frac{(\text{Crude protein \%} + \text{Crude fat \%})}{10} + \text{Pentosans \%} + \text{Fibre \%}$$

Statistical methods

Significance test

The significance of the difference between two means $\bar{x}_1$ and $\bar{x}_2$ was determined by the "t" test as given by Chambers (1958). The distribution of t was taken from the statistical tables of Fisher & Yates (1953). The lower level of significance was taken to be $P = 0.05$.

Standard deviation

The standard deviation of the mean was calculated from the equation:

$$S.D. = \sqrt{\frac{S(x^2)}{N} - \bar{x}^2}$$

Analysis of variance

This was carried out by the method of Snedecor (1953).

Calculation of regression equations

The method of least squares was used, as given by Johnson (1950)

to give the equation form:

$$y = a + bx$$

where y = oxygen consumption in ml/hr; a = a constant; b = regression coefficient; x = body weight in gm.

From this the logarithmic form was derived:

$$y = ax^b.$$

This equation was adopted as the standard form of expressing oxygen consumption.

I.5.iii Experimental designs

Embryonic respiration

Whilst many embryos exhibited the same trend, not all the results were suitable for inclusion in graphs. This was usually due to incomplete figures, or the inability to determine some point. At least six embryos have been used to determine each phenomenon. At all times the single egg has been used as the experimental unit, thus removing the possibility of the responses in one embryo being masked by others.

The termination of the embryonic existence

Each experiment involved a minimum of six embryos per treatment, but up to 36 embryos were often used. The time at which the onset of pulmonary respiration, pipping or hatching was exhibited by 50% of the embryos of each treatment was taken as the representative time for that treatment. The experiment was repeated until a minimum of 24 embryos was subjected to each treatment to ensure repeatibility. The mean time was calculated and used in subsequent comparisons.

| | | | | | |
|---|---|---|---|---|---|
| 8a | 18b | 15c | 6a | 11b | 16c |
| 12b | 13c | 5a | 14c | 9a | 10b |
| 24c | 7a | 17b | 22b | 1c | 2a |
| 21b | 19c | 23a | 4c | 20a | 3b |

Fig. 5. A typical experimental randomized block as used in experiments upon factors concerned in the termination of the embryonic existence. a, b and c are the treatments.

The eggs within any experimental block were distributed at random, by numbering each egg as its state of development was checked, and then distributing the eggs in a randomized number pattern. The treatments were determined by a Latin square of the appropriate dimensions. A typical experimental block is illustrated in fig. 5. In this way any positional effects were reduced to a minimum.

Respiration of hatched birds

A minimum of 12 birds from a single hatch was used. Larger groups were occasionally made up by amalgamating the chicks of two or more consecutive hatches from the same parents. In the experiments concerned with the effect of nutrition upon oxygen consumption, each pair of experimental groups was obtained by dividing at random the chicks from one hatch into two equal groups of approximately equal weight.

Effects of diet on the metabolism and certain endocrine glands

Food consumption was measured weekly. A number of birds from each group was selected at random at 3, 5 or 8 weeks of age and the weights of the thyroid and adrenal glands were determined.

I.5.iv Presentation of oxygen consumption data

Embryonic respiration

Preliminary studies showed that it was not possible to plot the results of the oxygen consumption of the embryo against the body weight or the time of incubation if pooling of the results were to be carried out. The following technique was therefore developed.

The times of pipping and hatching were noted, and 1 hour after the chick had hatched its body weight was determined. No correction was made for the weight of the yolk sac. This "hatching weight" was assumed to be the weight of the chick throughout the period of observation. The oxygen consumption of the chick in ml/hr and ml/gm/hr was then calculated and plotted against hours before pipping and after hatching. The period between pipping and hatching was given a variable time scale, i.e. the times at which the two events occurred were fixed reference points on the graph. The distance (time interval) between pipping and hatching was equal to the average time between these two events for the embryos used in that experiment.

This technique allowed the results of several embryos to be plotted together with a good correlation between them (see fig. 6 (page 48)).

Oxygen consumption of the hatched bird

The oxygen consumption (y) was plotted with reference to the body weight (x) in most experiments. Where three or more figures were collected for any particular weight the average was taken and used in subsequent calculations. This enabled the bulk of the data to be decreased with little effect on the value of the regression coefficient.

Methods of expressing oxygen uptake have been extensively discussed (Tanner, 1950; Kleiber, 1950; Chiu & Hsieh, 1960). Chiu and Hsieh, in a study of the metabolism of the rat, concluded that basing the oxygen consumption on body weight (kg), "metabolic

size" ($kg^{0.75}$ or $kg^{0.667}$), or surface area ($m^2$) - the latter three generally calculated from body weight - could lead to errors, especially when comparisons of the oxygen consumption of rats of different weights were made. The problem is difficult to resolve, and in the sections dealing with the comparisons of metabolic rate as affected by the diet, both oxygen uptake based on weight and metabolic rate related to age have been used.

In most of the experiments on oxygen uptake there were more data than could be conveniently included in a table in the text. It was felt, however, that these data should be available. They were therefore brought into as concise a tabular form as possible and placed together in a separate section - Part V. These tables are identified in the text by Roman numerals; tables necessary to the text have been given Arabic numerals and appear in the relevant part of the text.

All oxygen consumption data were corrected to STP.

# PART II

## The transition from the embryonic to post-embryonic existence

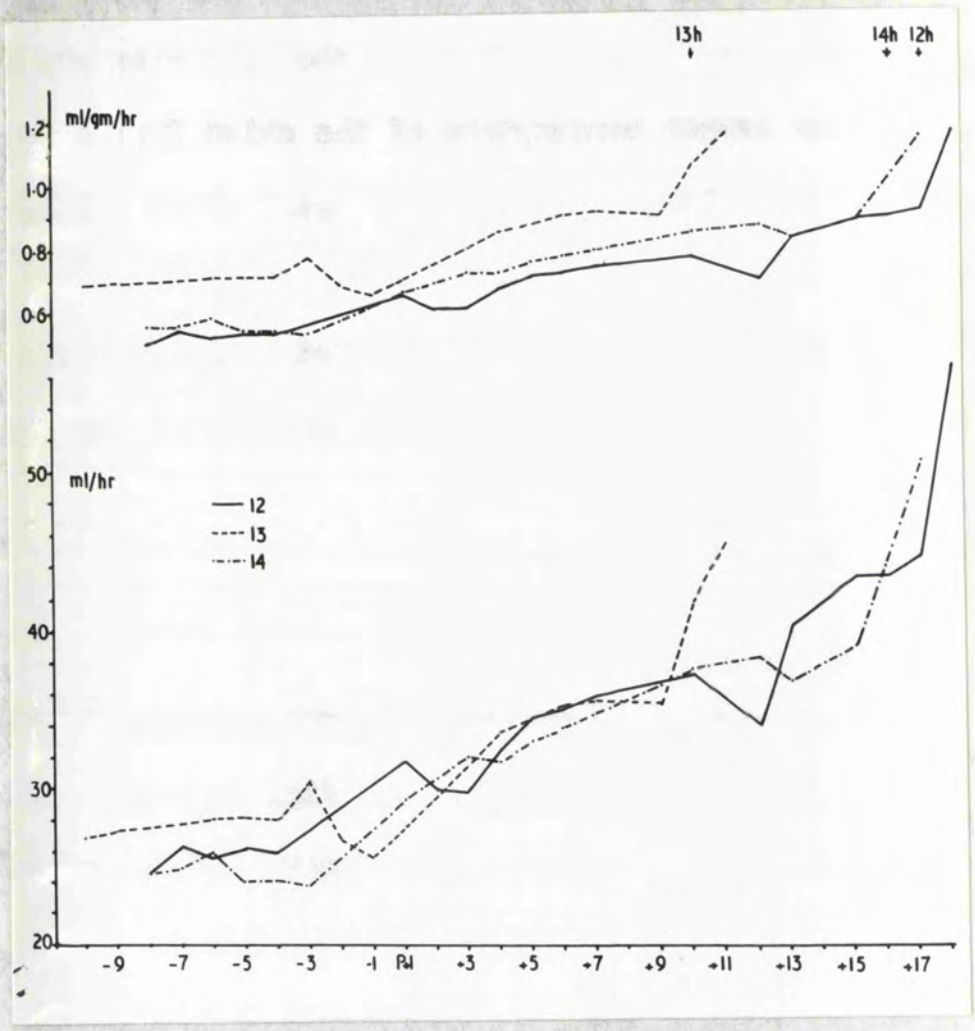



Fig. 6. The oxygen uptake of the hatching White Leghorn embryo, a) ml.$O_2$/gm/hr; b) ml.$O_2$/hr. The ordinate gives the time in hours before and after pipping (P). h indicates the time of hatching.

## II.1 The oxygen requirements of the hatching embryo

Giaja & Jovancic (1950) have shown that the rise in oxygen consumption at hatching can probably be divided into two distinct parts, correlated with the stage of hatching. These findings have been re-investigated in greater detail in the following experiments by determining the oxygen consumption of the chick from a few hours before the onset of pulmonary respiration until about six hours after hatching.

White Leghorn (WL) and Rhode Island Red x Light Sussex (RIR x LS) embryos were used. During the periods of observation the eggs were not turned.

Results

a) White Leghorns

The oxygen requirements of three typical embryos are given in table I and shown graphically in fig. 6. Fig. 6a shows the respiratory intensity of the embryo in ml/gm/hr with reference to pipping, and fig. 6b shows the oxygen uptake per hour for the same embryos. By standardizing the period between pipping and hatching (fig. 7) the similarity of the metabolic patterns of these three embryos can readily be seen. The pattern is as follows:- oxygen consumption begins to rise 3 or 4 hours in advance of pipping and continues for several hours before levelling out again. Just before active hatching commences, a second, rapid rise in oxygen consumption is initiated, and continues until 1 hour after the chick has hatched. The level reached 1 hour after hatching is very similar to that of the

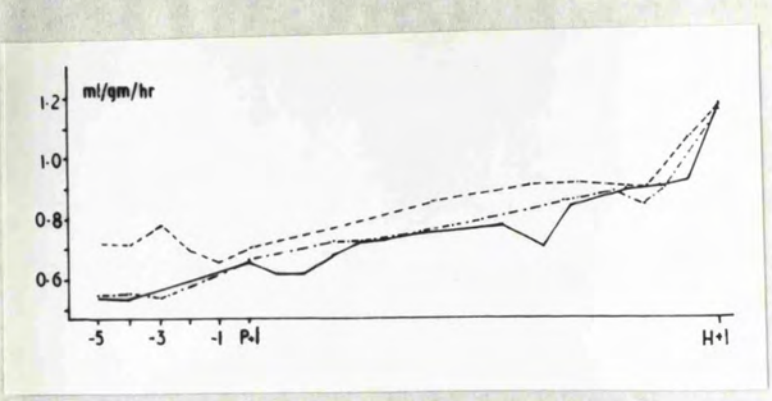



Fig. 7. The metabolic rates of three hatching embryos plotted against a fixed time interval between pipping (P) and hatching (H). Time before pipping in hours.

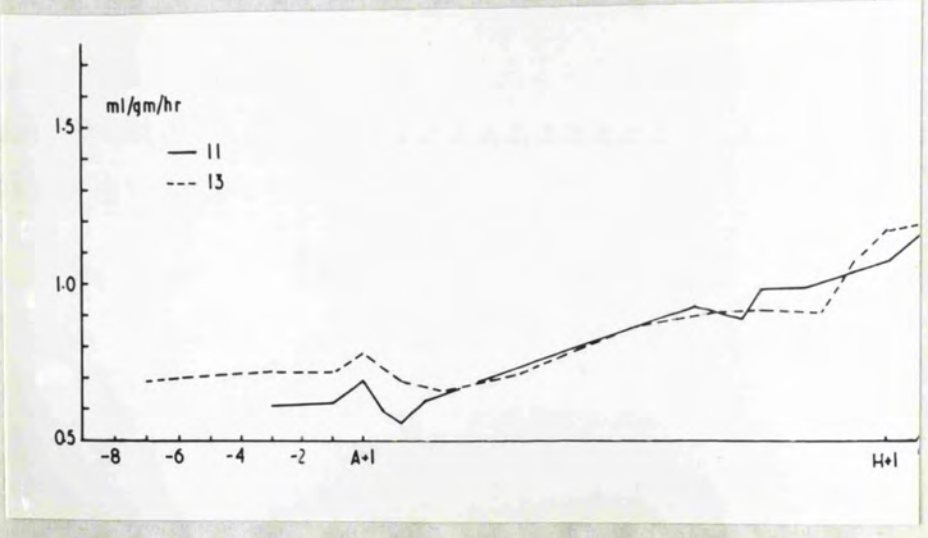



Fig. 8. The oxygen requirements of two embryos during hatching. Time scale as for fig. 7, but the onset of pulmonary respiration taken as one reference point. Note fall in metabolic rate after the onset of breathing (A).

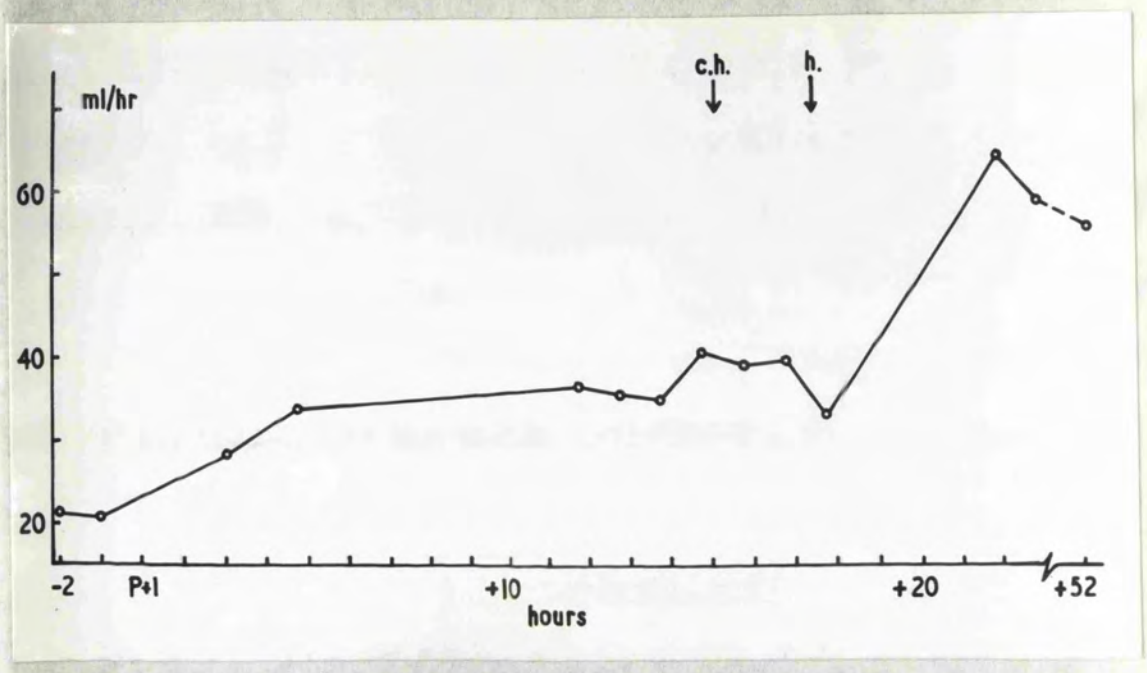



Fig. 9. The oxygen consumption of a typical RIR x LS hatching embryo. P = pipped; c.h. = commencement of active hatching; h = hatched. Note the rise in oxygen uptake after hatching: see also fig. 10.

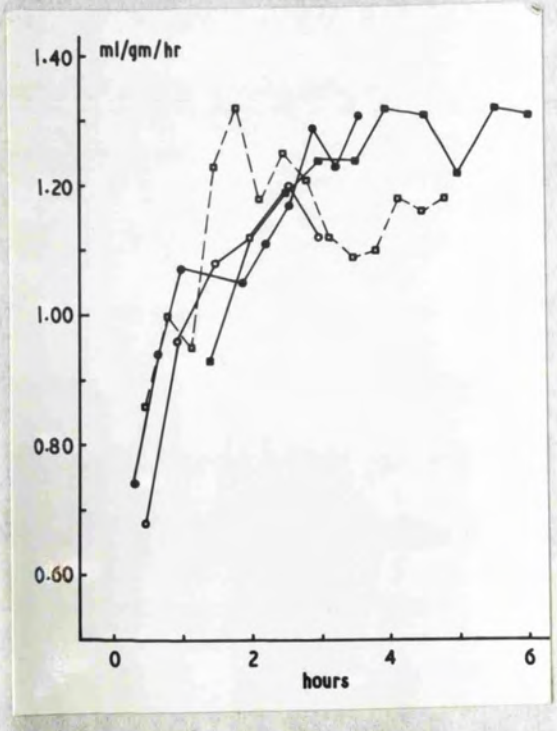



Fig. 10. The post-hatching rise in the metabolic rate of RIRxLS chicks

day-old chick.

There was slight evidence to suggest that, immediately following the initiation of pulmonary respiration, the oxygen consumption falls until pipping takes place (fig. 8). This may possibly be due to the diversion of blood from the chorio-allantois to the lungs. Since the oxygen content of the air space is below 13% at this time there would be a reduction in the oxygen available to the chick.

b) Rhode Island Red x Light Sussex

Similar results were obtained with this strain. The metabolic curve of a typical embryo is given in fig. 9, and the data in table II. In this strain there was a tendency for the oxygen consumption to rise with the commencement of active hatching and then to fall transiently at the moment of hatching. This was in contrast to the WL chicks where the rise was uninterrupted. Measurements of oxygen consumption were therefore continued for six hours after hatching. During the first half of this period the rate of uptake increased greatly and then became relatively constant during the remaining 5 hours of observation. Results are given in fig. 10 and table III. Although the chick exhibited little physical activity during the observational period, its oxygen consumption rose from an average of 0.90 ml/gm/hr half an hour after hatching to an average of 1.22 ml/gm/hr 3 hours after hatching. After this the metabolic rate became relatively constant.

Discussion

The rise in oxygen consumption begins before pipping. This would appear, at least in part, to be a result of the commencement of pulmonary respiration, since ventilation of the lungs is an active process and extra energy must therefore be expended. However, the lungs do not immediately become the sole site of gaseous exchange because the chorio-allantois continues to function for several hours (the parafoetal period). It may be expected that as the chick becomes more dependent upon its pulmonary circulation, oxygen consumption will rise proportionately. Such a relationship has been demonstrated by Visschedijk (1962a).

There is little evidence to identify the stimulus which actually initiates breathing. In these experiments there was a mortality of about 70% and it is significant that over 50% of these embryos died after pipping. Macroscopic examination showed them to be normal with no visible lesions and no evidence of malpositioning. Since the method of determining the oxygen consumption was one employing closed circuit principles, it seems likely that carbon dioxide may have been a contributory factor to the stimulation. Whilst the shell is intact the partial pressure of carbon dioxide within the air space will increase progressively. Pipping, however, removes the barrier to free diffusion and since the partial pressure of carbon dioxide within the respirometer itself is zero, there will be a rapid disappearance of carbon dioxide from the air space. A fall in the partial pressure of carbon dioxide in the blood would therefore be

| Breed | n | H + 1hr | H + 3hr |
|---|---|---|---|
| WL | 5 | 1.17±0.06 | --- |
| RIR x LS | 8 | 0.93±0.10 | 1.22±0.09 |

Table 10. The metabolic rates of two breeds 1 and 3 hours after hatching. Figures in ml/gm/hr ± SD.

expected as a result of breathing an atmosphere free of this gas. The lowered partial pressure of carbon dioxide in the blood might well be sufficient to reduce or even lose, the stimulatory effect of the gas upon the respiratory centres. Because of the high tolerance of the parafoetus to carbon dioxide (the partial pressure in the air space may reach 70 mm of mercury) only a small reduction may be necessary for the failure of the respiratory movements.

Since the second rise in the oxygen uptake of the W.L. chick was initiated before there was any increase in physical activity, it seems likely that the rise was due to an increase in the secretion rate of a metabolic-accelerating hormone. The post-hatching level of oxygen consumption was very similar between both individuals and breeds (see table 10). This finding supports the view that a hormone is responsible for the rise. The thyroid hormones seem to be the most likely to be concerned in raising the metabolic rate and the findings of Sun (1932) that the thyroid gland is particularly active at hatching is in agreement with this conclusion. Furthermore, since the rise in oxygen uptake preceded the onset of active hatching the hypothesis is advanced that active hatching is initiated by an increase in the rate of thyroid hormone secretion.

Giaja & Jovancic (1950) have also suggested that the rise in metabolic rate may be partly linked with the emergence of homeothermy. According to Pembrey et al. (1895) the homeothermic response is developed during hatching. However, little regulation of the body heat production would be expected when the environmental temperature

is within the zone of thermal neutrality. Therefore, the second rise may be associated with some other unknown physiological phenomenon.

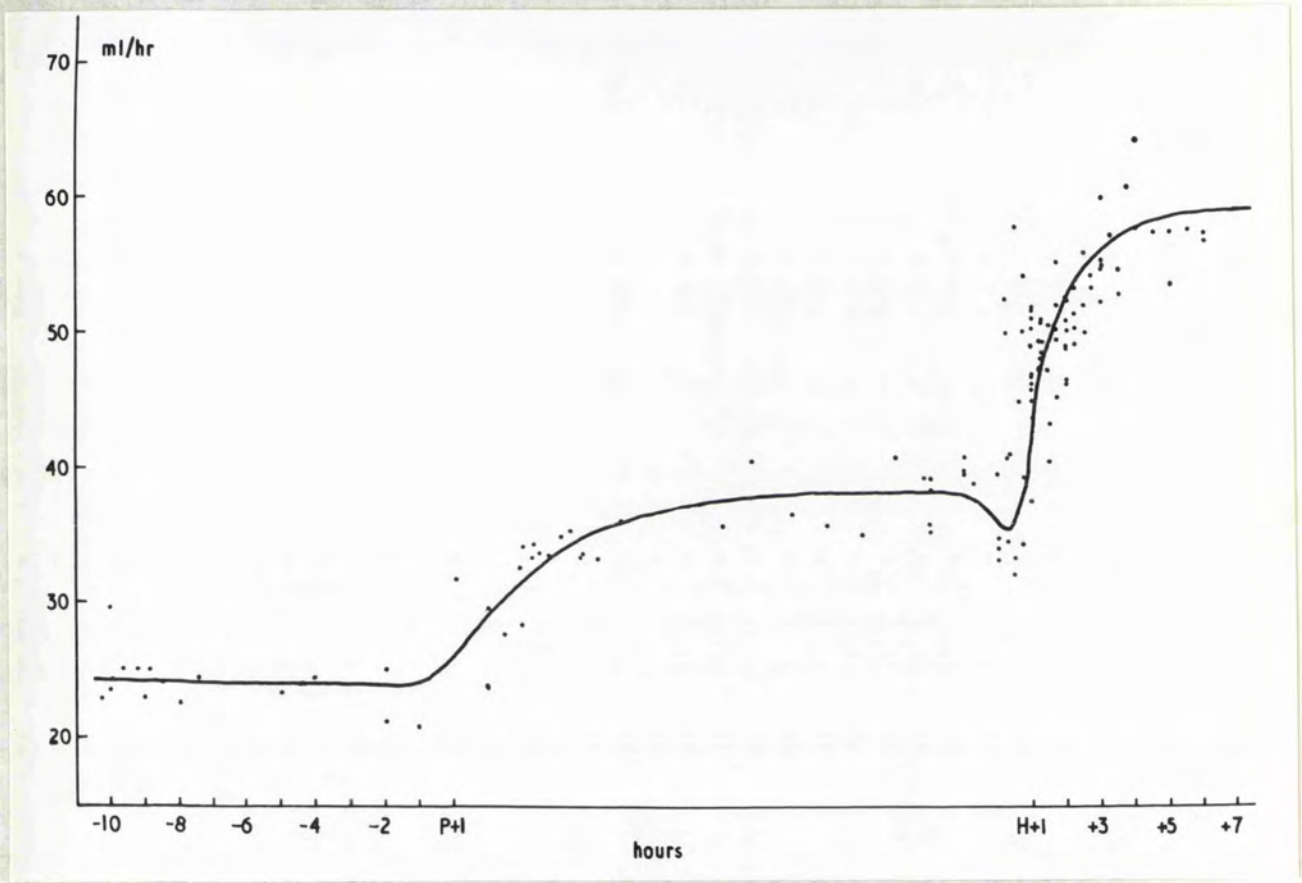



Fig. 11. The oxygen requirements of the normal hatching RIR x LS embryo at an environmental temperature of 37.7°C. The time scale between pipping (P) and hatching (H) is variable. Results obtained from 12 embryos, each point being the oxygen uptake of one embryo at that time.

| | Age hrs | $O_2$ uptake |
|---|---|---|
| Full-term embryo | - | 24.0 |
| Parafoetus | 1 | 24.0 |
| | 5 | 38.0 |
| Hatched chick | 1 | 38.0 |
| | 5 | 59.0 |

**Table 11.** The oxygen requirements of the RIR x LS chick during the termination of its embryonic existence. Figures (in ml/hr) calculated from fig. 11 .

## II.2 The development of the homeothermic response in the fowl

The time at which the homeothermic response is developed is not known precisely. The metabolic rate of the hatching chick has already been shown to be variable, and this makes it difficult to determine whether a rise in the rate is linked with the hatching process or is a response to a lowered environmental temperature. The normal metabolic pattern must therefore be accurately determined for the whole period of hatching in order to clarify this point.

In these experiments RIR x LS embryos and chicks were used. The normal environmental temperatures were $37.7^{\circ} \pm 0.1^{\circ}C$ for the embryo and just hatched chick, and $35^{\circ} \pm 0.1^{\circ}C$ for the day-old chick. All experimental temperatures were realized within 10 minutes. After each temperature change one hour was allowed to elapse before the determinations of oxygen consumption were recommenced, thereby permitting the apparatus to come into thermal equilibrium with the environment.

All measurements were carried out on single chicks and each chick was used once only.

### Results

#### a) The oxygen requirements of the hatching embryo

The metabolic pattern and mean oxygen requirements of the hatching embryo were determined by pooling the results from twelve embryos (fig. 11). The mean oxygen uptake of the chick just prior to the onset of pulmonary respiration, 1 and 5 hours after the onset of breathing, and 1 and 5 hours after hatching, has been computed

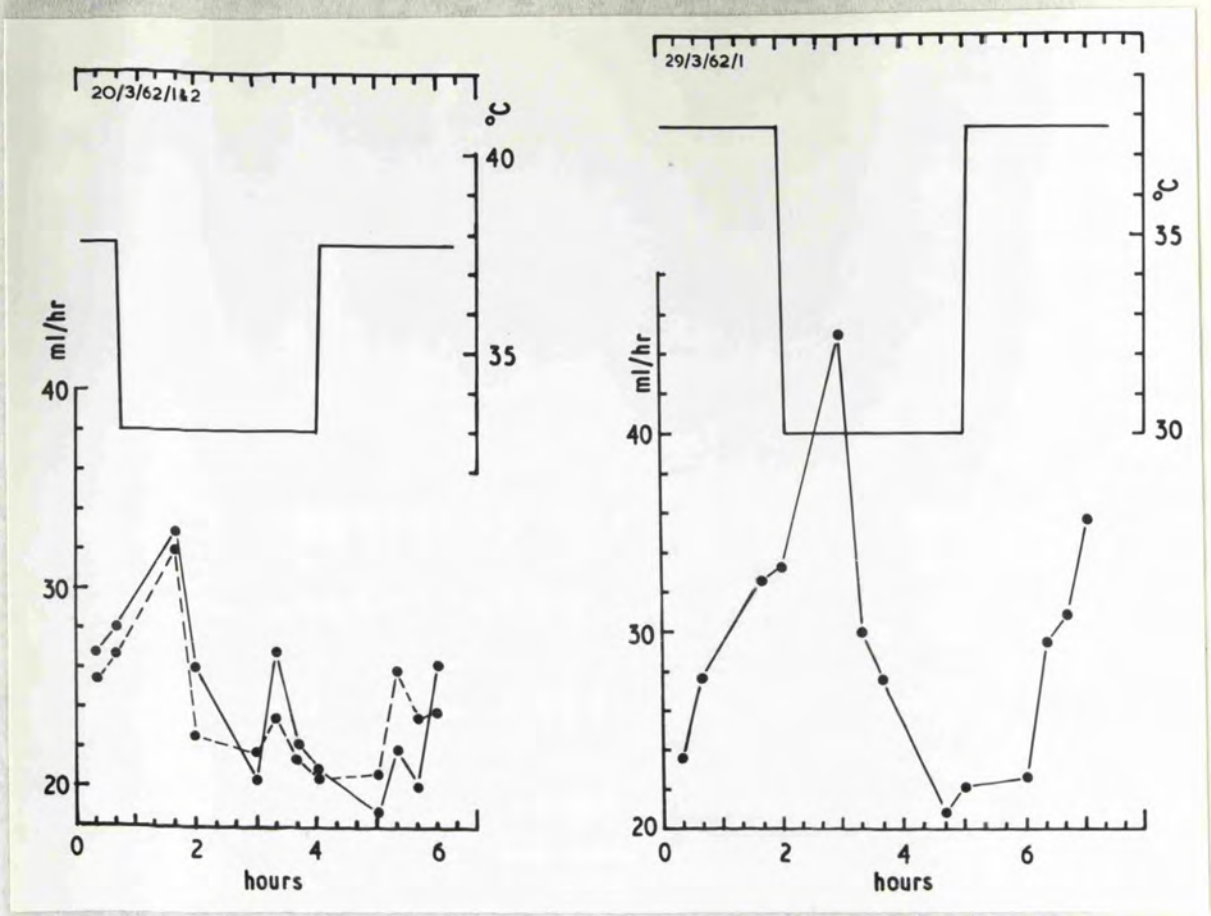



Fig. 12. Fig. 13.

Fig. 12. The effect of lowering the environmental temperature upon the 19 day old embryo. The results from 2 individuals are given. Note the slight, transient homeothermic response.

Fig. 13. The response of the parafoetus to cold. In spite of the transient rise in oxygen consumption, the parafoetus is essentially poikilothermic.

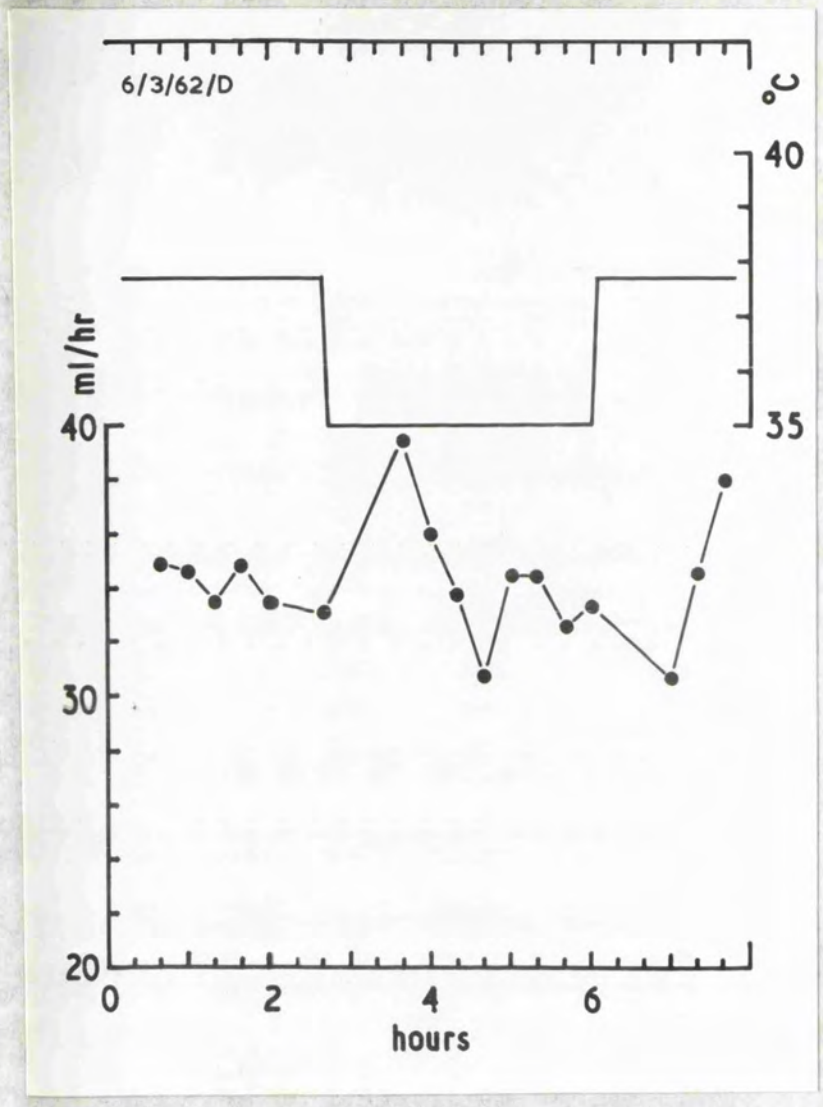



Fig. 14. If the fall in environmental temperature is less than 3°C then the parafoetus exhibits little change in oxygen consumption. This corresponds to the neutral condition first described by Pembrey et al. (1895)

from this figure and the results are given in table 11.

An increase in oxygen consumption above the appropriate normal figure in response to a reduction in the environmental temperature was taken to be indicative of a homeothermic response.

b) The effect of temperature on the oxygen consumption of the full-term and hatching embryo

i) Full-term embryo, 19 days

On reduction of the environmental temperature from 37.7°C to 33°C there was a slight, but transient, homeothermic response. Thereafter the oxygen consumption fell. A definite lag was found between the re-establishment of the normal environmental temperature and the restoration of the initial metabolic rate (fig. 12).

ii) The parafoetus

The pattern was very similar to that shown by the 19 day old embryo. After a transient homeothermic response, the rate of oxygen consumption declined progressively (fig. 13). On returning to the normal temperature, this rate increased, but did not attain its original level until 1½ hours had elapsed.

The degree of response appeared to be related to the amount of "stimulation". If the environmental temperature was reduced by 2.7°C (to 35°C) there was very little change in the metabolic rate (fig. 14). This is similar to the neutral condition first described by Pembrey et al. (1895).

iii) The hatching chick

The metabolic rate changes rapidly with the onset of active hatching. In fig. 15 the results from two chicks which hatched

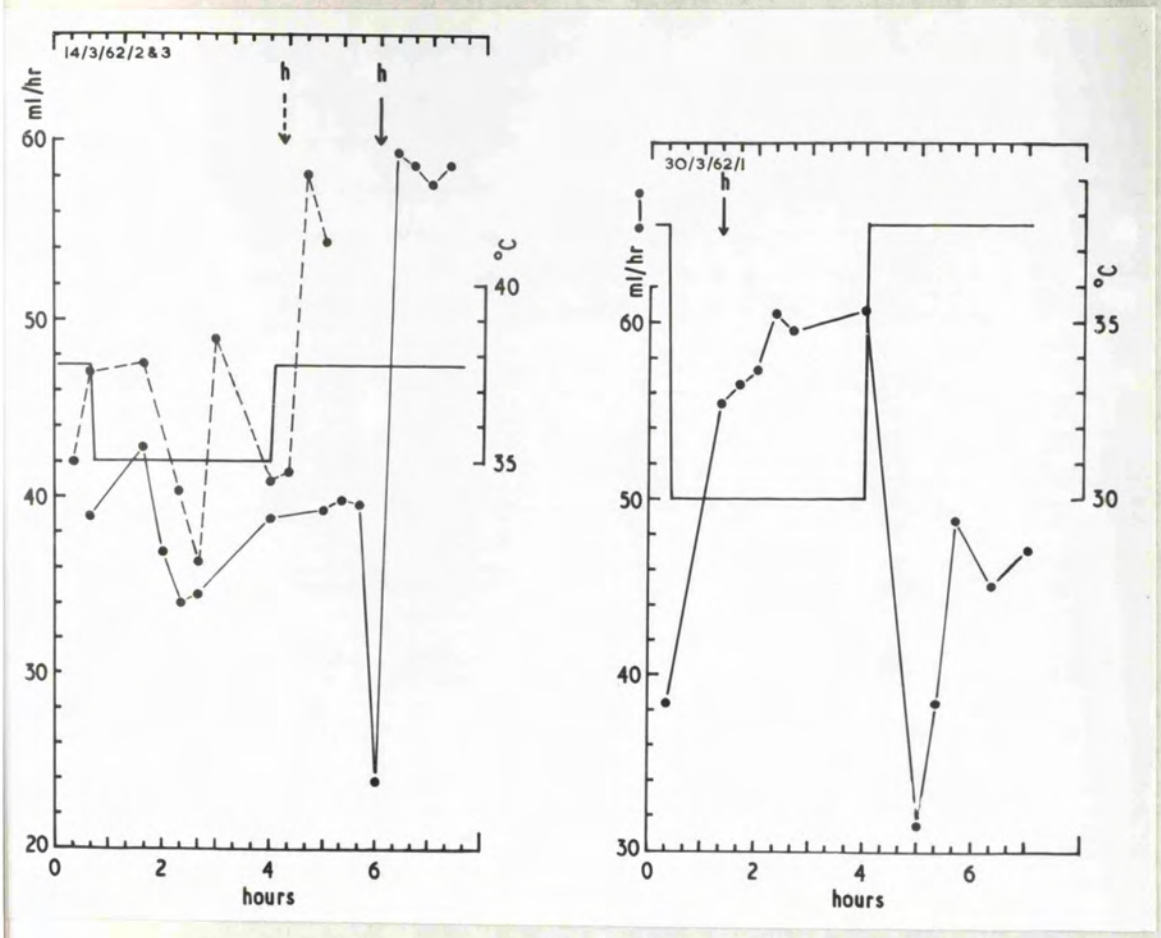



Fig. 15.

Fig. 16.

Fig. 15. The chick during the period of active hatching is seen to be still a poikilotherm. 2 individual records.

Fig. 16. The chick escaped from the shell membranes during the cold stimulation. A sustained homeothermic response is evident.

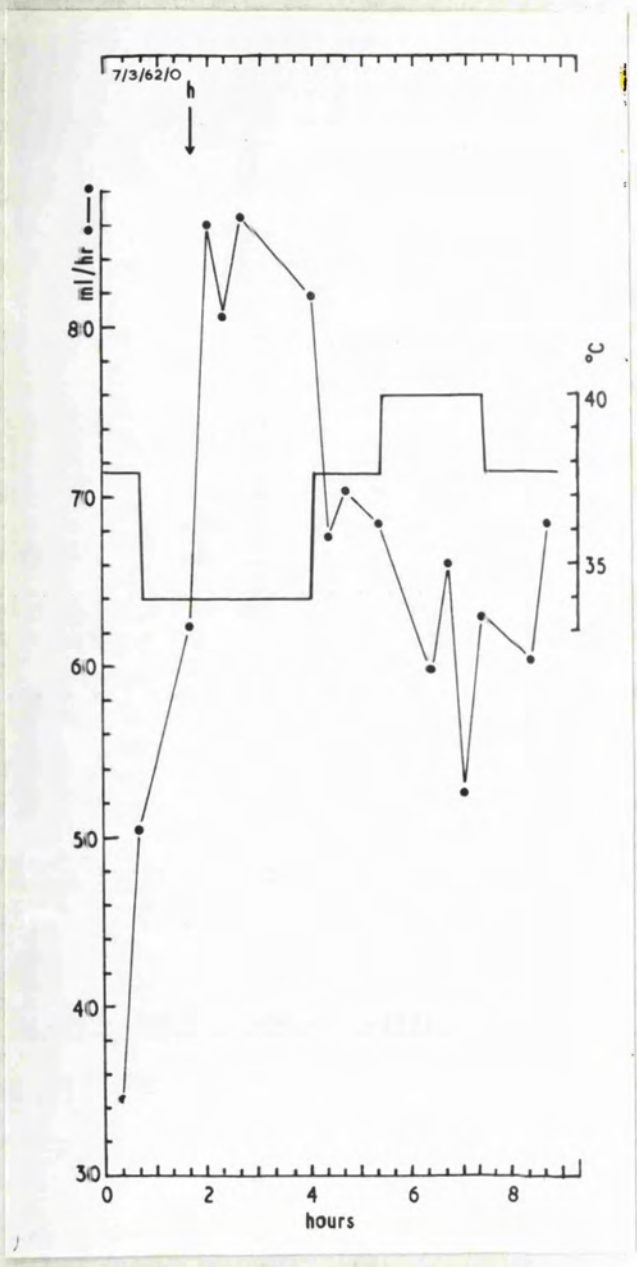



Fig. 17. The homeothermic response is developed in the chick as it escapes from the shell. This suggests that the response is mainly peripheral and probably nervous.

during the observational period are shown. The metabolism of the chick that hatched first appeared to be relatively unaffected by the decrease in temperature, but the second embryo showed a transient homeothermic response and then the poikilothermic response.

The results obtained at a slightly later stage of development are shown in fig. 16. After reducing the environmental temperature to 30°C the oxygen consumption of the chick rose by some 47% within 1 hour. At this point the chick hatched. One hour later oxygen consumption was 60 ml/hr or about 43% above the normal requirement. This was a sustained homeothermic response and the interpretation is further supported by the fact that the metabolic rate fell dramatically after the environmental temperature had been returned to normal.

That the newly hatched chick is able to give a sustained response to temperature changes is again demonstrated in fig. 17. Upon reduction of the environmental temperature to 34°C, oxygen consumption rose to 83 ml/hr within 80 minutes and fell to 69 ml/hr immediately the normal temperature was restored. When the temperature was raised to 40°C the oxygen consumption fell to 62 ml/hr. Thus, the response to a lowered environmental temperature was more marked than for a similar increase in the temperature.

iv) <u>The hatched chick</u>

The chicks examined were 24-30 hours old. The response to a fall in temperature was rapid, though the increase was not so great as for the just-hatched chick (fig. 18). This was probably due to

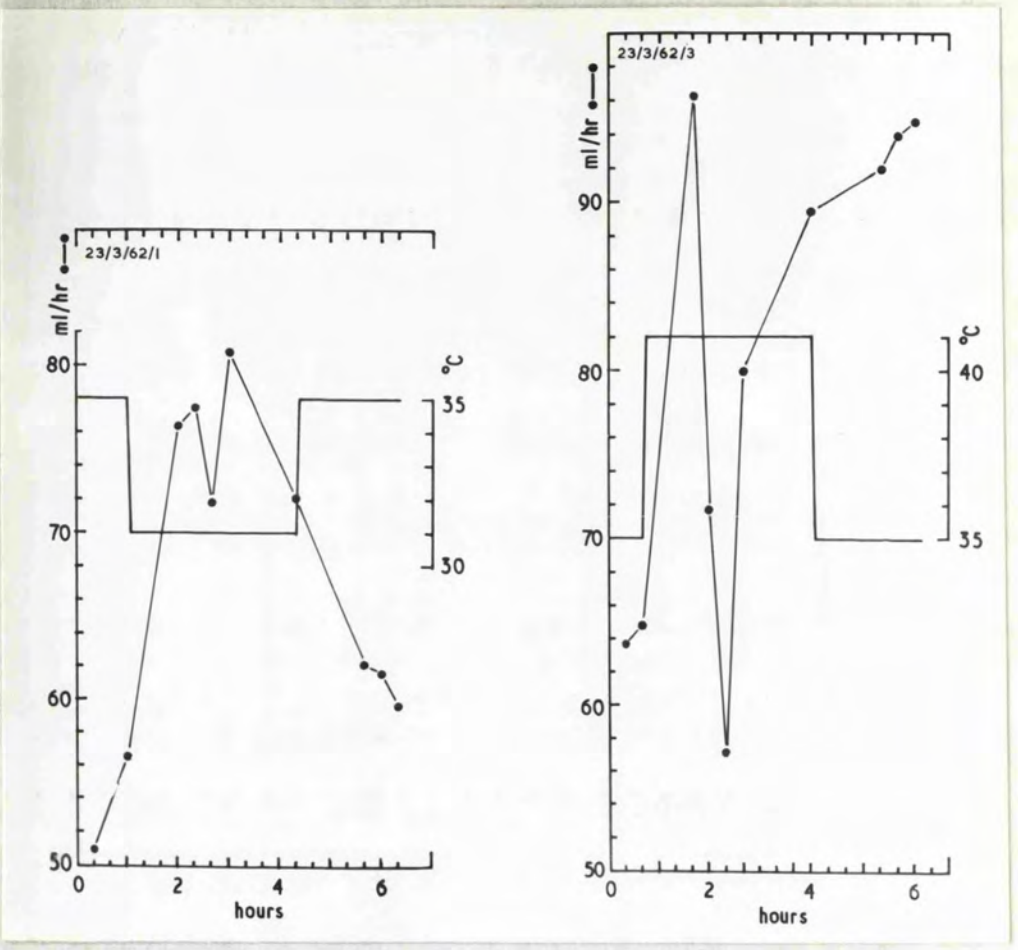



Fig. 18. Fig. 19.

Fig. 18. The day-old chick is a homeotherm and it will be seen that the response is immediate.

Fig. 19. The effect of a hot environment upon the day-old chick. It will be noted that the rising oxygen consumption continued after the temperature had been returned to normal. Thus hyperthermia was developing. It appears that the young chick is better able to deal with a cold environment.

the improved insulation of the dried down feathers.

The chick was found to be able to regulate its heat production at high environmental temperatures less well (fig. 19). The chick was subjected to a temperature of 41°C - that is 6°C above the zone of thermal neutrality. The rate of oxygen uptake rose as expected, but this rise continued after the environmental temperature had been returned to normal. This suggests that the homeothermic response had been completely broken down, and that hyperthermia was developing.

The bird develops the ability to regulate its heat production for long periods at the moment of hatching, though it appears that the response may be partially developed during the latter stages of incubation. The rapidity of the development is quite striking. The emergence of the response coincides with the second rise in the oxygen consumption and this fact lends support to the suggestion of Giaja and Jovancic (1950) that they are related.

| | n | ml/hr |
|---|---|---|
| Full-term embryo | 23 | 25.0 |
| Parafoetus (5hr) | 11 | 34.9 |
| Hatched chick (3 hr) | 18 | 55.6 |

**Table 12.** The oxygen requirements of the RIR x LS fowl during hatching.

| | n | $cm^2$ | | $ml/cm^2/hr$ |
|---|---|---|---|---|
| | | | Full-term | 0.37 |
| Egg | 30 | 67.9 | | |
| | | | Parafoetus | 0.51 |
| Chick | 10 | 92.8 | Hatched | 0.60 |

**Table 13.** The surface area of the egg and the hatching chick, and the relationship between the oxygen uptake and the surface area.

## II.3 The relationship between the metabolism and surface area in the hatching embryo

It has been pointed out (page 51) that the ability to regulate heat production *per se* may not be responsible for the rise in oxygen consumption at hatching since the observations were carried out in a thermally neutral environment. Dawes & Mott (1959), however, found that there is a relationship between the surface area and the metabolic rate of the new-born sheep and it was decided to investigate whether a similar relationship existed in the fowl. Full-term embryos (RIR x LS), chicks in the parafoetal period and three hour old chicks were used in this experiment. The number of individuals used are given in table 12.

### Results

The average oxygen requirements in ml/hr are summarized in table 12. The surface areas of the egg and chick, together with the computed oxygen consumption in $ml/cm^2/hr$, are given in table 13. It will be seen from this table that the oxygen uptake of the parafoetus and the hatched chick were of the same order, but that the figure for the full-term embryo was much lower.

That there is little correlation between the oxygen consumption per square centimetre of the full-term embryo and the hatched bird lends support to the conclusion that the rise in oxygen consumption associated with the establishment of pulmonary respiration is linked, at least in part, with the provision of the extra energy required to ventilate the lungs.

It is not surprising that the correlation between the oxygen uptake of the parafoetus and the hatched chick is not exact since the hatched bird is more active and is probably in an environment where the temperature is slightly above the upper critical temperature of the chick. However, it may be concluded that the rise in oxygen consumption (measured in ml/hr or ml/gm/hr) is a direct response by the chick to maintain its body temperature at its pre-hatching level.

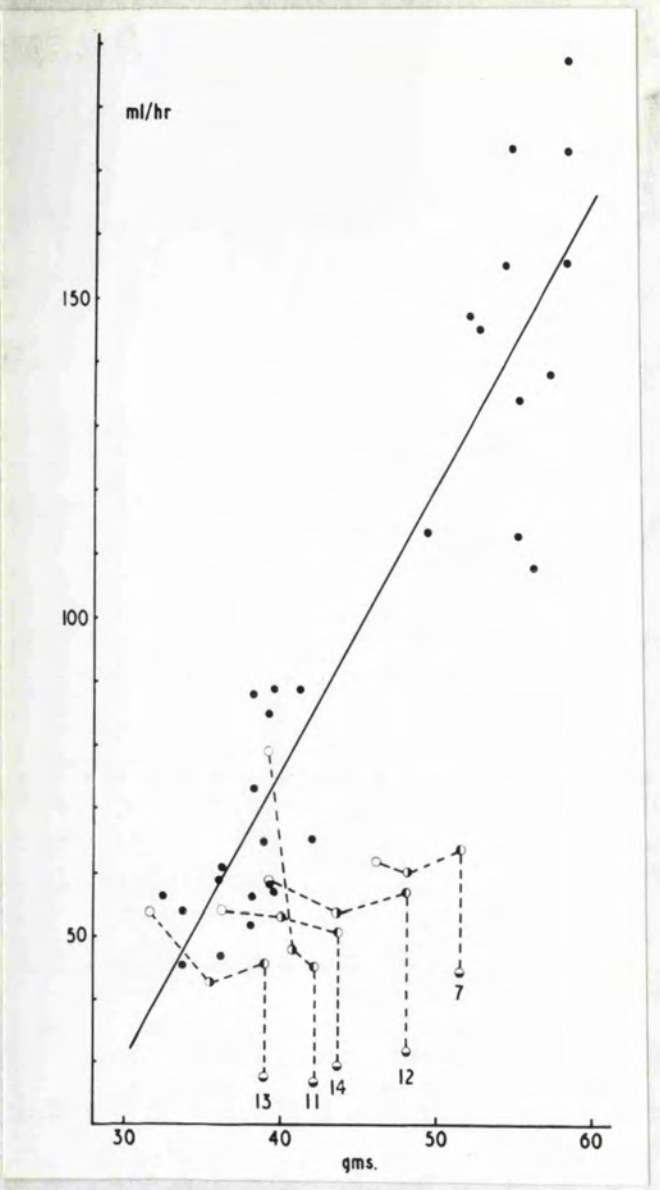



Fig. 20. The oxygen consumption of individual chicks during hatching and the first two days of post-embryonic life. ◒ = pipped; ◐ = hatched; ◑ = 1 day old; ○ = 2 days old; ● = normal chicks from "day-old". With the exception of no. 11 the oxygen uptake was fairly constant although the body weight fell. When 22 days old (2 days after hatching) the $O_2$ uptake was in the normal range. Also see text.

## II.4 The effect of the yolk sac on metabolism during hatching and the subsequent two days

The hatching chick has large (5 gm or more) food stores in the form of yolk. Yolk is metabolically inactive and the yolk sac itself contributes little to the total oxygen requirements: according to Needham (1932) it is responsible for less than 2% of the total oxygen consumption of the full-term embryo. The yolk sac and its contents are virtually "dead weight" and the calculated metabolic rate (ml/gm/hr) will therefore be depressed. Here this factor has been investigated using the parafoetus, newly hatched, 1 and 2 day old chick. Neither food nor water were available during the experiments. The chicks had to rely upon their yolk reserves to supply their energy requirements.

The oxygen consumption of the same breed (WL), which were hatched concurrently, were determined from the 22nd day after the commencement of incubation - i.e. "day-old" chicks - to the end of the first week after hatching. These data provided the normal oxygen requirements of the growing chick. The standard diet and water were available ad libitum.

All the embryos used in this study hatched after 20 days of incubation.

Results

The results of the normal growing chick are given in table IV and are shown in figs. 20 and 21. It is evident that the oxygen uptake increased at a greater rate than the body weight during the

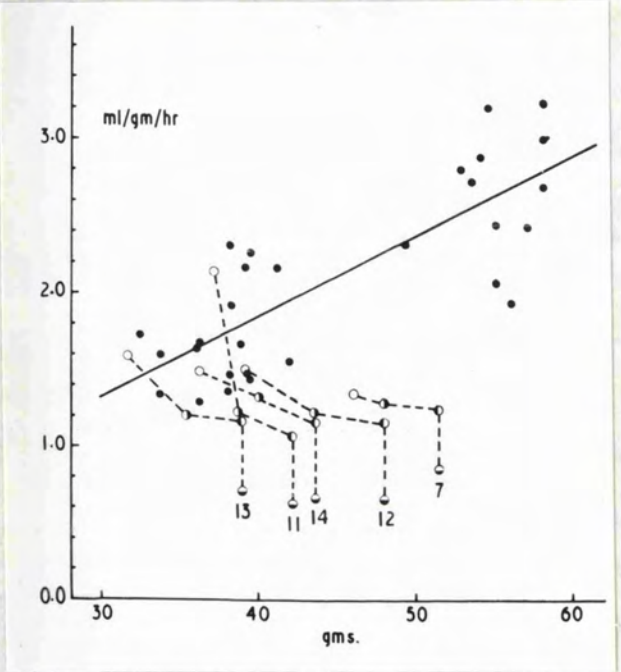



Fig. 21. The metabolic rate of the chick during hatching and the first two days of post-embryonic life. Symbols as for fig. 20. It will be seen that the loss in weight (through the utilization of yolk) was sufficient to bring the metabolic rate into the normal range of "day-old" chicks, i.e. after 22 days incubation.

| Embryo no. | Pipping | | Hatching | | H + 1 day | | H + 2 days | |
|---|---|---|---|---|---|---|---|---|
| | ml/hr | ml/gm/hr | ml/hr | ml/gm/hr | ml/hr | ml/gm/hr | ml/hr | ml/gm/hr |
| 7 | 44.3 | 0.86 | 64.0 | 1.25 | 60.4 | 1.29 | 61.9 | 1.35 |
| 11 | 26.6 | 0.63 | 45.0 | 1.07 | 47.8 | 1.23 | 79.2 | 2.14 |
| 12 | 31.7 | 0.66 | 56.8 | 1.18 | 53.6 | 1.23 | 58.9 | 1.50 |
| 13 | 27.6 | 0.71 | 45.5 | 1.17 | 42.6 | 1.20 | 53.8 | 1.59 |
| 14 | 29.4 | 0.67 | 50.8 | 1.16 | 53.0 | 1.32 | 54.0 | 1.49 |

Table 14. Oxygen consumption at pipping, hatching, 1 and 2 days after hatching. WL chicks, individual records.

period of observation - the equation of the regression line was calculated to be $y = 0.0005x^{3.216}$.

The oxygen requirements of individual chicks from pipping to 2 days after hatching are given in table 14 and in figs. 20 and 21. It will be seen from the data of individual chicks that, in general, the oxygen consumption of the hatched bird remained fairly constant when measured in ml/hr, but that the metabolic rate (ml/gm/hr) rose progressively.

Examination of the two figures shows that the progressive fall in the body weight resulted in the oxygen consumption moving, by the 22nd day, into the normal range expected for the "day-old" chick. Thus it may be concluded that the yolk sac and its contents cause the metabolic rate to be artificially depressed, the degree of depression being progressively reduced with the gradual absorption and utilization of the yolk. The rise in the metabolic rate at this time may therefore not be real.

## II.5 Factors concerned with the termination of the embryonic existence

In the section dealing with the oxygen requirements of the hatching embryo the hypothesis was advanced that the initiation and maintenance of pulmonary respiration and the act of pipping depend on the stimulatory effect of a high partial pressure of carbon dioxide in the blood and that active hatching is stimulated by a hormonal agent, possibly from the thyroid gland. The evidence for this hypothesis may be summarized as follows:-

i) Pulmonary respiration and pipping

a) Over 50% of the deaths occurred after pipping when the partial pressure of the carbon dioxide in the atmosphere was very low (approaching 0 mm Hg).

b) The air space of the full-term embryo contains 5 - 9% carbon dioxide.

c) Carbon dioxide, in moderate concentrations, is a respiratory stimulant.

ii) Active hatching

a) A rise in the oxygen consumption was detected, at least for White Leghorn chicks, before there was any increase in physical activity.

b) The metabolic rates of individual chicks were very similar after hatching.

c) The thyroid gland is active immediately after hatching.

## II.5.i The pulmonary and pipping stimulus

### The effect of altered diffusion rates across the shell of the air space

A certain partial pressure of carbon dioxide in the blood appears to be necessary for the initiation and maintenance of pulmonary respiration and for pipping. This may therefore be called the critical partial pressure. If this partial pressure, normally achieved on about the twentieth day of incubation, can be realised earlier, then, assuming the embryo is competent to respond at this earlier time, pulmonary respiration and pipping will be brought forward. Similarly, if the realization of the critical partial pressure can be delayed, then breathing and pipping may also be delayed. From such findings it may be inferred that the stimulus for both breathing and pipping is gaseous in nature. At the same time, if such experiments have no effect on the time of hatching, then it will be reasonable to suppose that active hatching is initiated by a stimulus of a different nature.

Firstly, the effect of changing the diffusion rates across the shell of the air space was investigated, using embryos from a RIR x LS cross. The diffusion rates were altered at 18½days' incubation by:

a) waxing the shell of the air space to reduce the rates and so accelerate the changes in gas concentrations;

or, b) perforating the air space to allow free diffusion between the space and the incubator environment, thereby preventing,

| | | Pipping | | Hatching | | Pip to hatching | | |
|---|---|---|---|---|---|---|---|---|
| Group | n | Wax | Perf | Wax | Perf | Wax | Norm | Perf |
| 1 | 24 | +22 | -4 | 0 | +1 | 41 | 19 | 14 |
| 2 | 24 | +12 | 0 | +2 | +2 | 31 | 21 | 19 |
| 3 | 12 | | -14 | | 0 | | 18 | 4 |
| 4 | 24 | +5 | -13 | +2 | -2 | 23 | 20 | 13 |
| 5 | 24 | +4 | -11 | +2 | +1 | 26 | 24 | 12 |
| 6 | 28 | +25 | | +2 | | 37 | 14 | |
| Mean | | +13.7 | -8.4 | +1.6 | +0.4 | 31.6 | 19.3 | 12.4 |

**Table 15.** The effect of waxing or perforating the shell of the air space of the embryo aged $18\frac{1}{2}$ days upon the times of pipping and hatching. Figures in hours; + = advancement; - = retardation.

or substantially delaying, the realisation of any critical concentration.

Observations were made hourly to determine the times of pipping and hatching.

Results

136 embryos, divided into six groups, were used. The results are given in table 15. Embryos with waxed shells pipped in advance of the controls by an average of 14 hours, whilst pipping by the embryos with perforated air spaces was retarded by 8 hours. The time of hatching was not affected by either of these treatments. The period between pipping and hatching was quite variable consequently, which suggests that the two events are not related.

It may be concluded therefore, that the pipping stimulus is gaseous in nature and is probably a high partial pressure of carbon dioxide, although the low oxygen tension within the air space may be a small contributory factor.

It was observed that as soon as the shell had been fractured, the chick became quiescent suggesting that pipping is only a reflex response to hypercapnia, and of little importance in the hatching process.

The effect of ventilating the air space with atmospheric air

It has been suggested above that pipping may only be a response to a noxious atmosphere and therefore cannot be regarded as a true ontological phenomenon, but the onset of pulmonary respiration is, and may well be a better criterion on which to measure the effects

| Group | n | Pul.resp. P | Pul.resp. A | Pipping P | Pipping A | Hatching P | Hatching A | Pul.resp to hatch P | Pul.resp to hatch Norm | Pul.resp to hatch A |
|---|---|---|---|---|---|---|---|---|---|---|
| 7 | 18 | 0 | -1 | -4 | -4 | 0 | 0 | 24 | 24 | 23 |
| 8 | 18 | -5 | +1 | -4 | -4 | -6 | +4 | 17 | 16 | 13 |
| 9 | 18 | -1 | -1 | -13 | -21 | -6 | -3 | 31 | 26 | 28 |
| 10 | 18 | -1 | -2 | -10 | -9 | 0 | 0 | 19 | 20 | 18 |
| 11 | 18 | 0 | -2 | -8 | -11 | +1 | -7 | 21 | 22 | 27 |
| Mean | | -1.4 | -1.0 | -7.8 | -9.8 | -2.2 | -1.2 | 22.4 | 21.6 | 21.8 |

Table 16. The effect of perforating (P) or aerating (A) the air space of the 18½ day old embryo upon the termination of the embryonic existence as compared to the normal embryo. Figures in hours; + = advancement; - = retardation.

of environmental changes on the termination of the embryonic existence. In these experiments the effects of ventilating the air space on the onset of pulmonary respiration, pipping and hatching were determined.

RIR x LS embryos aged 18½ days were used. There were two control groups - normal intact embryos and embryos with perforated air spaces.

Results

a) The initiation of pulmonary respiration

Examination of table 16 shows that whilst the times of pipping were substantially delayed by both perforation and ventilation of the air space, the onset of pulmonary respiration was not significantly altered.

Pulmonary respiration was delayed by only 1 hour in the aerated embryo, even though the gas mixture in the air space was quite dissimilar to the norm (about 9% carbon dioxide and 13% oxygen). This suggests that the composition of the gases in the air space do not influence the time of the onset of pulmonary respiration. However, pipping was advanced by an average of 13.7 hrs. in the waxed embryo (table 15) whilst the period between the onset of air breathing and pipping was found to be 8 hrs. for the normal bird, suggesting, indirectly, that an elevated carbon dioxide tension in the air space may advance the actual time of air breathing by about 5.7 hours.

b) Pipping

Pipping was delayed by an average of 7.8 hours in chicks with

| | Group | | | | | |
|---|---|---|---|---|---|---|
| | 7 | 8 | 9 | 10 | 11 | Mean |
| Pul. resp. to pip. | 5 | 12 | 6 | 8 | 9 | 8.0 |

Table 17. The time interval between the onset of pulmonary respiration and pipping in normal embryos. Figures in hours.

| | 7 | 8 | 9 | 10 | Mean |
|---|---|---|---|---|---|
| Controls | 464 | 467 | 468 | 468 | 467 |
| Perforated | 470 | 475 | 468 | 467 | 470 |
| Aerated | 460 | 470 | 468 | 475 | 468 |

Table 18. The effects of perforating or aerating the air space of the full-term embryo upon the time of hatching. Figures are number of hours incubation for the completion of development.

perforated air spaces and by 9.8 hours in those with aerated air spaces (table 16). The mean time interval between the onset of pulmonary respiration and pipping in normal embryos was 8 hours (see table 17). It is apparent from tables 16 and 17 that the action defined as pipping (page 2) was almost completely inhibited in embryos which had aerated air spaces and in several embryos it was observed that pipping did not occur although hatching was not impaired or delayed. Therefore it may be concluded that pipping is not essential to the hatching process and is only a reaction against a noxious environment.

c) Active hatching

The time at which hatching was completed was little influenced by the treatments (see tables 16 and 18) thus confirming the findings of the first experiments. Since all the chicks hatched at a similar time, and the metabolic rates were very similar between individuals (see tables 10 and 14) it seems likely that the hatching stimulus is hormonal in nature. Furthermore, since the period between the onset of pulmonary respiration and hatching was fairly constant (table 16), it may be possible that the pulmonary stimulus is, after all, hormonal.

The effect of waxing the shell of the air space upon the time of initiation of air breathing

The onset of pulmonary respiration is only slightly delayed by perforating or ventilating the air space, but indirect evidence has been presented to suggest that waxing the shell, thereby causing a rapid increase in the tension of carbon dioxide in the air space,

| Group | n | Pul.resp | Hatch | Pul.resp to hatch | |
|---|---|---|---|---|---|
| | | | | Waxed | Control |
| 9 | 12 | +8 | -5 | 32 | 19 |
| 18 | 40 | +6 | +3 | 27 | 24 |
| 19 | 36 | +2 | -1 | 25 | 22 |
| Mean | | +5.3 | -1.0 | 28.0 | 21.7 |

Table 19. The effect of waxing the shell of the air space of the $18\frac{1}{2}$ day old embryo upon the time of the initiation of pulmonary respiration and the time of hatching. Figures in hours.

may cause pulmonary respiration to be initiated at an earlier time. This has been investigated further using 18½ day old embryos.

Results

The results are summarized in table 19. It will be seen that although pulmonary respiration was initiated on average of 5.3 hours in advance of the controls, the time of hatching was not significantly affected. This agrees very well with the figure of 5.7 hours obtained by indirect means (see page 64).

These results indicate, therefore, that the pulmonary stimulus is probably a high partial pressure of carbon dioxide in the blood acting upon the respiratory centres which in turn initiate pulmonary respiration. A hormonal stimulus seems much less likely.

II.5.ii The hatching stimulus

Whilst the work of the above section was directed mainly at elucidating the nature of the pulmonary stimulus, some information was obtained from these experiments to strengthen the hypothesis that active hatching is stimulated by a hormone. These salient points are:

i) The length of the incubation period is very constant (table 18).

ii) Alteration of the gaseous environment within the air spaces was without effect on the time of hatching.

The hormone site may well be the thyroid (see page 61). Therefore the effects of the thyroid hormones, a goitrogen and the thyroid stimulating hormone of the pituitary on the time of hatching

| | | Pul.resp | | Hatch. | | Pul.resp to hatch | | |
|---|---|---|---|---|---|---|---|---|
| Group | n | T3 | T4 | T3 | T4 | T3 | SC | T4 |
| 11 | 108 | | +1 | | +4 | | 24 | 21 |
| 12 | 108 | +2 | +1 | +7 | +5 | 18 | 24 | 20 |
| 13 | 108 | 0 | 0 | +5 | +4 | 18 | 23 | 19 |
| 14 | 108 | 0 | 0 | +4 | +3 | 19 | 23 | 20 |
| Mean | | +0.7 | +0.5 | +5.3 | +4.0 | 18.3 | 23.5 | 20.0 |

T3 = l-triiodothyronine injected embryos; T4 = l-thyroxine injected embryos; SC = solvent injected embryos.

Table 20. The effects of injecting 1 µg of sodium-l-triiodothyronine or sodium-l-thyroxine into the air space of the $18\frac{1}{2}$ day old embryo upon the termination of the embryonic existence. Figures in hours; + = advancement.

have been determined. The times of the onset of pulmonary respiration have been noted concurrently to determine whether a hormonal stimulus might be involved.

The effect of exogenous thyroxine or triiodothyronine on the termination of the embryonic existence

In this series of experiments, the effects of injecting the sodium salts of l-thyroxine or l-triiodothyronine into the air space of the full-term embryo were determined. It must be emphasised that in these experiments, the hormones were administered to the normal and fully developed chick embryo (18½ days old), and not to the young embryo where the metabolism and subsequent development would be grossly affected. The aim of these experiments was to "trigger" normally developed mechanisms or precipitate natural events in the foetus. These hormones were considered to become active very shortly after injection since MacLagan, Sprott & Wilkinson (1952) have shown that the latent period of both drugs is 3 hrs. in the rat.

Results

Four groups, each of 108 eggs, were examined and the results are summarized in table 20. The time of the onset of pulmonary respiration was only very slightly affected, whereas the time of hatching was advanced by an average of 4.0 hours by the l-thyroxine, and 5.3 hours by the l-triiodothyronine.

The thyroid hormones did not affect mortality.

At first sight it might be thought that l-triiodothyronine had a slightly greater effect on the chick embryo than l-thyroxine.

However, this is probably not so for sodium-l-thyroxine has five molecules of water of crystallization whilst sodium-l-triiodothyronine is anhydrous. Thus 1 µg sodium-l-thyroxine is equivalent to 0.87 µg l-thyroxine and 1 µg sodium-l-triiodothyronine to 0.96 µg l-triiodothyronine. Therefore these two hormones have a similar potency in the embryo, confirming the work of Shellabarger (1955), Newcomer (1957) and Tata & Shellabarger (1959).

There was no significant effect upon the time of the initiation of pulmonary respiration. Since the hormones were injected into the air space at least twenty-two hours previously, the conclusion that the thyroid is not involved in the stimulation of pulmonary respiration would appear to be valid. The advancement in the time of hatching was brought about entirely by a reduction in the time interval between the onset of air-breathing and active hatching. Thus it may be concluded that the thyroid gland probably provides an essential factor of the stimulus for the onset of active hatching in the chick embryo.

The effect of 2-thiouracil on the termination of the embryonic existence

There is some doubt as to whether the inhibitory effect of the goitrogens is completely specific to the thyroid gland. Some evidence has been presented to show that the thiocarbamide drugs can affect the general metabolism of the cell. In order to validate conclusions based upon any positive results it is necessary to reduce or neutralize that effect by exogenous thyroid hormones. Here the effects of 2-thiouracil have been investigated, 2 mg or 4 mg being

| | | Pul.resp | | Hatch | | Pul.resp to hatch | | |
|---|---|---|---|---|---|---|---|---|
| Group | n | 2* | 4* | 2 | 4 | 2 | SC* | 4 |
| 15 | 108 | 0 | +1 | -6 | -8 | 27 | 21 | 30 |
| 16 | 84 | -1 | -2 | -14 | -21 | 36 | 23 | 42 |
| 17 | 72 | -2 | 0 | -12 | -23 | 40 | 30 | 53 |
| Mean | | -1.0 | -0.3 | -10.7 | -17.3 | 34.3 | 24.7 | 41.7 |

*2 = 2 mg 2-thiouracil; 4 = 4 mg 2-thiouracil; SC = solvent only.

+ = advancement; - = retardation.

Table 21. The effect of 2-thiouracil upon the termination of the embryonic existence. Injections were made into the air spaces of $18\frac{1}{2}$ day old embryos. Figures in hours.

| Group | % Hatch | | | % Mortality | | |
|---|---|---|---|---|---|---|
| | 2* | Cont* | 4* | 2 | Cont | 4 |
| 15 | 67 | 86 | 69 | 5 | 5 | 3 |
| 16 | 59 | 86 | 50 | 14 | 11 | 14 |
| 17 | 57 | 75 | 52 | 13 | 21 | 20 |
| Mean | 61 | 82 | 54 | 11 | 12 | 12 |

* 2 = 2 mg 2-thiouracil; 4 = 4 mg 2-thiouracil; Cont = solvent only.

Table 22. The percentage hatch and percentage mortality as affected by 2-thiouracil.

Both figures were determined 24 hours after 50% of the controls had hatched.

injected into the air space two days before the expected onset of pulmonary respiration. Some embryos treated with 2 mg 2-thiouracil on the eighteenth day were also injected with 2 µg sodium-l-thyroxine either at the same time or 1 day later, in an attempt to reduce any positive effect the goitrogen might have.

a) The effect of 2-thiouracil

Three groups totalling 264 embryos were examined. The results are given in table 21. Although the time of the onset of pulmonary respiration was not significantly altered by either dose level of thiouracil, the time of hatching was greatly delayed. The larger dose of thiouracil caused the greater delay in hatching. The percentage hatch was calculated twenty-four hours after 50% of the control embryos had hatched. The figures are given in table 22. It will be seen that whereas the majority of the control embryos had hatched by this time, only half of the thiouracil treated embryos had completed their development, whether 2 mg or 4 mg had been administered. Mortality, however, was not affected (table 22).

b) The effect of a mixture of 2-thiouracil and l-thyroxine

The time of the initiation of pulmonary respiration of the treated embryos was not affected in either group (see table 23). Hatching in both groups also occurred at the same time as the control group. Thus 2 µg sodium-l-thyroxine (equivalent to 1.74 µg l-thyroxine) completely neutralized the effect of 2 mg 2-thiouracil.

| Group | n | Pul.resp | Hatch | Pul.resp to hatch | | % Hatch | |
|---|---|---|---|---|---|---|---|
| | | | | Treated | Control | Treat | Cont |
| 18a | 58 | 0 | 0 | 23 | 23 | 80 | 81 |
| 18b | 58 | +1 | +1 | 22 | 23 | 86 | 80 |

Table 23. The effect of 2 mg 2-thiouracil and 2 µg sodium-l-thyroxine on the termination of the embryonic existence. Figures in hours; + = advancement. For further details see text.

Discussion

No goitrogen appears to influence the release of stored hormones. This may explain the variation in the results. Thus in group 15 2 mg of thiouracil delayed hatching by only 6 hours as compared with one day according to Grossowicz (1946). But over the next 18 hours only a further 17% of the thiouracil treated embryos hatched, whilst another 36% of the controls completed their development. Some embryos, therefore, may have sufficient stored thyroid hormones to allow hatching to take place at the normal time.

It has been consistently found that goitrogenic drugs, if injected before the seventeenth day of incubation, cause a marked rise in mortality (Grossowicz, 1946; Adams & Bull, 1949; Adams & Buss, 1952; Romanoff & Laufer, 1956; Rogler et al., 1959b). This is not so if the drug is administered after this age (see table 22).

That exogenous thyroxine reduced the effects of the thiouracil on the termination of the embryonic existence shows that the interpretations of the results obtained when thiouracil alone was injected into the embryo are valid.

The time at which pulmonary respiration was initiated was not significantly affected by any of these treatments. Again this supports the hypothesis that the pulmonary stimulus is not hormonal.

Since the thyroid gland is not a wholly autonomous gland but controlled by the anterior lobe of the pituitary gland, it must be supposed that active hatching is ultimately under the control of the pituitary. The effect of the thyrotrophic hormone on active hatching

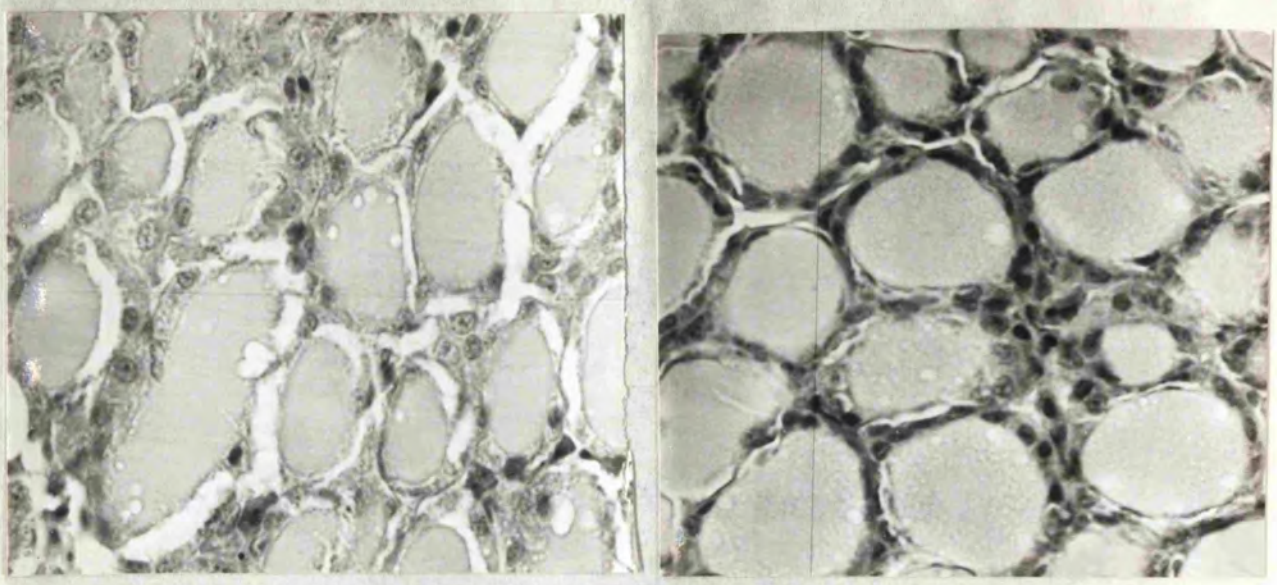
| | | Pul.resp | | Hatching | | Pul.resp to hatch | | |
|---|---|---|---|---|---|---|---|---|
| Group | n | 1* | 2* | 1 | 2 | 1 | SC | 2 |
| 19 | 40 | +1 | +1 | +5 | +6 | 22 | 26 | 21 |
| 20 | 40 | 0 | 0 | +2 | +2 | 22 | 24 | 22 |
| Mean | | +0.5 | +0,5 | +3.5 | +4.0 | 22.0 | 25.0 | 21.5 |

* 1 = 1 i.u.; 2 = 2 i.u; SC = saline injected control.

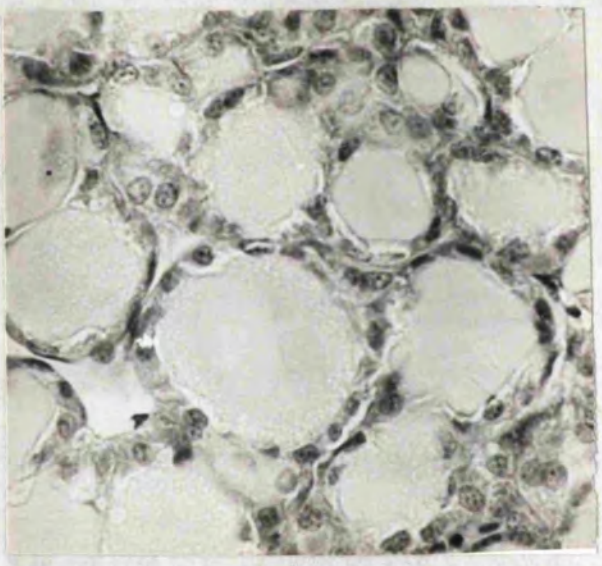
Table 24. The effect of 1 i.u. or 2 i.u. of thyrotrophic hormone on the termination of the embryonic existence. Injections made into the air space at $18\frac{3}{4}$ days. Figures in hours; + = advancement.

| Dose i.u. | n | Pul.resp | Hatch |
|---|---|---|---|
| 0.005 | 56 | 0 | +2 |
| 0.05 | 56 | 0 | 0 |
| 0.5 | 40 | +1 | -1 |
| 1.0 | 80 | +0.5 | +3.5 |
| 2.0 | 80 | +0.5 | +4.0 |

Table 25. The effect of thyrotrophic hormone upon the termination of the embryonic existence: the effect of dose. Figures in hours; + = advancement; - = retardation.



(a) (b)



(c)

Fig. 22. The thyroid gland of the hatching chick and the effect of thyrotrophic hormone upon its activity. (a) 19 day old embryo: little epithelial activity, much colloid. (b) 1 hour old chick: little increase in epithelial activity and some loss of colloid. (c) 1 hour old chick treated with 1 i.u. TSH at 18¾ days: epithelial activity slightly greater than (b), also increase in vacuolation of the colloid.

was therefore investigated.

The effect of thyrotrophic hormone on the termination of the embryonic existence

Embryos aged 18¾ days were used. The hormone was given at the following levels: 0.005, 0.05, 0.5, 1.0 and 2.0 i.u.

Results

The critical dose level was found to be 1.0 i.u. Neither 1.0 i.u. nor 2.0 i.u. had any effect on the time at which breathing began, but advanced the time of hatching by 3.5 and 4 hours respectively. The results are summarised in tables 24 and 25.

In spite of the demonstrated hypersensitivity of the chick thyroid gland to thyrotrophic hormone (Martindale, 1941), no response could be elicited with doses of below 1.0 i.u. and little increased activity in the thyroid gland was evident (fig. 22). However, the results did confirm that the thyroid is intimately concerned with the onset of active hatching, although ultimately, it must be the anterior hypophysis that supplies the stimulus. The pulmonary stimulus was again confirmed to be of a different nature.

# PART III

## The hatched bird

| Group | 1st. Metabolic phase | 2nd. Metabolic phase |
|---|---|---|
| A | $y = 0.0138x^{1.663}$ | $y = 3.684x^{0.913}$ |
| B | $y = 0.0383x^{1.951}$ | $y = 1.760x^{1.030}$ |
| C | $y = 0.0960x^{1.805}$ | $y = 2.073x^{1.038}$ |
| D | $y = 0.0297x^{2.091}$ | $y = 7.473x^{0.761}$ |

Table 26. The calculated regression equations for the curves shown in figs. 23 - 26.

## III.1 The oxygen requirements of three breeds of fowl during the first fortnight of post-embryonic life

The oxygen consumption of three strains of domestic fowl, Rhode Island Red x Light Sussex, a commercial broiler strain and a commercial laying strain, has been measured in order to determine whether the reported rise in the metabolic rate following hatching is a general phenomenon. The effect of the diet on the metabolic pattern and the absolute oxygen requirements of the laying strain during this period have also been investigated, by substituting for the standard ration a commercial broiler ration containing 23% crude protein and with a calculated metabolizable energy content of 1300 kcals/lb. All the conditions were otherwise identical.

No significant difference in the oxygen requirements of the male or female chicks could be detected and therefore no attempt was made to separate the results.

Oxygen consumption was measured at an environmental temperature of 35°C for the whole period.

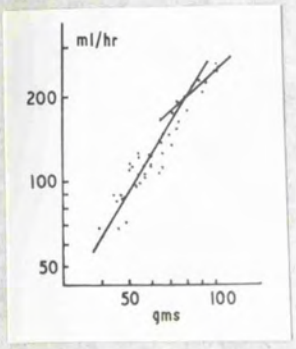
### Results

The data on oxygen consumption are given in tables V - VIII. The equations of the regression lines are given in table 26, and the growth curves of the groups are given in fig. 27.

i) Rhode - Sussex strain: Group A

The oxygen requirements were found to increase at a greater rate than the body weight until the latter had reached 80 gm (see fig. 23). At this weight a definite change in the metabolism could

Fig. 23



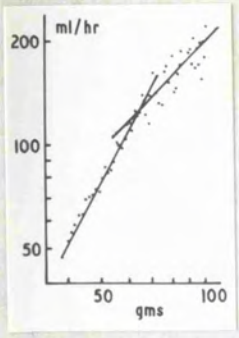
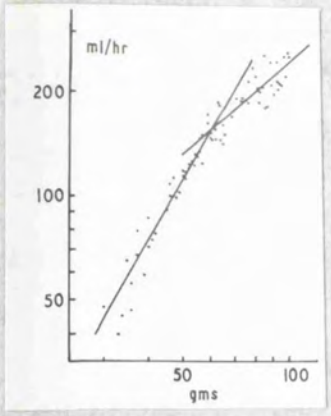




Fig. 24

Fig. 25



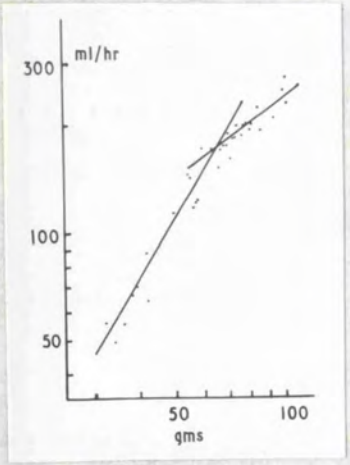




Fig. 26

Figs. 23-26. Oxygen consumption of three breeds during the first fortnight of post-embryonic life.

Fig. 23. RIR x LS, fed the standard ration: group A.

Fig. 24. Broiler strain, fed the standard ration: group B.

Fig. 25. Laying strain, fed the standard ration: group C.

Fig. 26. Laying strain, fed the broiler ration: group D.

| Wt. (gm) | Standard diet (groupC) | Broiler diet (groupD) |
|---|---|---|
| 40 | 75 | 76 |
| 60 | 155 | 168 |
| 100 | 247 | 248 |

**Table 27.** A comparison of the oxygen requirements of chicks of a laying strain fed either the standard diet or The broiler diet. Figures in ml/hr at STP.

| Group | Wt. (gm) | Age (days) |
|---|---|---|
| A | 80 | 9 |
| B | 63 | 7.5 |
| C | 57 | 8 |
| D | 65 | 9 |

**Table 28.** The body weights and ages at which the metabolic rates of the three breeds became constant.

be detected, the rates of growth and increase in oxygen uptake being approximately equal thereafter.

ii) Broiler strain: Group B

The metabolic pattern is shown in fig. 24. Two phases could be clearly defined, firstly, a rising metabolic rate (b = 1.951) extending from hatching to a body weight of 63 gm, whilst the second phase was characterized by an almost constant metabolism.

iii) Laying strain: Group C

Again two phases were differentiated; oxygen uptake rose at a greater rate than body weight to 57 gm and then became almost directly proportional to the latter (fig. 25).

iv) Laying strain: Group D

This group came from the same parents as group C. Its diet, however, was a broiler ration and not the standard ration. Examination of fig. 26 shows that the metabolic pattern was unaffected.

v) The effect of diet on the oxygen requirements and metabolic pattern of the laying strain

There was no significant difference between the oxygen requirements of the two groups (see table 27). The metabolic pattern was little affected. The regression coefficients of the group fed the standard ration (group C) were slightly higher (table 26), but this may have been due to the fact that there were fewer observations for Group D.

There was also a slight difference in the growth rate (fig. 27).

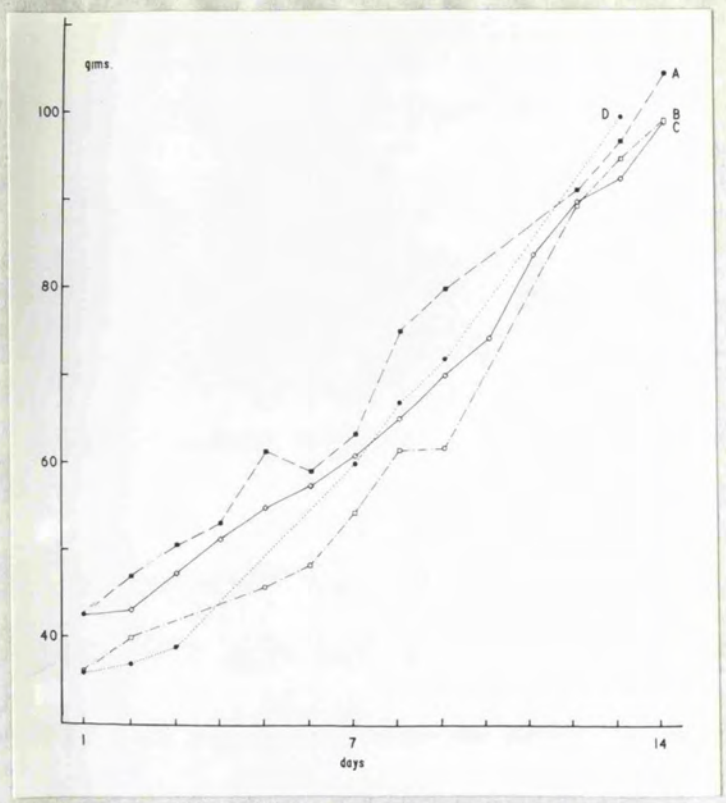



Fig. 27. Growth curves during the first fortnight of post-embryonic life. ■ = group A; □ = group B; ◇ = group C; ● = group D.

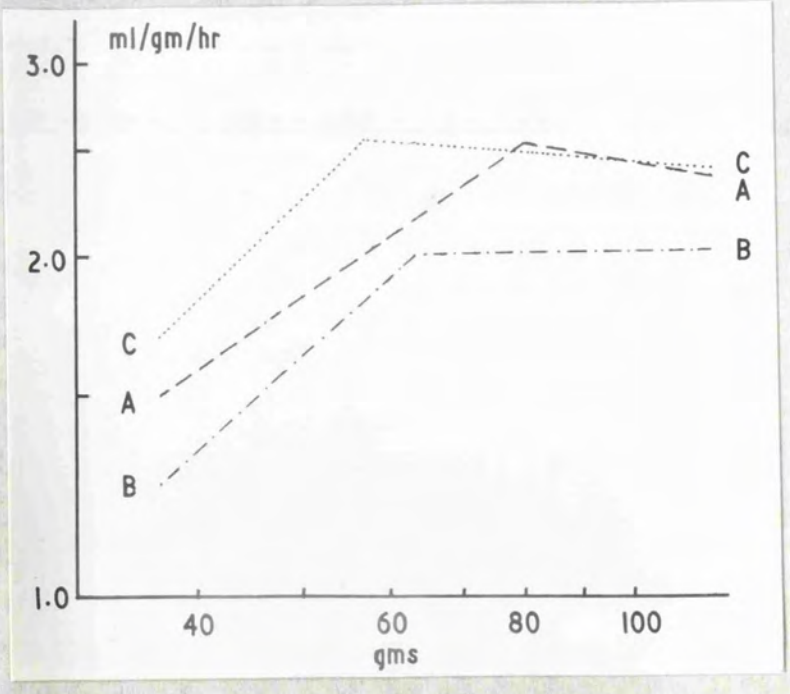



Fig. 28. Comparison of the metabolic rates of the three breeds. For further explanation see legends for figs. 23-26.

vi) A comparison of metabolic rates and patterns as influenced by strain

The metabolic rates of the three strains have been compared in fig. 28. It will be seen that the body weights at which the metabolic changes occurred were variable (see also table 28) but the ages at which these changes occurred were similar (table 28).

Discussion

The oxygen requirements of the fowl increase at a greater rate than the body weight (i.e. the metabolic rate increases) for some time after hatching. It will be seen in figs. 23 - 26 and 28 that the weight at which the metabolic rate became constant, although precise within the group, was variable between each group. Reference to table 28, however, shows that the age at which the metabolism became constant was similar and it is therefore concluded that the rising metabolic rate is related to the age of the bird rather than its body weight.

It has already been shown that the yolk sac and its contents are "dead weight", and cause a depression in the real metabolic rate. This depression progressively decreases to the fifth day when yolk absorption is completed (Virchow, 1891; Romanoff & Romanoff, 1933; Entenman, Lorenz & Chaikoff, 1940; Romanoff, 1944). Thus the overall rise in metabolic rate might not be real at the cellular level.

The post-hatching rise in the body temperature might be explained as a result of a rise in the metabolic rate. However, since it has been suggested above that there may be no real rise in

the metabolic rate some alternative explanation must be sought. The suggestion of Kendeigh & Baldwin (1928) and Randall (1943) that the mass of the bird increases at a greater rate than the surface area might resolve the problem.

| Age (days) | 1 ♂ | 1 ♀ | 2 ♂ & ♀ | 3 ♂ & ♀ | Mean ♂ & ♀ |
|---|---|---|---|---|---|
| 1 | 40.48 | 40.49 | 39.89 | 40.32 | 40.23 |
| 2 | 40.69 | 40.55 | 40.35 | 40.42 | 40.46 |
| 3 | | | | 40.77 | 40.77 |
| 4 | 41.00 | 40.91 | 41.00 | 40.85 | 40.94 |
| 5 | 40.83 | 40.78 | 40.58 | 40.95 | 40.78* |
| 6 | 40.96 | 41.04 | 40.91 | 40.95 | 40.95 |
| 7 | 41.01 | 41.08 | 40.84 | 40.97 | 40.95 |
| 8 | 40.92 | 40.99 | 40.80 | 41.05 | 40.94 |
| 9 | 40.90 | 40.94 | 40.97 | 40.95 | 40.94 |
| 10 | 40.93 | 40.96 | | | 40.95 |
| 11 | 40.89 | 40.90 | 41.19 | 40.97 | 41.02 |
| 12 | 40.95 | 40.97 | 41.10 | 40.94 | 41.00 |
| 13 | 41.01 | 40.93 | 40.95 | 40.99 | 40.97 |
| 14 | 40.87 | 40.92 | 41.00 | 40.93 | 40.94 |

* Significant fall between 4 and 5 days (P = 0.05).

**Table 29.** The body temperature of three consecutive batches of RIR x LS chicks from the same parent stock during the first fortnight of post-embryonic life. Group 1: each figure is the mean of 6 observations; groups 2 and 3: mean of 12 observations.

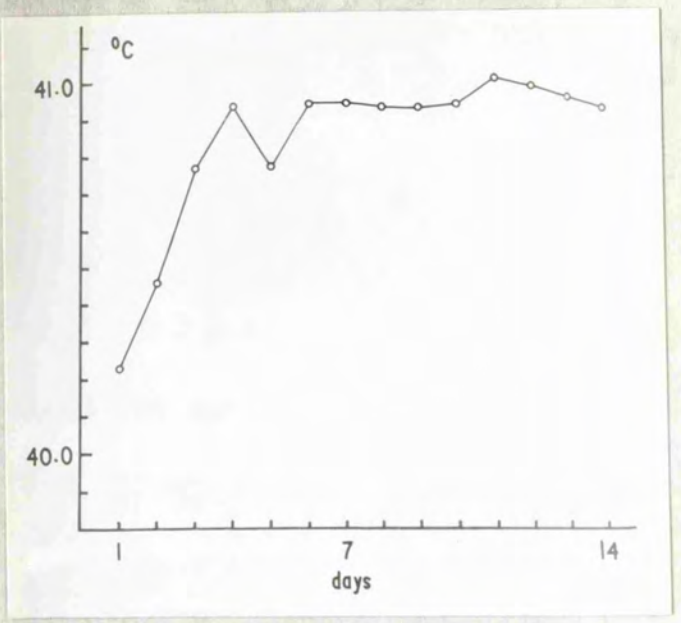



Fig. 29. The body temperature curve during the first two weeks after hatching. Each point is the mean of 36 observations.

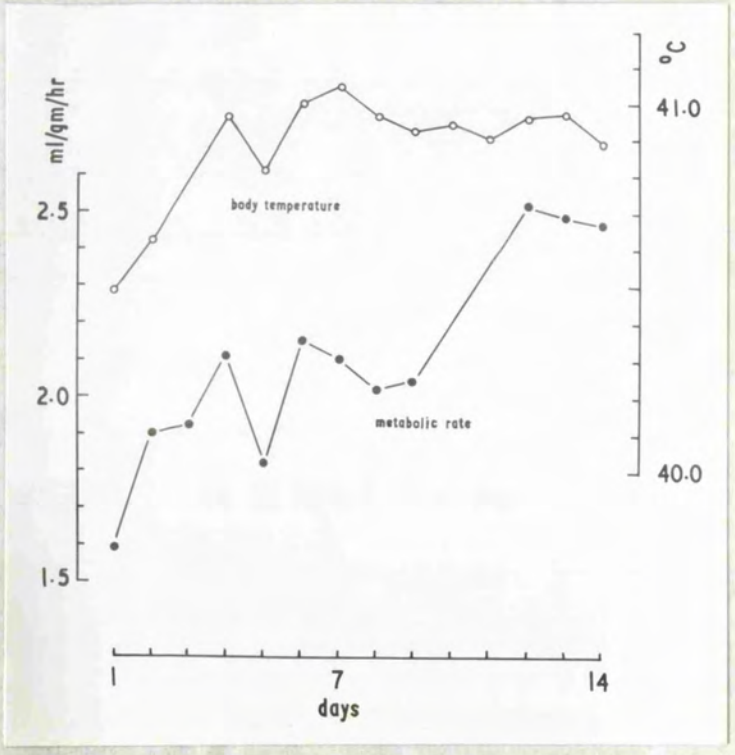



Fig. 30. The metabolic rate and the body temperature of chicks during the first fortnight. Batch 1; mean of 12 birds.

## III.2 The establishment of a constant body temperature and the relationship between the surface area and the mass of the chick

RIR x IS chicks were used. Three consecutive batches of 12 chicks were used to determine the normal body temperature curve during the first fortnight after hatching. The metabolic rate of the first batch was determined concurrently at an environmental temperature of 35°C.

Another group of RIR x IS chicks, 35 in number, were used to determine the yolk sac and body weights, the "active mass" (defined here as the body weight minus the yolk sac weight) and the surface area. 5 chicks were used daily.

The standard diet was available ad libitum.

Results

a) Body temperature and the metabolic rate

The mean daily temperatures of the three batches are given in table 29. The results of batch 1 confirmed that there is little or no difference between the body temperature of the male and female chick during the first fortnight after hatching. In view of this finding the sex of the bird was ignored in the other two batches.

In both males and females of batch 1 and in batch 2 there was a fall in the body temperature between the fourth and fifth days. Combination of all the results - to give mean values for 36 chicks (fig. 29) - shows that the body temperature of the chick became constant from the sixth day after hatching. The fall in the

| Age (days) | n | Wt. (gm) | YS (gm) | Active mass | SA $cm^2$ | $\frac{SA}{Active\ mass}$ | $\frac{SA}{Wt}$ |
|---|---|---|---|---|---|---|---|
| 1 | 5 | 41.4 | 5.3 | 36.1 | 87.8 | 2.43 | 2.12 |
| 2 | 5 | 41.7 | 3.7 | 38.0 | 85.0 | 2.24 | 2.04 |
| 3 | 5 | 43.6 | 1.5 | 42.1 | 89.0 | 2.11 | 2.04 |
| 4 | 5 | 45.0 | 0.6 | 44.4 | 91.0 | 2.05 | 2.02 |
| 5 | 5 | 48.8 | 0.3 | 48.5 | 92.4 | 1.91 | 1.89 |
| 6 | 5 | 51.3 | 0.0 | 51.3 | 106.2 | 2.07 | 2.07 |
| 7 | 5 | 53.8 | 0.0 | 53.8 | 102.8 | 1.91 | 1.91 |

YS = yolk sac; Active mass = body weight - yolk sac weight; SA = surface area.

Table 30. The body weight, yolk sac weight and the surface area of the chick during the first week of post-embryonic life.

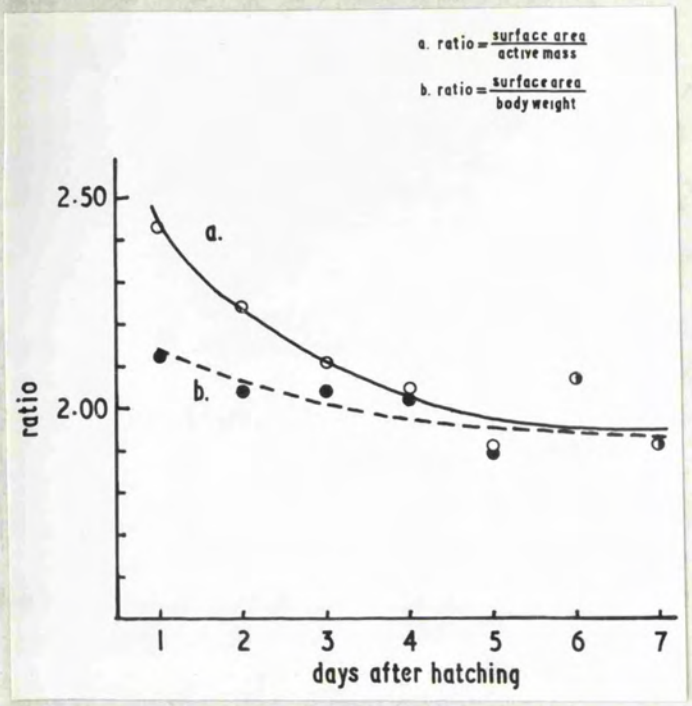



Fig. 31. The relationship between surface area, body weight or active mass and age. Active mass is defined as body weight minus yolk weight. Each point is the mean of 5 observations. It will be seen that the active mass of the chick increases at a greater rate than the surface area, whilst body weight and surface area are almost proportional throughout the period of observation.

temperature on the fifth day was significant (P = 0.05).

The daily metabolic rate and the body temperature of batch 1 are shown in fig. 30. The fall in the body temperature on the fifth day was paralleled by a fall in the metabolic rate. This probably coincided with the exhaustion of the yolk reserves and with a change from an essentially fat metabolism to a mixed metabolism. It will be seen that the two curves are very similar, suggesting that they are related.

b) Surface area and mass

The results are summarized in table 30 and illustrated in fig. 31. Whereas there was only a slight fall in the ratio of surface area to body weight (curve b), there was a definite and progressive fall in the ratio between surface area and active mass (curve a). This indicates that the active mass rather than the body weight, increased at a greater rate than the surface area.

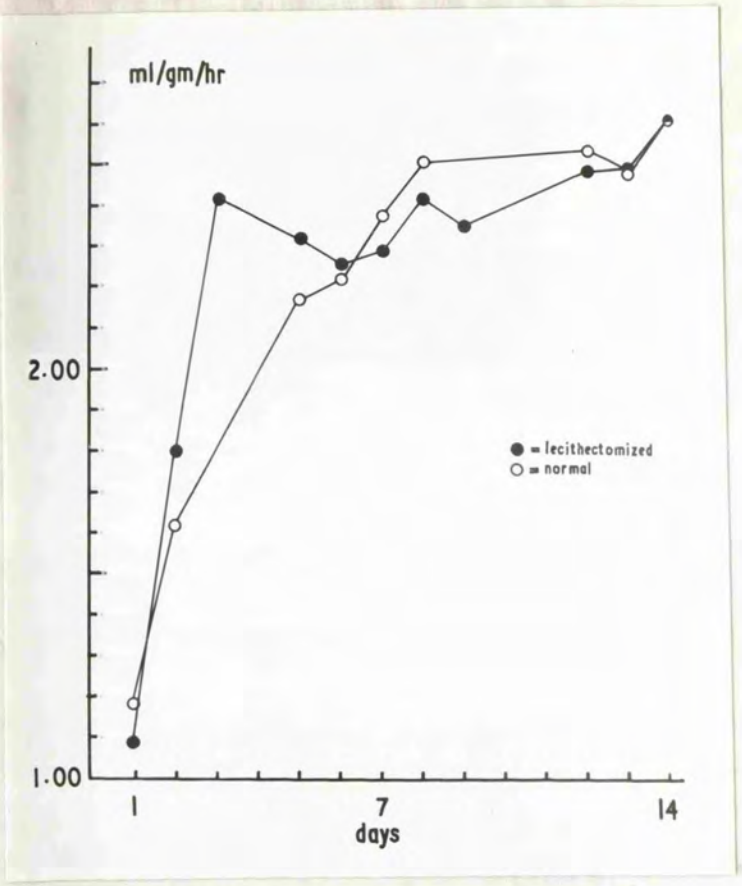



Fig. 32. The daily metabolic rate of lecithectomized and normal chicks. Each point is the mean of the daily determinations.

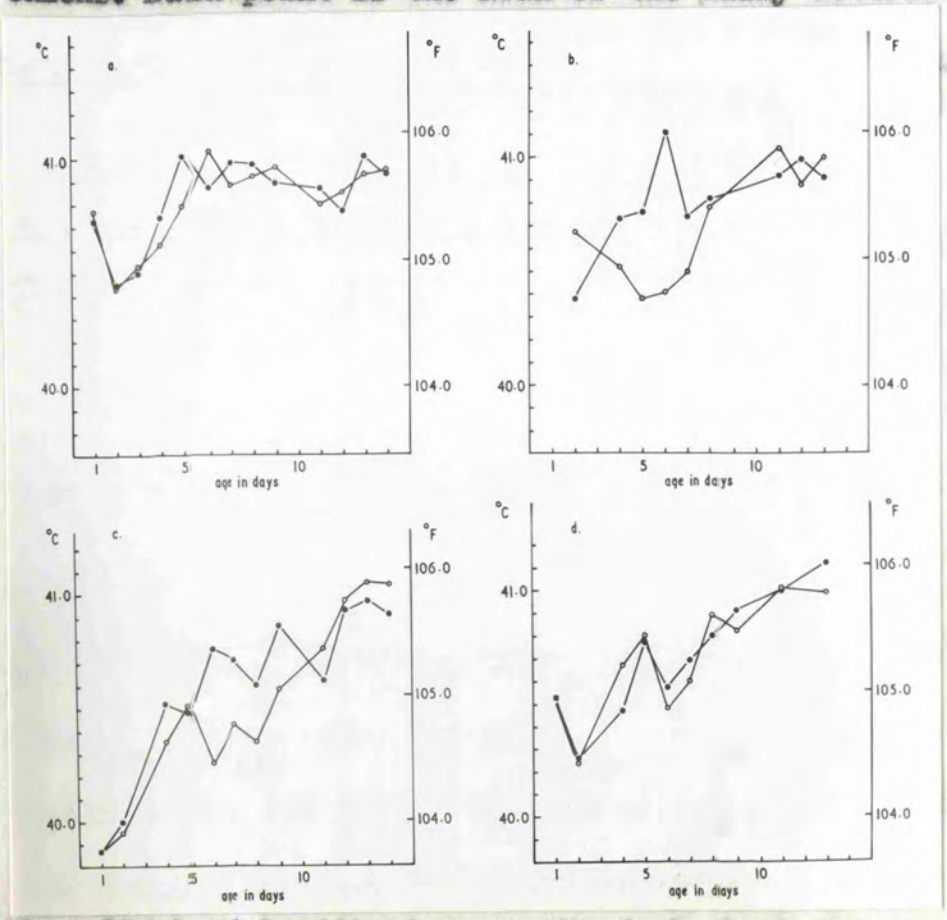



Fig. 33. The effect of lecithectomy on the body temperature of 4 consecutive batches of chicks.

## III.3 The effect of lecithectomy on the metabolism and body temperature

In the above experiment it was suggested that the fall in the metabolic rate on the fifth day might be due to a change in metabolism brought about by the exhaustion of the fat store. The effect of lecithectomy upon oxygen uptake was therefore investigated. RIR x LS chicks were used; 26 birds, from four consecutive batches from a single mating were lecithectomized on the first day after hatching. Another 26 normal birds, hatched at the same time, acted as controls.

The temperatures at which the oxygen consumption was determined were as follows:

| Weight range | T°C |
|---|---|
| to 100 gms | 35 |
| 100 - 150 | 33 |
| 150 - 200 | 31 |
| 200 - 250 | 29 |
| 250 - | 27 |

Results

a) The metabolic rate and body temperature during the first fortnight

Both groups showed a rise in the metabolic rate to about the eighth day, that of the lecithectomized group was slightly greater up to the fifth day, but not significantly greater at any time (fig. 32). Lecithectomy did not affect the growth rate (fig. 35a).

The body temperature curves of the four batches are given

| Age (days) | Lecithec. | Normal |
|---|---|---|
| 1 | 40.48±0.50 | 40.48±0.50 |
| 2 | 40.32±0.34 | 40.27±0.31 |
| 3 | 40.53±0.34 | 40.50±0.33 |
| 4 | 40.54±0.28 | 40.61±0.18 |
| 5 | 40.62±0.27 | 40.75±0.25 |
| 6 | 40.54±0.39 | 40.83±0.26 |
| 7 | 40.60±0.25 | 40.78±0.24 |
| 8 | 40.74±0.28 | 40.80±0.15 |
| 9 | 40.79±0.26 | 40.89±0.15 |
| 11 | 40.90±0.24 | 40.85±0.23 |
| 12 | 40.90±0.16 | 40.90±0.20 |
| 13 | 40.99±0.15 | 41.00±0.19 |
| 14 | 40.99±0.15 | 40.99±0.13 |

Table 31. The effect of lecithectomy upon the mean body temperature of 26 birds during the first fortnight of post-embryonic life. Figures in $^{o}C \pm SD$.

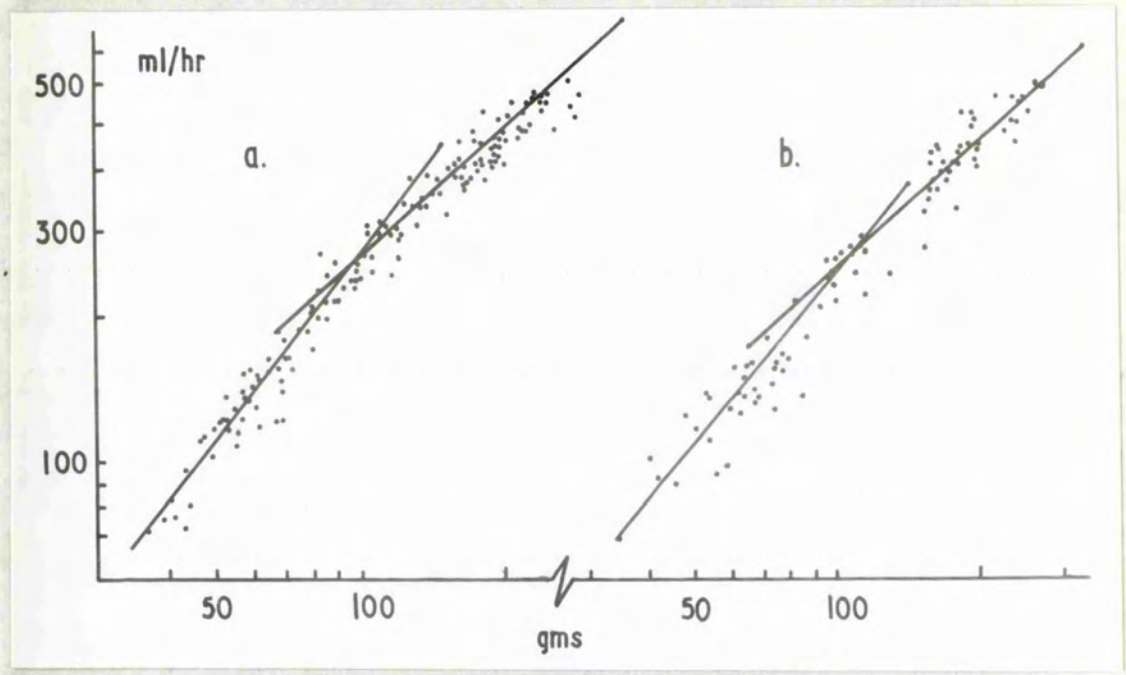



Fig. 34. Oxygen requirements of lecithectomized and normal birds during the first month of post-embryonic life.
a = lecithectomized; b = normal chicks.

| | Metabolic phase 1 | Metabolic phase 2 |
|---|---|---|
| Lecithectomized | $y = 0.504x^{1.376}$ | $y = 4.789x^{0.874}$ |
| Normal | $y = 1.035x^{1.185}$ | $y = 4.500x^{0.878}$ |

Table 32. Regression equations for the oxygen consumption curves of lecithectomized and normal RIR x LS chicks. See also fig. 34.

separately in fig. 33 and the mean values in table 31. It will be noted that the intergroup variation was quite large, but that the intragroup variation became progressively less with age. The abnormally high body temperature on the first day may have been due to slight hyperthermia as body temperatures were determined immediately after the chicks were removed from the incubator. It was noted in a previous section (see also below) that the upper critical temperature of the hatched bird is 3°C below the temperature of the incubator and consequently hyperthermia may well have resulted.

The body temperatures of the lecithectomized birds, although similar throughout the whole period of observation, did fall below those of the control chicks between the fifth and eighth days after hatching. The difference was significant ($P < 0.05$) on the sixth day only.

b) Effect of lecithectomy on the oxygen requirements and growth: Group E

The results are shown in fig. 34 and are given fully in table IX. The regression equations were similar (table 32) showing that lecithectomy had no effect on the absolute oxygen requirements during the month of observation. The metabolic pattern was also unaffected, as was the growth rate (fig. 35b). In both groups the metabolic rate became constant on about the thirteenth day after hatching.

It seems likely, therefore, that the yolk is not an essential source of energy after hatching. Furthermore, the rise in metabolic rate, although accentuated by the progressive reduction in the amount

Fig. 35a.

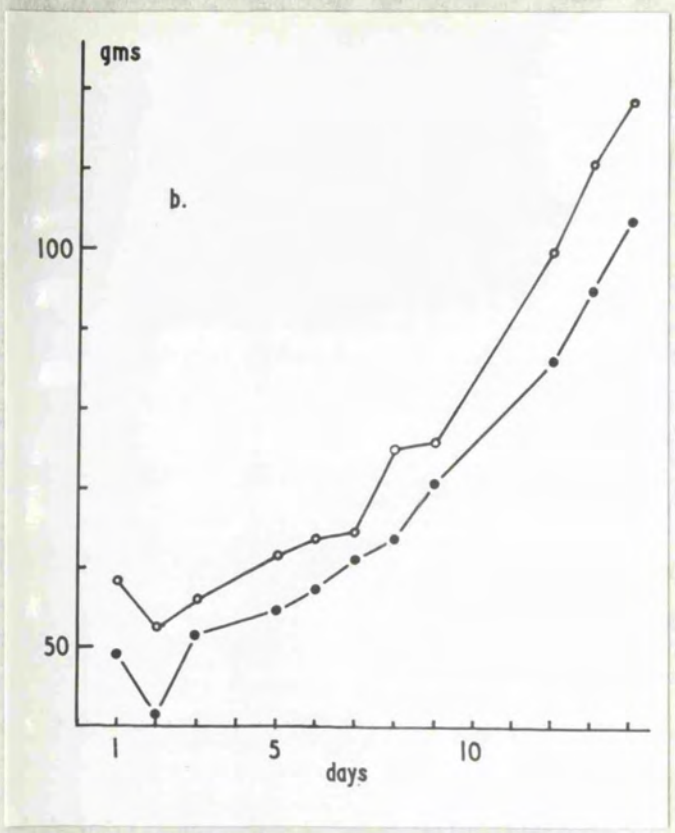



Fig. 35b.

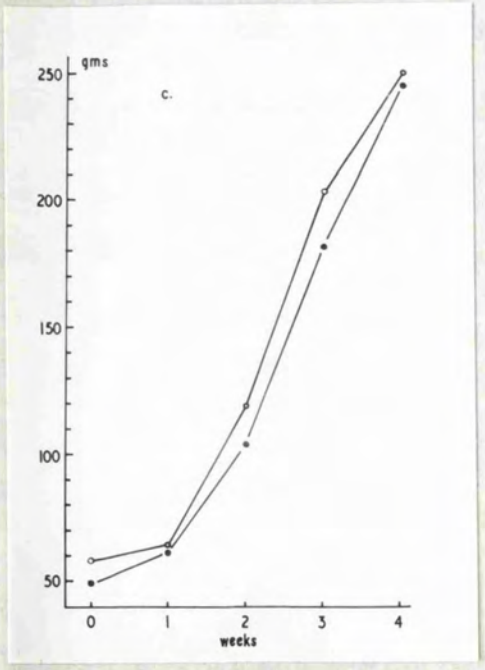



Fig. 35. Growth curves of lecithectomized and normal chicks.
a) 0 - 2 weeks. b) 0 - 4 weeks.
It will be seen that the growth rate was unaffected by lecithectomy.

of yolk, would appear, from these results, to be a definite phenomenon since it was not abolished by lecithectomy.

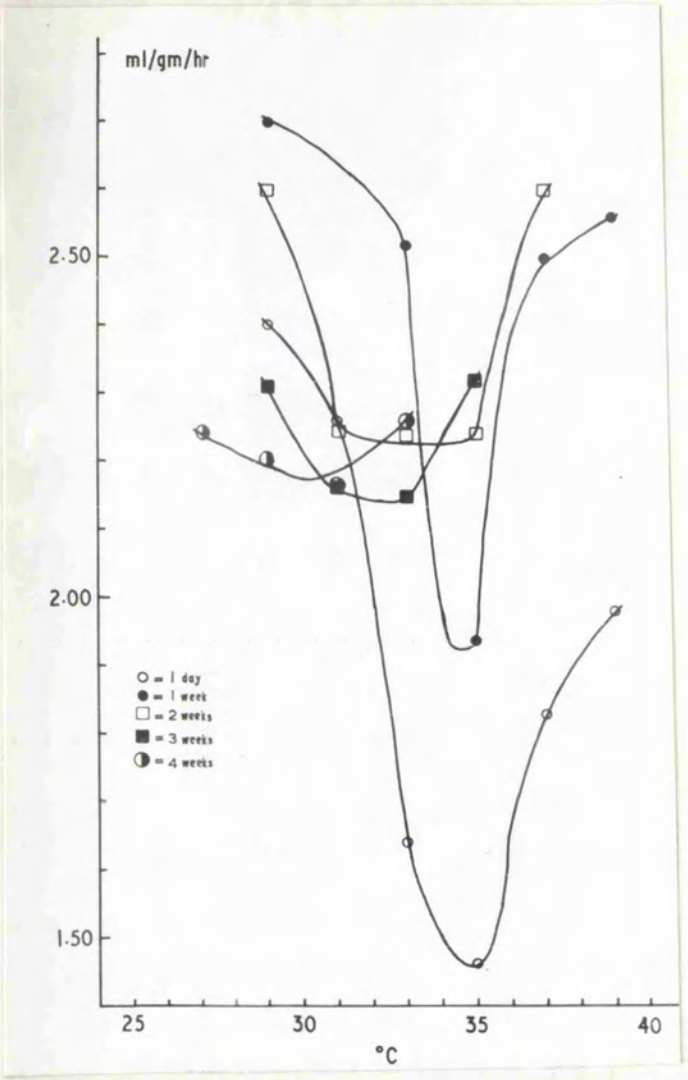



Fig. 36. The effect of temperature upon the metabolism of the growing fowl (0 - 4 weeks). Each point represents the mean of a variable number of observations (see table 33). The rise in metabolic rate during the first fortnight is evident in this figure.

## III.4 The effect of temperature on the resting metabolism of the fowl

From a practical husbandry point of view it seems likely that the greatest changes in the zone of thermal neutrality occur during the first month after hatching; published data, however, show that the zone of thermal neutrality alters little. The whole matter was therefore reinvestigated. The standard ration was available *ad libitum*. The chicks (RIR x LS) were kept in brooders with supplementary heating so placed to maintain a temperature gradient within the cages, allowing the birds to place themselves in their optimal environment.

### Results

The results are summarized for the five age groups in table 33 and illustrated in fig. 36.

The zone of thermal neutrality for the day-old chick was found to be very narrow. The rise in the metabolic rate between 35° and 37°C was highly significant ($P < 0.01$) but that between temperatures of 35° and 33°C was not significant. However, a highly significant rise between 35° and 31°C suggested that the lower critical temperature was certainly not less than 33°C, and, allowing for the fairly high degree of variation at this age, even closer to 35°C.

At 1 week of age a rise in temperature from 35° to 37°C resulted in a highly significant rise in the metabolic rate ($P < 0.01$) and a lowering of the temperature to 33°C gave a similar result, although the degree of significance was slightly reduced.

| Age (days) | Environmental temperature °C | | | | | | |
|---|---|---|---|---|---|---|---|
| | 39 | 37 | 35 | 33 | 31 | 29 | 27 |
| 1 | 1.98±0.01(15) | 1.83±0.06(9) | 1.46±0.04(12) | 1.64±0.09(18) | 2.27±0.11(9) | 2.40±0.18(6) | |
| 7 | 2.56±0.08(10) | 2.50±0.04(48) | 1.64±0.03(50) | 2.52±0.11(7) | | 2.70±0.13(6) | |
| 14 | | 2.60±0.04(26) | 2.24±0.03(46) | 2.24±0.04(50) | 2.25±0.05(33) | 2.60±0.05(9) | |
| 21 | | | | 2.15±0.04(41) | 2.16±0.05(20) | 2.31±0.09(5) | |
| 28 | | | | 2.26±0.11(11) | 2.17±0.10(6) | 2.26±0.05(7) | 2.24±0.08(9) |

Table 33. The effect of temperature upon the resting metabolic rate of the fowl (0 - 4 weeks of age).

Figures in ml.$O_2$/gm/hr ± SE (number of observations).

| Age | Zone |
|---|---|
| 1 day | 35 |
| 1 week | 34 - 35 |
| 2 weeks | 31 - 35 |
| 3 weeks | 30 - 33 |
| 4 weeks | 26 - 32 |

Table 34. The zones of thermal neutrality of the fully fed fowl during the first month after hatching. Figures in °C.

The variation in metabolism within the groups of 2 week old chicks at 35°, 33° and 31°C was much greater than the variation between the groups. Therefore the statistically significant difference ($P < 0.01$) between groups was not thought to be biologically significant.

The lower critical temperature of 3 week old birds was certainly between 31° and 29°C, whilst at 4 weeks it was probably a little below 27°C. Intragroup variation in birds of this age was considerably smaller than in younger chicks.

The zones of thermal neutrality for the age groups considered are given in table 34.

The rise in the metabolic rate during the first week or so was also evident (fig. 36). The metabolic rate, when measured at temperatures within the zones of thermal neutrality, was virtually constant from 2 to 4 weeks of age. It will be noted that the metabolic response to temperature decreased with age.

## III.5 The oxygen requirements of the fowl during the period of rapid growth

The only work on the oxygen requirements of the unstarved fowl from hatching to sexual maturity has been carried out by Kibler & Brody (1944). Unfortunately they failed to describe the exact parameters of their experiments. Here the requirements of a commercial laying strain have been determined using three different feeding programmes in order to determine the effect, if any, of the diet upon respiratory metabolism.

Three groups, totalling 87 birds, were used in these experiments. Except for the difference in the diet, experimental conditions were the same for all the groups. The results for each sex have been recorded separately. Food was available *ad libitum*.

The temperatures at which the determinations were carried out were based on the recommended figures of Russian workers for general husbandry (Gordon, 1960):

| Weight range | T°C |
|---|---|
| to 150 gm | 29 |
| 150 - 300 | 27 |
| 300 - | 21 |

The diets were quite different in composition: one was the standard ration (for formula see table 9, page 32); the other was a commercial broiler diet with an estimated crude protein content of 23% and a metabolizable energy content of 1300 kcals/lb. The exact formulation, however, was not available. The broiler diet

| Group | | Metabolic phase 1 | 2 | 3 | 4 |
|---|---|---|---|---|---|
| F | ♂ | $y = 0.403x^{1.511}$ | $y = 3.832x^{0.920}$ | $y = 91.73x^{0.397}$ | |
| | ♀ | $y = 0.403x^{1.511}$ | $y = 5.211x^{0.845}$ | $y = 6.985x^{0.782}$ * | $y = 149.55x^{0.279}$ *** |
| G | ♂ | $y = 1.650x^{1.137}$ | $y = 3.190x^{0.938}$ | $y = 203.03x^{0.181}$ | $y = 7.477x^{0.747}$ |
| | ♀ | $y = 1.650x^{1.137}$ | $y = 3.756x^{0.923}$ | $y = 34.06x^{0.501}$ | |
| H | ♂ | $y = 0.080x^{1.315}$ | $y = 4.776x^{0.873}$ | $y = 45.89x^{0.475}$ | |
| | ♀ | $y = 0.080x^{1.315}$ | $y = 5.636x^{0.830}$ | $y = 113.97x^{0.311}$ | |

* This is still part of the 2nd phase, the fall in the metabolic rate is intensified slightly.

*** Actually the 3rd phase; see note above.

Table 35. Equations of the regression lines of oxygen consumption of groups F, G and H.

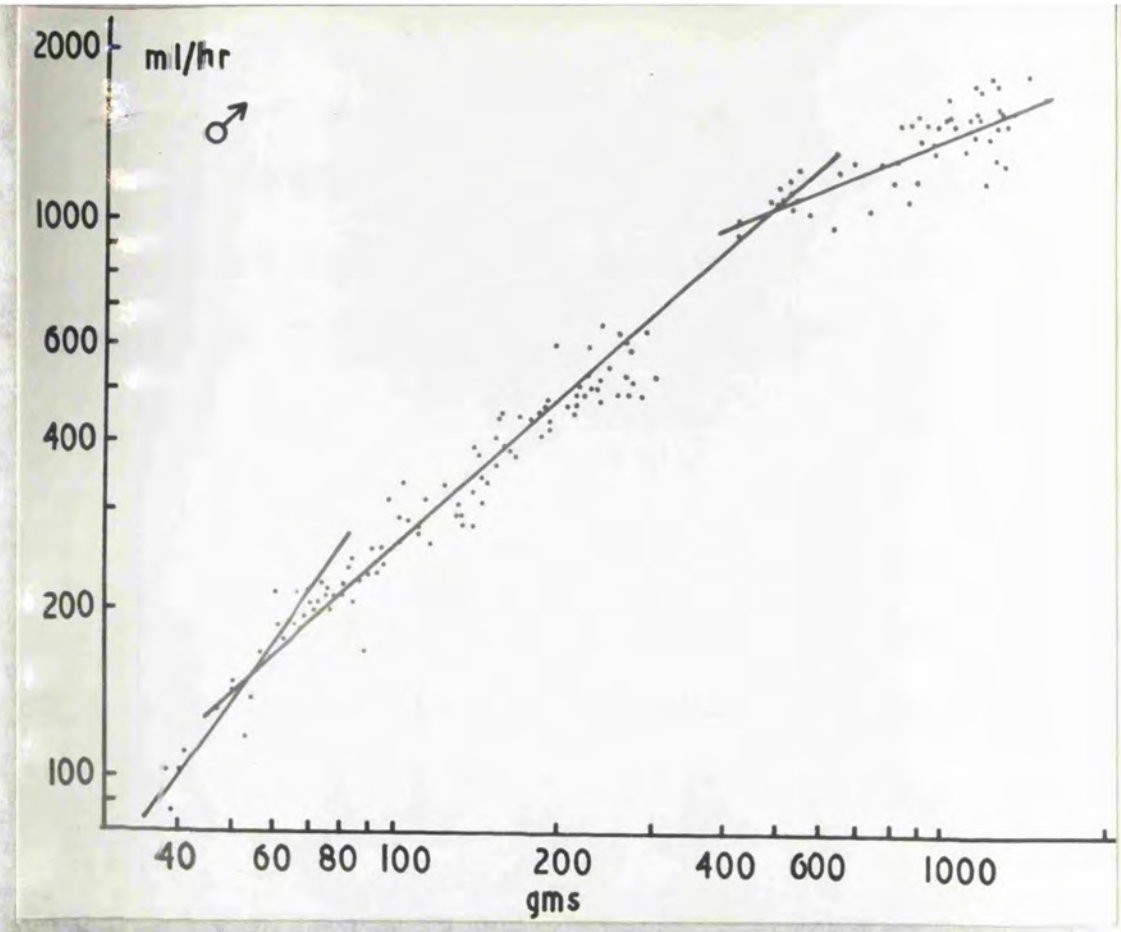



Fig. 37. Oxygen requirements of males of a laying strain fed the broiler ration: group F.

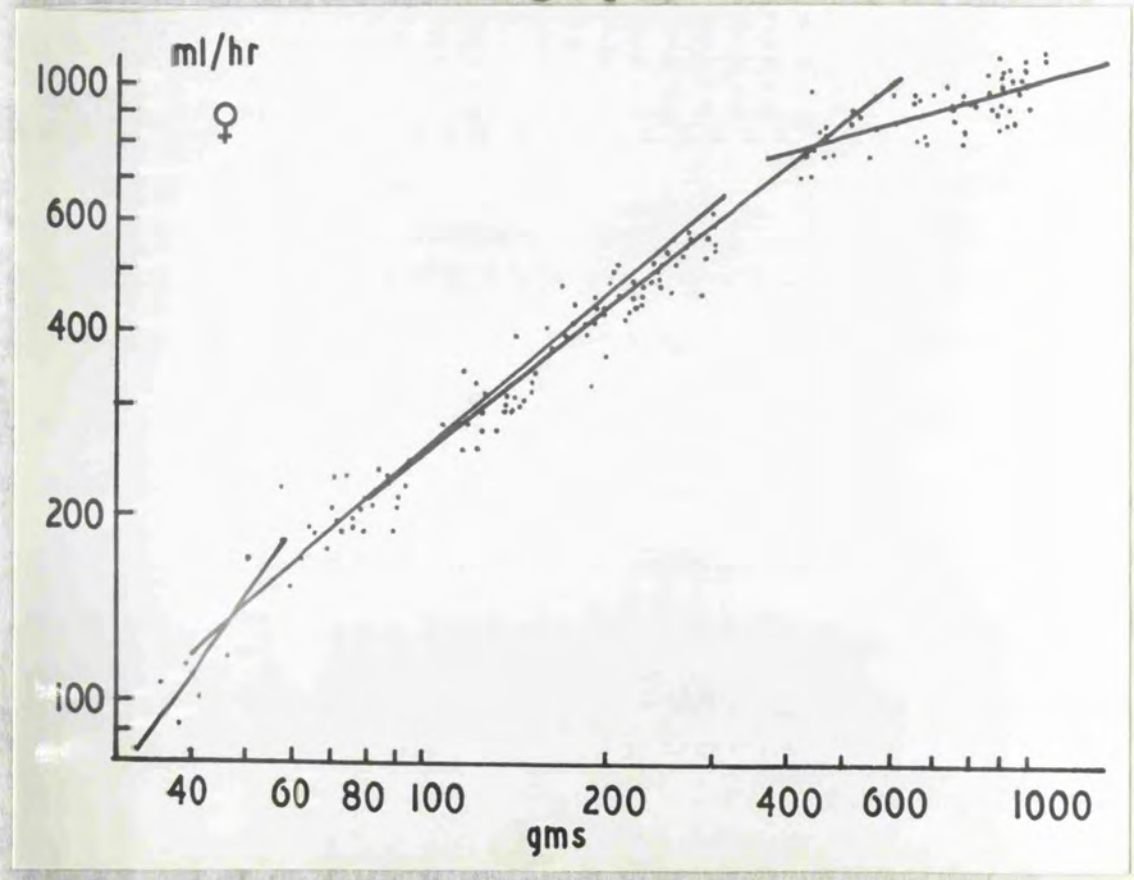



Fig. 38. Oxygen requirements of females of a laying strain fed the broiler ration: group F.

and the standard diet were fed throughout the period to groups F and G respectively, whilst with group H the standard diet was substituted for the broiler diet at the beginning of the third week.

Results

All the data are included in tables X, XI and XII.

a) The broiler diet: Group F (36 birds)

A rise in the metabolic rate during the first week after hatching was still evident in spite of the low environmental temperature. As no significant difference in the requirements of the sexes at this stage could be detected, the results have been amalgamated. During the second metabolic phase, when the metabolism and the growth were proportional (see table 35 for the regression equations) the males required progressively more oxygen than the females. At a body weight of about 150 gm it was found (statistically) that the metabolism of the females began to fall at a slightly greater rate. This change could not be detected in the males although there was some suggestion of a reduction in oxygen uptake at about 200 gm (fig. 37). At a body weight of 500 gm the rate of oxygen uptake, compared with the growth rate, was greatly reduced in both sexes (see figs. 37 and 38). This third metabolic phase continued to the end of the observations.

The growth curves are given in fig. 43b.

b) The standard diet: Group G (15 birds)

Again the metabolic rate rose immediately after hatching.

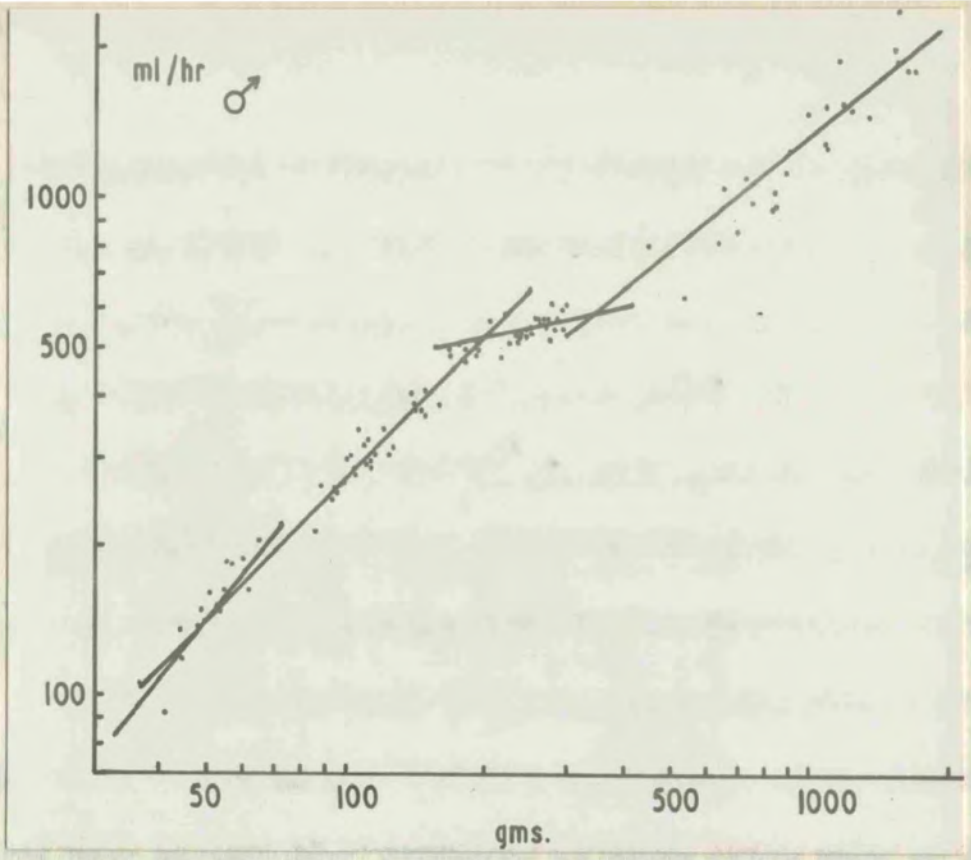



Fig. 39. Oxygen consumption curve of the males of a laying strain fed the standard ration: group G.

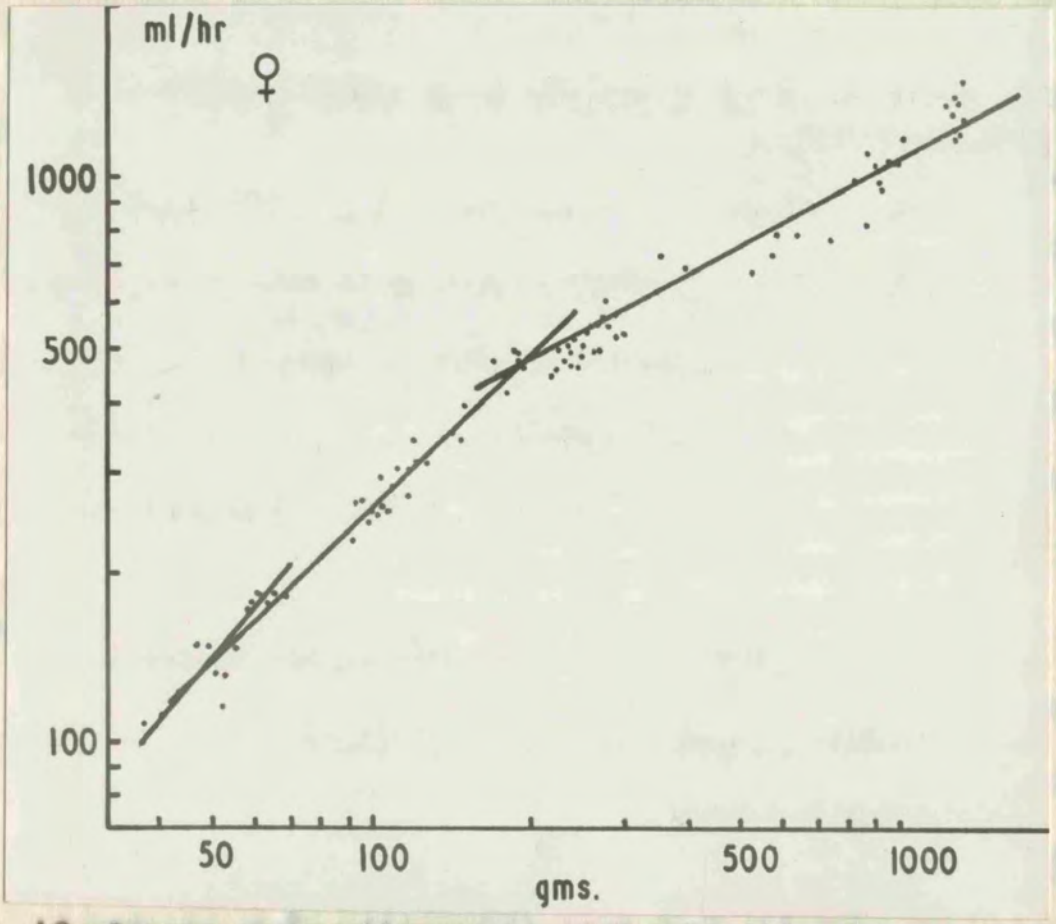



Fig. 40. Oxygen consumption curve of females of a laying strain fed the standard ration: group G.

During the second phase the rate was approximately constant for both sexes. At 200 gm body weight (3 weeks) (see fig. 43a) a third metabolic phase was initiated. The rate of oxygen uptake fell dramatically for both sexes, especially for the males. This phase for the males was, however, transient. Reference to fig. 39 shows that it was soon superseded by a fourth and final metabolic phase, when the rate of oxygen uptake increased again. In the females the third metabolic phase persisted to the end of the observational period (fig. 40). There were no significant sexual differences in absolute oxygen requirements in the weight period 200 - 500 gm although weight differences could be detected from 300 gm.

c) The broiler ration followed by the standard ration: Group H (36 birds)

The results are shown in figs. 41 and 42. The metabolic pattern was found to be essentially the same as that of birds fed the broiler diet for the whole observational period (group F), even though this diet had only been fed for two weeks. The third metabolic phase was initiated at a body weight of 400 gm in both sexes.

The course of sexual divergence in oxygen requirements was similar to that shown by group F. It was slight until the third phase of metabolism had begun.

d) A comparison of the absolute oxygen requirements at different weights

The oxygen requirements have been calculated for different

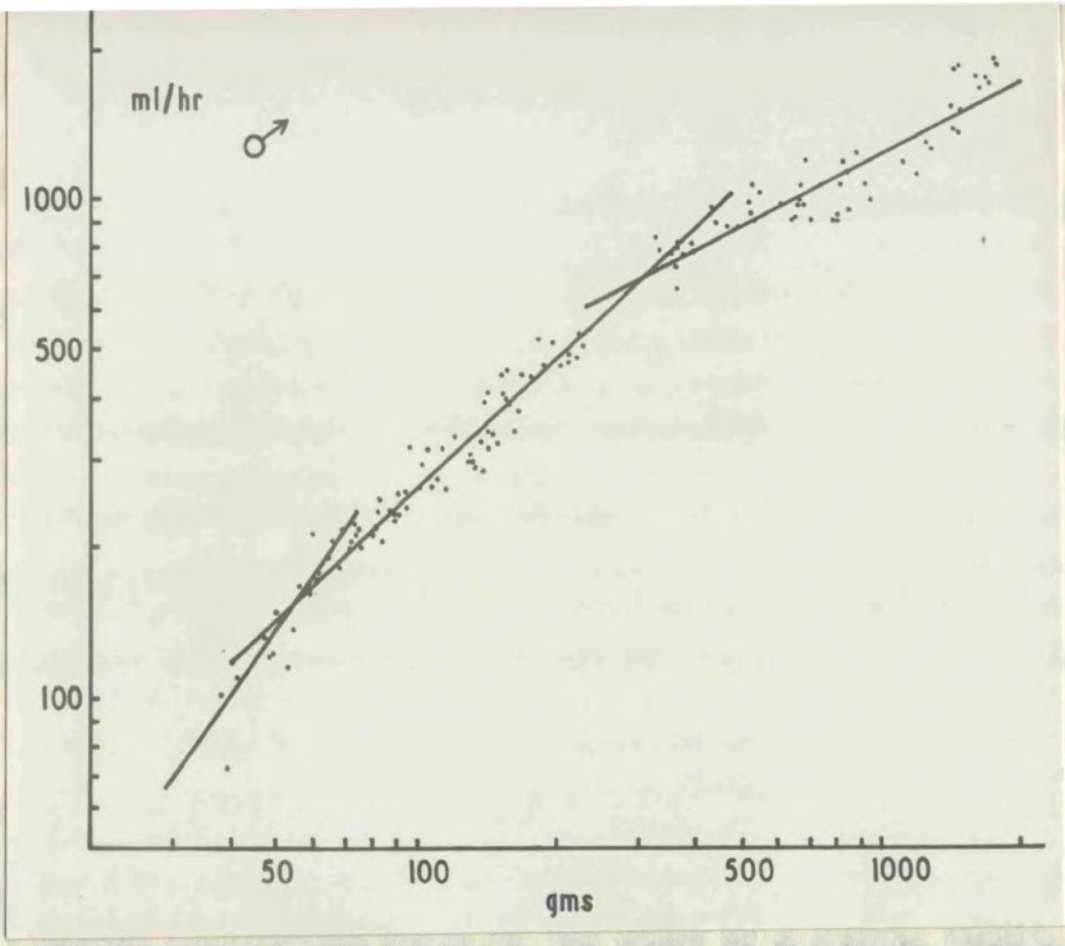



Fig. 41. Oxygen consumption curve of males of the laying strain fed the broiler diet for 2 weeks followed by the standard ration for the rest of the experiment: group H.

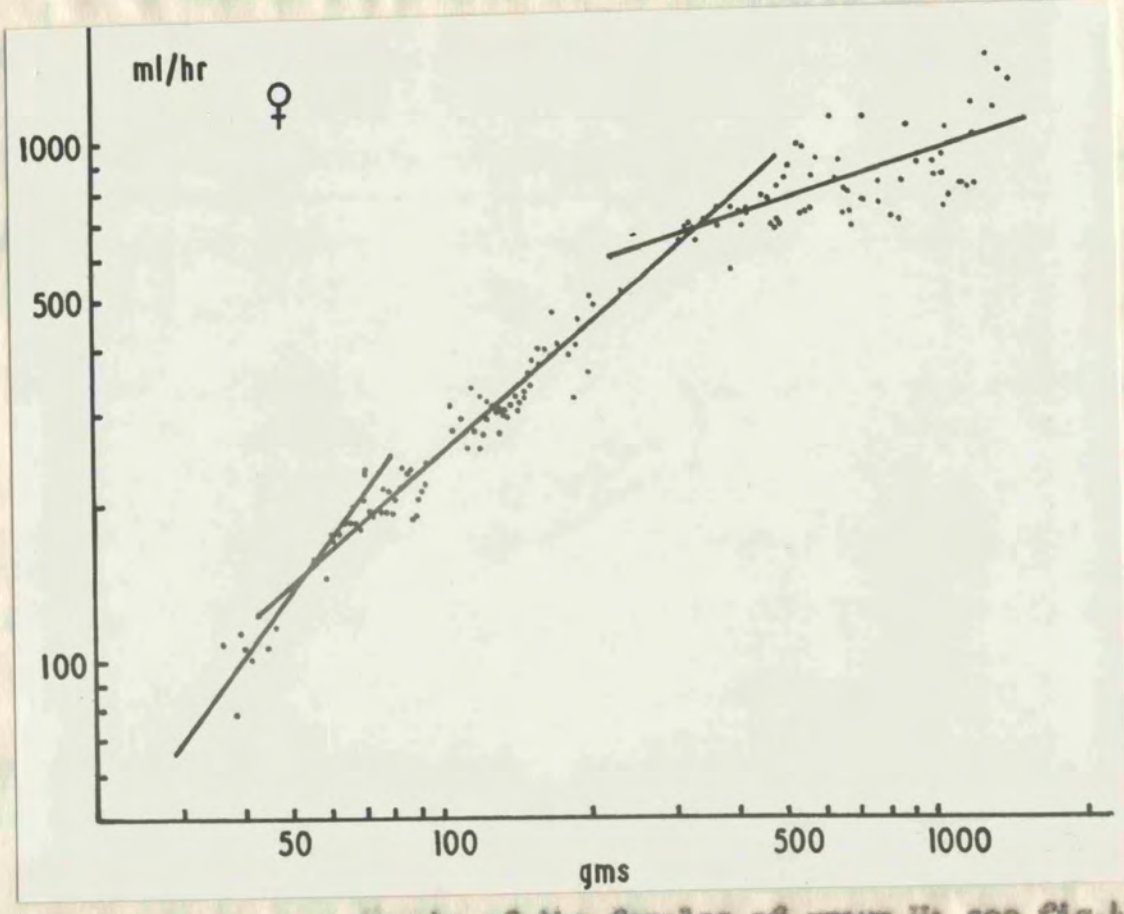



Fig. 42. Oxygen requirements of the females of group H: see fig.41.

| Wt. (gm) | F | | G | | H | |
|---|---|---|---|---|---|---|
| | ♂ | ♀ | ♂ | ♀ | ♂ | ♀ |
| 40 | 106 | 106 | 103 | 103 | 109 | 109 |
| 60 | 184 | 177 | 187 | 183 | 175 | 170 |
| 100 | 265 | 255 | 266 | 258 | 277 | 264 |
| 300 | 670 | 594 | 664 | 628 | 570 | 559 |
| 500 | 1097 | 847 | 862 | 775 | 764 | 709 |
| 1000 | 1431 | 1028 | 1221 | 977 | 1302 | 1084 |

Table 36. The effect of the diet upon the oxygen requirements of the laying strain at different body weights. Group F was fed the broiler diet; group G the standard diet and group H the broiler diet for 2 weeks and then the standard diet for the rest of the period of observation.

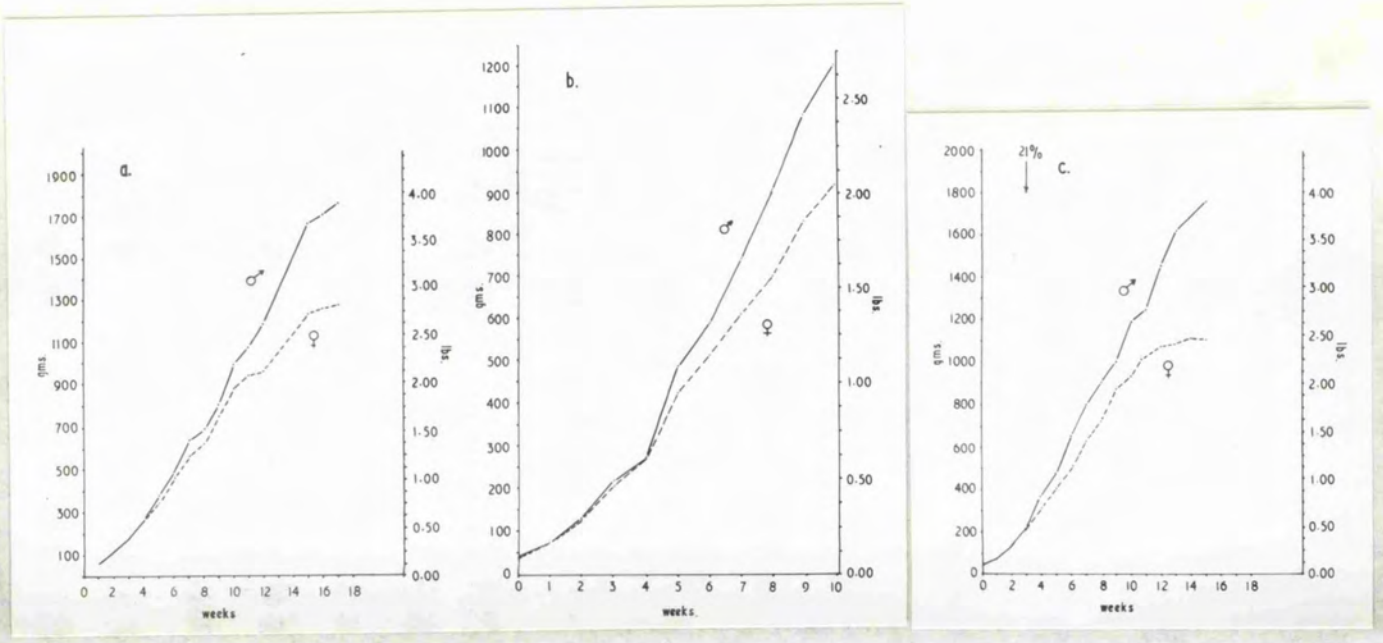



Fig. 43. Growth curves of the laying strain with reference to the diet. (a) Standard diet - group G; (b) Broiler diet - group F; (c) Broiler diet followed by the standard diet - group H.

body weights from the regression equations and are included in table 36. There was no significant difference between the requirements of the three groups during the first metabolic phase. Differences began to appear when the body weight exceeded 100 gm. In group H, where the standard diet was substituted at 210 gm for the broiler diet, oxygen requirements remained similar to those of the birds fed the broiler diet alone (group F) until about 400 gm body weight. During the third metabolic phase group F required more oxygen than group G, whilst group H males took an intermediate position. The females of group H, however, fell below the females of group G by about 10%.

| Ingredient | H.E.% | N.E.% |
|---|---|---|
| Oat feed | | 17.5 |
| Ground oats | | 20.0 |
| Ground wheat | 47.5 | 25.0 |
| Ground maize | 20.0 | |
| Soyabean meal (44%) | 20.0 | 25.0 |
| Fish meal (66%) | 5.0 | 5.0 |
| Grass meal | 2.5 | 2.5 |
| Unextracted dried yeast | 2.5 | 2.5 |
| Minerals | 2.5 | 2.5 |
| Vitamin A 4m i.u./ton | for both diets. | |
| Vitamin $D_3$ 1m i.u./ton | | |
| Analytical data | | |
| Crude protein % | 22.9 | 23.5 |
| Metabolizable energy kcals/lb | 1354 | 1000 |
| Oil (ether extract) % | 2.3 | 1.5 |
| Crude fibre % | 3.5 | 4.9 |
| IOM % | 11.4 | 17.6 |

Table 37. Formulae of the high energy (H.E.) and normal energy (N.E.) diets.

| Age (days) | °C |
|---|---|
| 1 - 7 | 35 |
| 8 - 14 | 33 |
| 15 - 18 | 31 |
| 19 - 21 | 29 |
| 22 - 28 | 27 |
| 29 - 35 | 25 |
| 36 - 49 | 23 |
| 50 - 56 | 21 |

Table 38. Temperatures at which the resting metabolism was determined in experiments III.6.i, ii and iii.

| Phase | High energy | Normal energy |
|---|---|---|
| 1 | $y = 0.0232x^{2.083}$ | $y = 0.0049x^{2.507}$ |
| 2 | $y = 5.591x^{0.767}$ | $y = 2.165x^{0.880}$ |
| 3 | $y = 59.494x^{0.328}$ | $y = 200.2x^{0.086}$ |
| 4 | $y = 2.776x^{0.879}$ | $y = 2.532x^{0.900}$ |
| 5 | $y = 29.171x^{0.506}$ | $y = 9.238x^{0.335}$ |

Table 39. Equations for the regression lines for group I (Figs. 44 and 45).

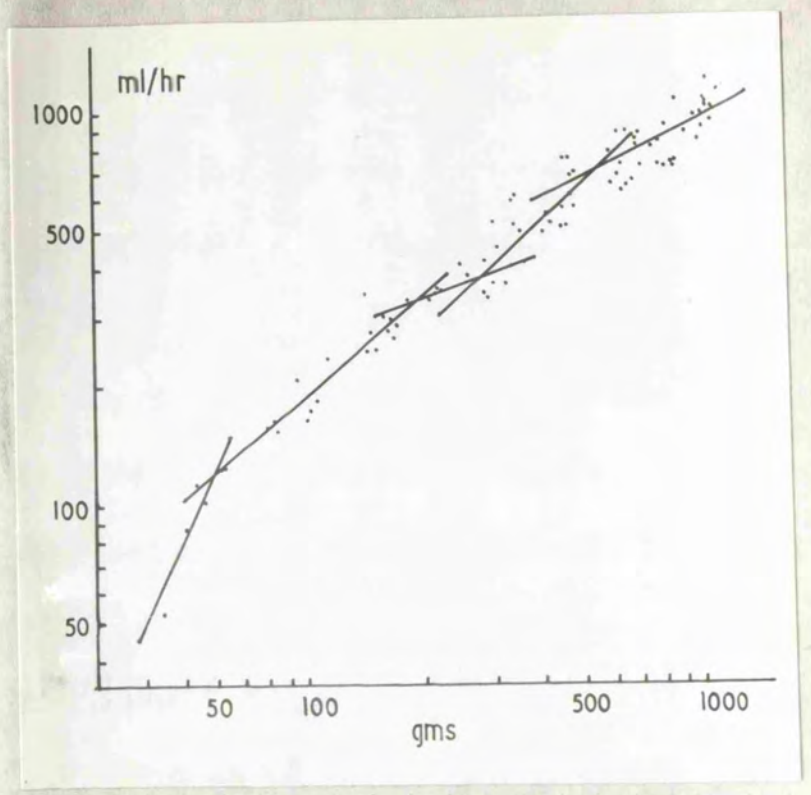



Fig. 44. The metabolic pattern of RIR x LS ♂♂ fed a high energy diet: group I.

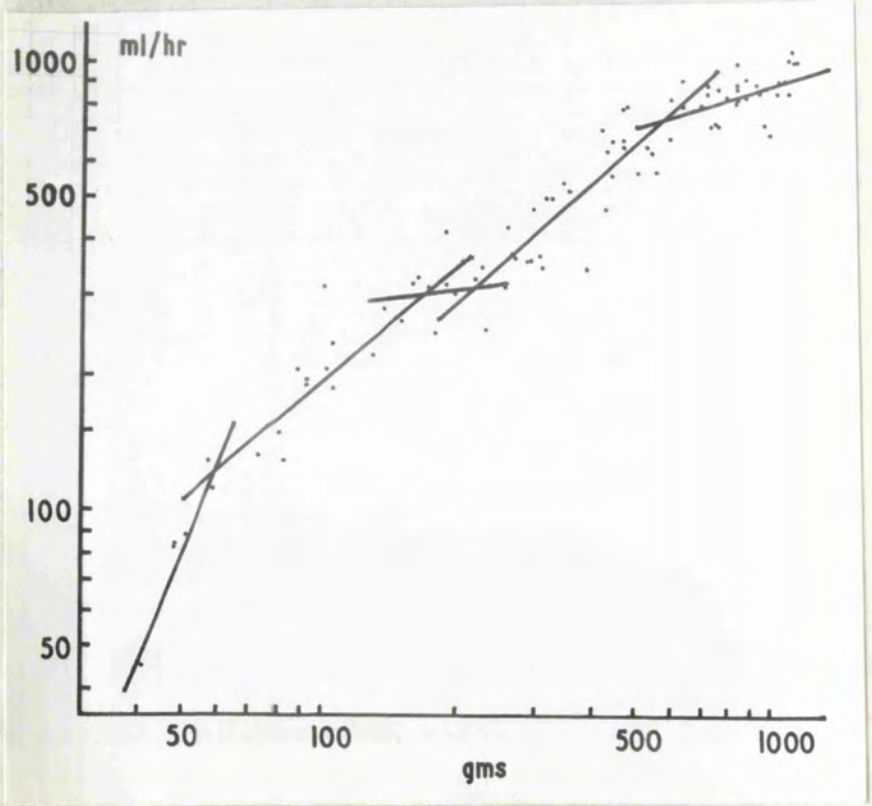



Fig. 45. The metabolic pattern of RIR x LS ♂♂ fed a normal energy diet: group I. Note the similarity of this pattern to that of birds fed the high energy diet (fig. 44).

## III.6 The effect of certain dietary factors upon the metabolism of the growing male chicken

### III.6.i The effect of the calorific content of the diet: Group I

In the above experiments it was found that the diet had a pronounced effect upon both the absolute volume of oxygen required by the bird and the metabolic pattern. Furthermore the sexes behaved differently.

The dietary factors, protein, fat and metabolizable energy content were therefore investigated singly in order to determine the effect of each upon oxygen consumption of the growing male chicken. Here the latter factor was investigated.

Day-old chicks (RIR x LS) were divided into two groups of twelve. Group A was fed a high energy diet whilst group B was fed a normal energy diet. Food and water were available ad libitum. The formula and the analytical data of these two diets are set out in table 37.

The temperatures at which oxygen requirements were determined are given in table 38.

Results

The data of oxygen consumption are given in table XIII and are shown in figs. 44 and 45. On analysis it was found that the metabolic pattern was not significantly influenced by the calorific content of the diet. Five metabolic phases could be differentiated for both groups. During the first phase oxygen uptake increased at a greater rate than the body weight. From 60 gm to about 150 gm

| Age (days) | High energy diet n | ml/gm/hr±SD | Normal energy diet n | ml/gm/hr±SD |
|---|---|---|---|---|
| 1 | 3 | 1.51 ± 0.20 | 3 | 1.57 ± 0.31 |
| 7 | 7 | 1.91 ± 0.20 | 7 | 1.98 ± 0.17 |
| 14 | 3 | 1.92 ± 0.21 | 4 | 2.09 ± 0.12 |
| 21 | 8 | 1.94 ± 0.30 | 7 | 1.79 ± 0.16 |
| 28 | 8 | 1.35 ± 0.15*** | 8 | 1.43 ± 0.17* |
| 35 | 9 | 1.35 ± 0.17 | 9 | 1.44 ± 0.25 |
| 42 | 8 | 1.16 ± 0.15 | 8 | 1.23 ± 0.06 |
| 49 | 6 | 1.02 ± 0.14 | 6 | 1.09 ± 0.07 |
| 56 | 7 | 0.99 ± 0.05 | 9 | 0.88 ± 0.02 |

* $P < 0.01$; *** $P < 0.001$.

Table 40. The effect of age and the calorific content of the diet upon the metabolic rate of the growing RIR x LS ♂ fowl.

| Age (weeks) | High energy Food intake | High energy Wt. (gm) | Normal energy Food intake | Normal energy Wt. (gm) |
|---|---|---|---|---|
| 0 | | 43 | | 39 |
| 1 | 110 | 55 | 105 | 51 |
| 2 | 343 | 98 | 292 | 93 |
| 3 | 658 | 167 | 500 | 164 |
| 4 | 1003 | 273 | 701 | 283 |
| 5 | 1415 | 464 | 1026 | 442 |
| 6 | 1975 | 637 | 1656 | 663 |
| 7 | 2553 | 835 | 2246 | 851 |
| 8 | 3227 | 1031 | 2980 | 1060 |

Table 41. The effect of the calorific content of the diet upon food consumption and growth rate during the 8 weeks after hatching: group I

body weight both the oxygen consumption and the body weight increased proportionately. There then followed a short period of approximately constant oxygen uptake - from 170-210 gm for the normal energy group and from 190-250 gm for the high energy group. The rate of uptake increased greatly again until a weight of 500 gm had been attained in both groups. Finally there was a phase of a reduced rate of oxygen uptake. The equations of the regression lines are given in table 39.

The influence of age and diet upon the metabolic rate is shown in table 40. The pattern was little affected by the diet, the only difference being a significant fall ($P < 0.05$) in the rate between the second and third weeks in birds fed the normal energy diet. Both groups showed significant weekly falls in metabolic rate from the third week, with the exception of the fifth week and at 7 and 8 weeks in the high energy group.

It is interesting to note that the group fed the normal energy diet should have a consistently, though not significantly, higher metabolic rate during the greater part of the experimental period. Only at 8 weeks of age was a significant difference measurable ($P < 0.05$) when the high energy group had the higher metabolic rate.

In spite of a somewhat greater intake of the high energy diet, the two groups had almost indistinguishable body weights throughout the experiments (see table 41).

The very significant fall ($P < 0.001$, high energy group; $P < 0.01$, normal energy group) in the metabolic rate between the

| Ingredient | H.F. % | N.F. % |
|---|---|---|
| Ground wheat | 34.0 | 47.5 |
| Ground maize | 10.0 | 20.0 |
| Middlings | 11.5 | |
| Soyabean meal (44%) | 20.0 | 20.0 |
| Fish meal (66%) | 5.0 | 5.0 |
| Unextracted dried yeast | 2.5 | 2.5 |
| Fat (stabilized tallow) | 12.0 | |
| Minerals | 2.5 | 2.5 |
| Vitamin A 4m i.u./ton<br>Vitamin $D_3$ 1m i.u./ton | for both diets | |
| **Analytical data** | | |
| Crude protein % | 21.8 | 21.7 |
| Metabolizable energy kcals/lb. | 1347 | 1354 |
| Oil (ether extract) % | 6.8 | 2.3 |
| Crude fibre % | 4.5 | 3.2 |
| IOM % | 13.7 | 11.3 |

Table 42. Formulae of the high fat (H.F.) and normal fat (N.F.) diets.

| Phase | High fat diet | Normal fat diet |
|---|---|---|
| 1 | $y = 0.0195x^{2.149}$ | $y = 0.320x^{1.438}$ |
| 2 | $y = 5.314x^{0.806}$ | $y = 2.822x^{0.936}$ |
| 3 | $y = 87.370x^{0.274}$ | $y = 89.894x^{0.265}$ |
| 4 | $y = 0.0017x^{2.020}$ | $y = 0.0459x^{2.017}$ |
| 5 | $y = 240.9x^{0.227}$ | $y = 37.585x^{0.514}$ |

Table 43. Regression equations for the oxygen consumption curves of group J: see figs. 46 and 47.

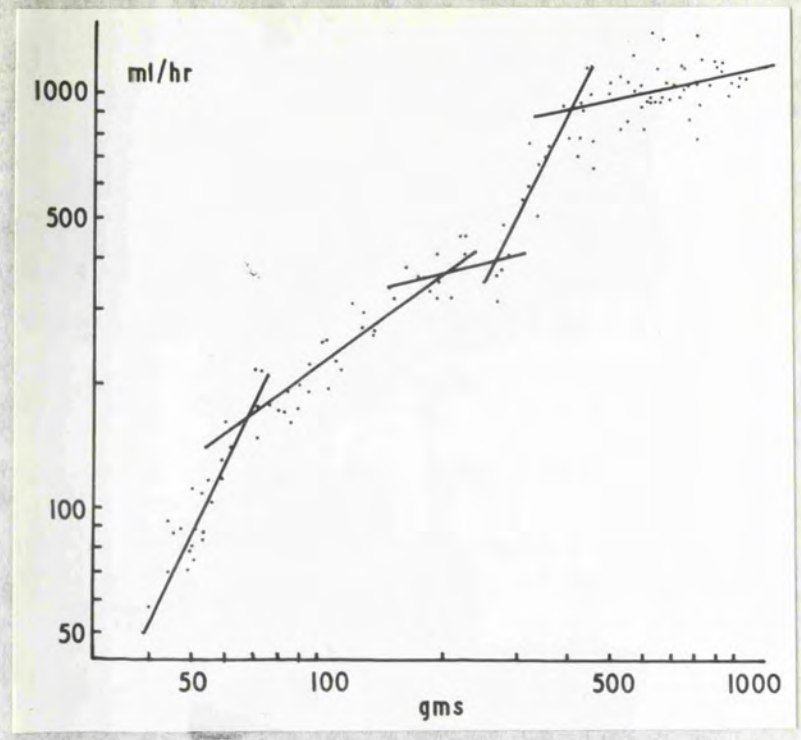



Fig. 46. The oxygen consumption curve of RIR x LS ♂♂ fed a high fat diet: group J.

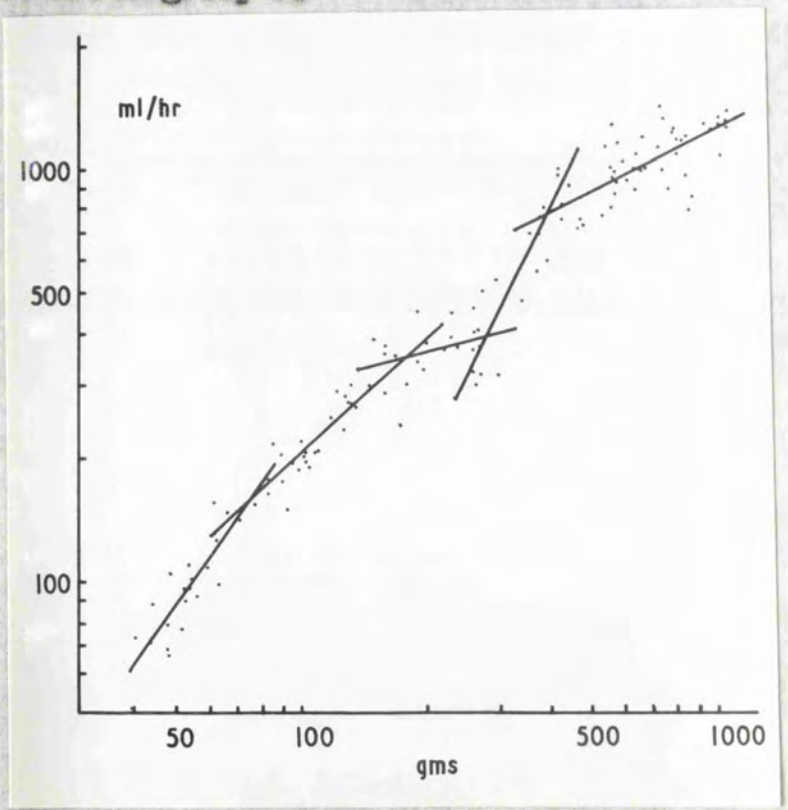



Fig. 47. The oxygen consumption curve of RIR x LS ♂♂ fed a normal fat diet: group J.

third and fourth week is of great interest but cannot be explained. A definite physiological change is certainly indicated.

III.6.ii The effect of the fat content of the diet: Group J

Two levels of fat were compared in this experiment whilst the metabolizable energy and protein levels were the same (table 42).

The high fat content was realized by using a special stabilized form of tallow. The analytical data of the diets are also given in table 42. Food consumption was determined weekly.

The temperatures at which the resting oxygen uptake of the 36 RIR x LS male chicks was determined are given in table 38.

The weights of the thyroid and adrenal glands were determined at 3, 5 and 8 weeks from birds selected at random from each group.

Results

The data are presented in table XIV. The metabolic patterns of the two groups are shown in figs. 46 and 47, and the equations of the regression lines are given in table 43. It will be noted that the fat content of the diet did not have any effect upon the metabolic pattern and had only a slight effect on the absolute oxygen requirements during the latter phases of the experiment. Growth was not consistently affected (table 44) although the birds on the high fat diet consumed about 10% more food during the 8 weeks (table 44).

Apart from a significant ($P < 0.01$) fall in the absolute thyroid weight of the high fat group at 5 weeks, there was no other measurable effect on either thyroid or adrenal weight (see table 45).

The effect of age on the metabolic rate is summarized in

| Age (weeks) | High fat diet Food (gm) | Wt. (gm) | Normal fat diet Food (gm) | Wt. (gm) |
|---|---|---|---|---|
| 0 | | 42 | | 41 |
| 1 | 44 | 56 | 40 | 57 |
| 2 | 175 | 88 | 167 | 101 |
| 3 | 352 | 148 | 336 | 163 |
| 4 | 704 | 280 | 612 | 257 |
| 5 | 1162 | 398 | 1001 | 409 |
| 6 | 1778 | 614 | 1541 | 601 |
| 7 | 2568 | 800 | 2305 | 832 |
| 8 | 3352 | 992 | 2921 | 1049 |

Table 44. The effect of the fat content of the diet upon the accumulative food consumption and growth of the RIR x LS male chick: group J.

| (weeks) | Group | n | Body weight (gm) | Thyroid weight (mg) | Thyroid weight (mg/100gm) | Adrenal weight (mg) | Adrenal weight (mg/100gm) |
|---|---|---|---|---|---|---|---|
| 3 | High fat | 6 | 136.0 ± 37.1* | 8.4 ± 3.8 | 6.0 ± 1.8 | 22.1 ± 3.4 | 18.1 ± 6.9 |
| | Normal fat | 6 | 148.9 ± 18.5 | 7.7 ± 2.3 | 5.3 ± 1.8 | 22.2 ± 5.4 | 14.9 ± 3.0 |
| 5 | High fat | 5 | 365.6 ± 36.5 | 21.5 ± 4.4** | 5.8 ± 1.2 | 50.6 ± 6.0 | 13.9 ± 2.0 |
| | Normal fat | 6 | 397.1 ± 42.5 | 25.4 ± 3.9 | 6.4 ± 1.3 | 53.1 ± 9.6 | 13.2 ± 1.9 |
| 8 | High fat | 5 | 992.0 ± 57.9 | 61.1 ± 16.1 | 0.06 ± 0.03 | 110.6 ± 17.0 | 0.11 ± 0.02 |
| | Normal fat | 6 | 1049.0 ± 34.2 | 65.0 ± 12.1 | 0.06 ± 0.02 | 110.4 ± 21.1 | 0.10 ± 0.03 |

* SD; ** $P < 0.01$

Table 45. The effect of dietary fat upon the body weight, thyroid and adrenal weights.

| Age (weeks) | n | High fat ml/gm/hr ± SD | n | Normal fat ml/gm/hr ± SD |
|---|---|---|---|---|
| 1 | 4 | 1.73 ± 0.18 | 4 | 1.99 ± 0.29 |
| 2 | 4 | 1.99 ± 0.35 | 4 | 2.05 ± 0.13 |
| 3 | 8 | 2.12 ± 0.27 | 8 | 1.83 ± 0.24 |
| 4 | 4 | 1.66 ± 0.23 | 5 | 1.55 ± 0.18 |
| 5 | 7 | 2.01 ± 0.36 | 7 | 1.66 ± 0.36 |
| 6 | 7 | 1.52 ± 0.70 | 7 | 1.71 ± 0.11 |
| 7 | 7 | 1.29 ± 0.22 | 7 | 1.34 ± 0.32 |
| 8 | 5 | 1.13 ± 0.24 | 5 | 1.32 ± 0.18 |

Table 46. The effect of dietary fat upon the metabolic rate of the growing fowl (RIR x LS ♂♂).

| Ingredient | H.P. % | N.P. % |
|---|---|---|
| Ground wheat | 20.0 | 47.5 |
| Ground maize | 40.0 | 20.0 |
| Soyabean meal (44%) | 15.0 | 20.0 |
| Fish meal (66%) | 17.5 | 5.0 |
| Unextracted dried yeast | 2.5 | 2.5 |
| Grass meal | 2.5 | 2.5 |
| Minerals | 2.5 | 2.5 |
| Vitamin A 4m i.u./ton<br>Vitamin $D_3$ 1m i.u./ton | for both diets | |
| Analytical data | | |
| Crude protein % | 26.1 | 21.7 |
| Metabolizable energy kcals/lb | 1357 | 1354 |
| Oil (ether extract) % | 3.7 | 2.5 |
| Crude fibre % | 3.1 | 3.4 |
| IOM % | 11.1 | 10.0 |

Table 47. Formulae of the high protein (H.P.) and low protein (L.P.) diets.

| Phase | High protein | Normal protein |
|---|---|---|
| 1 | $y = 0.397x^{1.396}$ | $y = 0.005x^{2.412}$ |
| 2 | $y = 3.199x^{0.932}$ | $y = 3.841x^{0.885}$ |
| 3 | $y = 53.873x^{0.413}$ | $y = 390.01x^{0.042}$ |
| 4 | $y = 1.099x^{1.032}$ | $y = 4.286x^{0.828}$ |

Table 48. Equations of the lines for the oxygen uptake of group K.

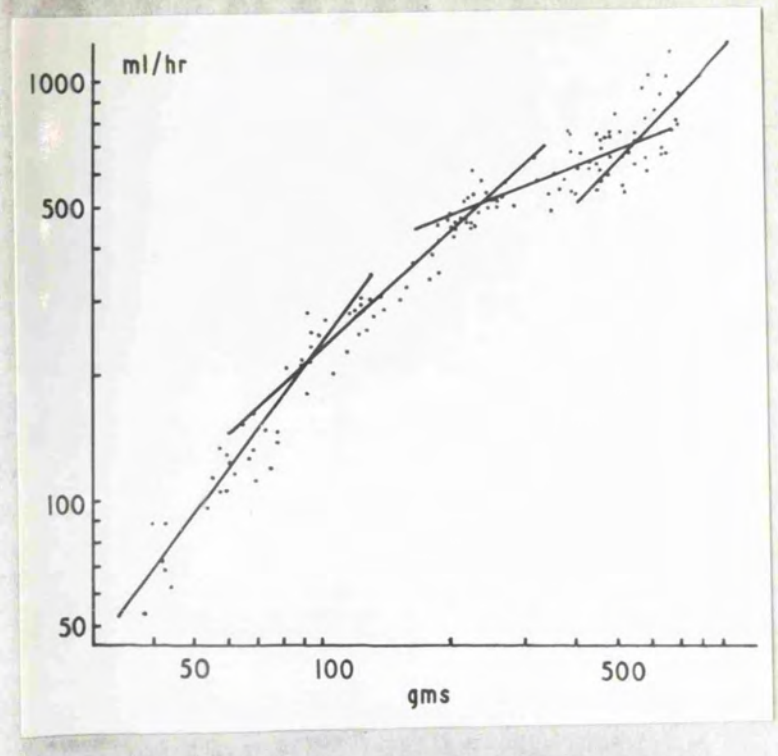



Fig. 48. Oxygen requirements of RIR x LS ♂♂ fed a high protein diet: group K.

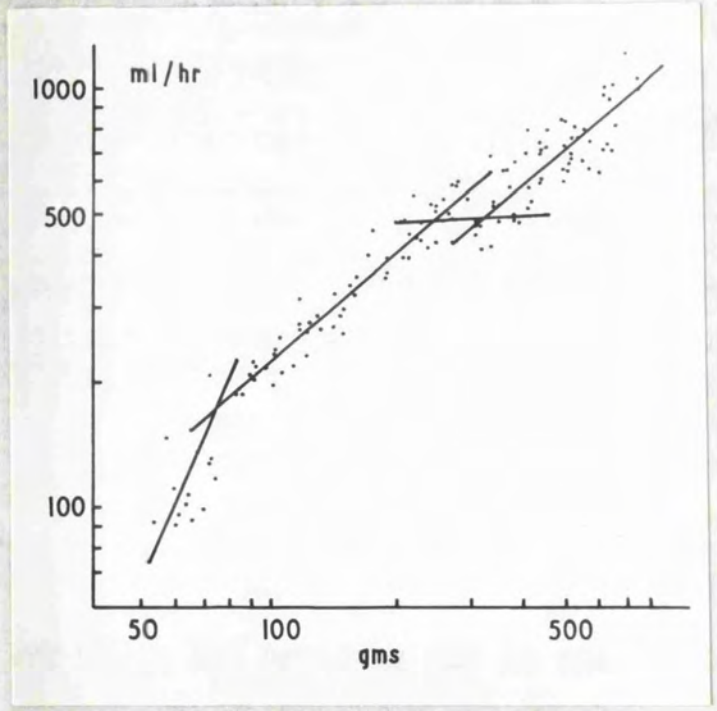



Fig. 49. Oxygen requirements of RIR x LS ♂♂ fed a normal protein diet: group K. Note that the oxygen uptake was more markedly depressed during the third phase than in the group fed the high protein diet (fig. 48).

table 46. Only at 3 and 8 weeks were there significant differences; at 3 weeks the high fat group had the higher rate (P = 0.05) whilst at 8 weeks the reverse was found. There was a trend in both groups for the metabolic rate to fall after the second or third week.

III.6.iii The effect of dietary protein of the diets: Group K

In the previous two experiments the effects of the source of the metabolizable energy upon oxygen uptake were investigated, whilst the protein content was similar in all the diets. Here two diets containing different levels of dietary protein but with similar metabolizable energy and oil contents have been investigated.

The environmental temperatures at which oxygen requirements were measured are given in table 38. Owing to a major technical breakdown at six weeks the measurements had to be virtually concluded but a few data were collected at eight weeks.

The adrenal and thyroid weights of a representative sample from each group were determined at 5 and 8 weeks of age.

Results

The data are given in table XV, and the metabolic pattern of each group is shown in figs. 48 and 49. It will be noted that there was very little difference in the patterns and apart from the third phase, little difference in the actual slope of the regression lines (see table 48 for regression equations). The third metabolic phase, characterized by a levelling off in oxygen uptake in ml/hr when compared with the body weight was observed in both groups but it was notably less marked in the high protein group. Furthermore, this

| Age (weeks) | High protein Food (gm) | Wt. (gm) | Normal protein Food (gm) | Wt. (gm) |
|---|---|---|---|---|
| 0 | | 40.5 | | 40.6 |
| 1 | 58 | 73 | 56 | 71 |
| 2 | 194 | 108 | 170 | 113 |
| 3 | 399 | 193 | 355 | 192 |
| 4 | 766 | 336 | 737 | 316 |
| 5 | 1130 | 469 | 1074 | 449 |
| 6 | 1650 | 652 | 1581 | 602 |
| 7 | 2221 | 874 | 2119 | 859 |
| 8 | 2788 | 1067 | 2624 | 1076 |

Table 49. The effect of protein on the accumulative food intake and growth of the growing fowl (RIR x LS ♂♂): group K.

| Age (weeks) | High protein n | ml/gm/hr±SD | Normal protein n | ml/gm/hr±SD |
|---|---|---|---|---|
| 1 | 10 | 1.85 ± 0.27 | 9 | 1.81 ± 0.51 |
| 2 | 12 | 2.25 ± 0.26 | 12 | 2.26 ± 0.22 |
| 3 | 15 | 2.27 ± 0.18 | 15 | 2.15 ± 0.24 |
| 4 | 15 | 1.49 ± 0.29* | 14 | 1.41 ± 0.39* |
| 5 | 18 | 1.39 ± 0.19 | 17 | 1.48 ± 0.29 |
| 6 | 9 | 1.32 ± 0.25 | 9 | 1.30 ± 0.24 |
| 8 | 3 | 1.15 ± 0.08 | 3 | 1.23 ± 0.14 |

* Significant fall ($P < 0.01$) between 3 and 4 weeks.

Table 50. The effect of dietary protein upon the metabolic rate of the growing fowl (RIR x LS ♂♂).

| Age (weeks) | Group | n | Body weight (gm) | Thyroid weight (mg) | Thyroid weight (mg/100gm) | Adrenal weight (mg) | Adrenal weight (mg/100gm) |
|---|---|---|---|---|---|---|---|
| 5 | High protein | 7 | 448.7±46.9[1] | 36.3±7.7 | 8.1±0.9 | 64.4±7.9 | 14.3±1.6 |
| | Normal protein | 7 | 416.4±24.2 | 28.4±3.0** | 6.8±0.7* | 51.2±4.0**** | 12.3±1.3*** |
| 8 | High protein | 7 | 1067.0±60.8 | 68.4±12.7 | 0.064±0.014 | 101.2±14.5 | 0.093±0.001 |
| | Normal protein | 7 | 1075.9±60.8 | 85.4±18.1 | 0.078±0.014 | 106.1±28.4 | 0.099±0.022 |

[1] SD; * $P < 0.05$; ** $P < 0.02$; *** $P < 0.01$.

Table 51. The effect of dietary protein upon the weights of the thyroid and adrenal glands: group K

phase was observed in the small weight range of 240-320 gm in the normal protein group in contrast to the wider range of 230-520 gm in the high protein group. The weekly metabolic rate was not significantly affected by the protein content of the diet. It is interesting to note that there was a highly significant fall in metabolic rate ($P < 0.01$) between the third and fourth weeks in both groups (table 50).

Although the group fed the high protein diet did show a better growth rate from the third to seventh week, it was at no time significant (see table 49). Birds on the high protein diet also tended to consume rather more food.

It will be seen from table 51 that both thyroid and adrenal weights were significantly affected by the protein content of the diet at 5 weeks of age but not at 8 weeks. Whether measured in milligrammes or mg/100 gm the thyroids and adrenals were significantly larger in the high protein group. That the thyroids and adrenals should be larger is consistent with the higher growth rate, although at no time did the body weight of the high protein group significantly exceed that of the normal protein group, and in fact at the end of the experimental period (8 weeks) the normal protein group were slightly heavier. This was paralleled by the loss of the significant differences in the weights of the thyroids and adrenals.

# PART IV

## Discussion and Summary

| Author | Breed | Full term | P* | H+1hr | H+2hr | H+6hr | H+24hr |
|---|---|---|---|---|---|---|---|
| Giaja & Jovancic (1950) | WL | 18.0 | 33.0 | — | 44.0 | — | 54.0 |
| Romijn & Lokhorst (1956) | ? | 36.1 | 36.8 | 45.1 | — | — | — |
| Visschedijk (1962a) | WL | 26.4 | 41.0 | 33.1 | — | — | — |
| | RIR | 22.2 | 28.5 | 32.6 | — | — | — |
| | WLxRIR | 23.5 | 43.4 | 51.6 | — | — | — |
| Present work | WL | 26.0 | 36.0 | 51.0 | — | — | 49.7 |
| | RIRxLS | 24.5 | 38.0 | 42.0 | 52.5 | 59.4 | 58.1 |

* Measured about 5 hours after pipping; P = pipped; H = hatched.

Table 52. The oxygen consumption of the chick during hatching: a comparison of published results. Figures in ml/hr at STP.

## IV.1 Discussion

### IV.1.i The termination of the embryonic existence

It is necessary to use the single egg as the experimental unit if the metabolic changes which occur during hatching are to be accurately established. Only Giaja & Jovancic (1950) and Visschedijk (1962a) have examined the respiratory metabolism of the chick during this period in any detail, but unfortunately these authors have used small numbers of embryos. Their results, together with data taken from Romijn & Lokhorst (1956) have been brought together in table 52 to facilitate a comparison with the results presented in this thesis. It will be seen that there is general agreement as to the volume of oxygen required by the chick at various stages of development, although the figure published by Romijn & Lokhorst (1956) for the full-term embryo is considerably higher. The reason for this is unknown.

The first rise in the oxygen uptake by the full-term embryo was found to be initiated a few hours before pipping occurred. This would seem to be a result of the onset of pulmonary respiration. Indeed Brody remarked in 1927 that " ... the break in curve at this time might be (due to) a change in the mode of respiration". The oxygen consumption of both the breeds - White Leghorns and RIR x IS - was found to continue to gradually increase for about 6 hours after pipping. During this period (the parafoetal period) the site of gaseous exchange is transferred from the chorio-allantoic membrane to the lungs (Kuo, 1932). Since pulmonary respiration is an

active process, it may be assumed that at least part of this rise in oxygen uptake is a direct result of the increased muscular activity. Further support for this conclusion is offered by the observation that whilst there was a great similarity between the oxygen consumption per square centimetre of the parafoetus and the hatched bird (0.51 ml/cm$^2$/hr and 0.60 ml/cm$^2$/hr) the oxygen uptake of the full-term embryo was quite different (0.37 ml/cm$^2$/hr). The main difference between the parafoetus and the full-term embryo is that the former has functional lungs.

Oxygen uptake was fairly constant from about 6 hours after pipping to about 2 hours before the onset of active hatching. The metabolic patterns of the two breeds were found to be somewhat different during active hatching. Whereas the oxygen requirements of the White Leghorn chick immediately after hatching became constant for at least twenty four hours, the RIR x LS chicks showed a rapid rise in oxygen uptake after hatching so that it was not until the third hour of post-embryonic life that the oxygen requirements rose to the level of the day-old chick (tables 10 and 52). Once the metabolism had become constant, however, the oxygen requirements of the two breeds were very similar.

It is well known that the embryo is poikilothermic whilst the hatched chick is essentially homeothermic (Pembrey et al., 1895; Giaja, 1925; Romijn, 1954b, Romijn & Lokhorst, 1955), but the exact time that the homeothermic response emerges was not known. From the results of the present experiments it appears that the

full-term embryo and the parafoetus are capable of limited, transient thermoregulation but that immediately upon escaping from the shell membranes the chick is able to give sustained responses. Transient homeothermy has not previously been described for the fowl. It was only elicited when the cooling was greater than 3°C for the parafoetus and the full-term embryo. When the cooling of the parafoetus was limited to less than 3°C, the well-known "neutral condition" could be demonstrated. Whether it is the neutral condition or the transient homeothermic response that is elicited from the hatching embryo would therefore appear to depend upon the degree of cooling. Transient homeothermy is certainly a developmental advance upon "chemical shivering" (Romijn & Lokhorst, 1955) and the slow thyroid response (Tixier-Vidal, 1957), both of which were found to be insufficient to prevent the decline in the metabolic rate.

In altricial birds and mammals generally, the ability to regulate heat production is developed over several days, months or even years. Buchanan & Hill (1947, 1949) have suggested that the development of homeothermy is related to the degree of myelination of the hypothalameal tracts. Such a mechanism is most unlikely to be responsible for the emergence of homeothermy in the fowl, although it may well be linked with the perfection of the response which occurs at the end of the first week of post-embryonic life (Romanoff, 1941b; Romijn, 1954b; Romijn & Lokhorst, 1955).

The rapidity with which homeothermy is developed suggests that the mechanism in the fowl is essentially nervous and probably

peripheral, for although at lowered environmental temperatures the blood of the embryo was cooled as it passed through the chorio-allantoic membrane, it failed to stimulate more than a transient response in the hatching embryo. Although the thyroid must be active immediately after hatching in that the "elevated" metabolic rate becomes the normal rate, it does not appear to be involved in the immediate homeothermic response. At least 2 days' continuous stimulation are required to elicit the normal histological changes in the thyroid of the full-term embryo (Tixier-Vidal, 1957) and 10 days in that of the mature fowl (Stahl et al., 1961). The importance of nor-adrenaline in the thermoregulatory responses of the new-born has recently been emphasized by Moore (1960), Moore & Underwood (1960a, b) and Scopes & Tizard (1963). It therefore seems more likely that the adrenal gland is involved in thermoregulation in the newly hatched chick.

The chick, although it develops the homeothermic response extremely rapidly, is not unique in this respect. A similar emergence of the response has been demonstrated for the sheep (Dawes & Mott, 1959), the pig (Mount, 1958, 1959: cf. Holub, Forman & Jeskova, 1957), the dog (Gelineo, 1954; McIntyre & Ederstrom, 1958), the rat (Taylor, 1960) and the duck (Khaskin, 1960).

The average rise in the oxygen consumption between the onset of active hatching and the third hour of post-embryonic life was found to be 20.7 ml/hr. However, when expressed in relation to the surface area, there was a rise in the oxygen requirements of only

0.09 ml/cm$^2$/hr. It may therefore be concluded that this rise in oxygen uptake is the result of the chick attempting to maintain its body temperature at the pre-hatching level.

During the termination of the embryonic existence the oxygen consumption was found to increase by 125-150% confirming the results of several authors (Lusanna, 1906; Romanoff, 1941a; Romijn & Lokhorst, 1951, 1960; Visschedijk, 1962a). According to Bogue (1932) the heart rate increases by only 25% during this same period. His data have been confirmed for the embryo by Romanoff & Sochen (1936) and for the day-old chick by Ringer, Weiss & Sturkie (1957) and Francis (1962). The oxygen carrying capacity of haemoglobin does not increase during the period of hatching but probably falls slightly (Hall, 1934). This is due to there being embryonic and adult forms of haemoglobin (Hall, 1934; Saha, Dutta & Ghosh, 1957; Datta, Ghosh & Guha, 1958; van der Helm & Huisman, 1958), the embryonic haemoglobin having a greater affinity for oxygen. Neither the erythrocyte count nor the haemoglobin content of the blood change markedly at hatching (Romanoff, 1960).

The partial pressure gradient of oxygen between the atmosphere and the venous blood of the full-term embryo is about 70-80 mm of mercury (Romijn, 1950a, 1954a). This is sufficient to allow the complete oxygenation of the blood (Hall, 1934), and therefore it seems likely that the embryo receives sufficient oxygen for its needs. Consequently the discrepancy cannot be explained simply in terms of the removal of an anoxic state. However, there is some indirect

| Age (days) | SA egg $cm^2$ | Diffusion rate $ml/cm^2/hr$* | Vol. of $CO_2$ ml/egg/hr |
|---|---|---|---|
| 20 | 62.4 | 17.53 | 5500 |

*Measured at an environmental temperature of 30°C and a pressure gradient of 100 mm $H_2O$. SA = surface area

Table 53. The volume of carbon dioxide that is able to diffuse across the egg shell under natural conditions. Calculations based on data of Romijn (1954a).

evidence to suggest that the chorio-allantois becomes less efficient as a site for gaseous exchange during the latter stages of incubation. Kuo & Shen (1937) noted that the colour of the blood became progressively darker which they interpreted as indicative of an increase in its carbon dioxide content. Thus since the metabolism is constant, the loss of carbon dioxide from the embryo must be reduced by a decrease in the efficiency of the chorio-allantois for the permeability of the shell is such as not to interfere with the loss of the gas at this time (table 53). The uptake of oxygen might be similarly impaired. Therefore the removal of anoxia might allow some increase in oxygen uptake without a compensatory increase in the heart rate. The significance of this factor, however, is not known.

More oxygen could be made available to the tissues by either increasing the minute volume of blood, or by increasing the difference between the partial pressure of oxygen between the arterial and venous blood, or by a combination of both. There are no data available on the effect of hatching upon the stroke volume of the heart, but it seems unlikely that there is any great increase. Thus the minute volume can only parallel the increase in the heart rate - i.e. 25%. Greater abstraction of oxygen from the blood can be accomplished by two methods, increasing the partial pressure of carbon dioxide in the tissues or lowering the pH of the blood. Again there is no evidence available to support or refute either of these suggestions. The problem, therefore, remains substantially unsolved.

There are two essential phenomena in the termination of the embryonic existence each activated by a different stimulus. The first stage of hatching begins with the establishment of pulmonary respiration and ends when the lungs become the sole site of gaseous exchange. The second stage is concerned with the breakdown and escape from the shell membranes.

The pulmonary stimulus does not appear to be hormonal, or at least thyroidal. l-thyroxine, l-triiodothyronine, 2-thiouracil and thyrotrophic hormone were all without effect upon the time at which pulmonary respiration was initiated. The hypothesis that carbon dioxide is the pulmonary stimulus (Windle & Barcroft, 1937, 1938; Windle & Nelson, 1938; Windle et al., 1938) has been a subject of some controversy. From early experiments indirect, confirmatory evidence was supplied by the observation that many embryos died after pipping when they were maintained for the latter part of incubation in an atmosphere free of carbon dioxide. Prior to pipping development was quite normal.

Waxing the shell of the air space substantially reduces the rates of diffusion of gases over this part of the shell. Several authors have investigated the effects of such a treatment upon the embryo (Byerly & Olsen, 1931; Windle & Barcroft, 1938; Windle et al., 1938; Visschedijk, 1962a). None of these authors has reported that the time of the onset of pulmonary respiration is affected in any way, although Visschedijk (1962b) is of the opinion that respiration might be initiated at an earlier time. In the present experiments

it was found that the onset of breathing was advanced by an average of 5.3 hours when the waxing was carried out at 18½ days' incubation. It was concluded that the increase in the partial pressure of carbon dioxide in the air space had been reflected by a similar rise in the blood, thereby causing that critical partial pressure of the gas necessary to stimulate the respiratory centres being reached at an earlier time. The lowered oxygen tension within the air space was not considered to be of importance in the stimulation, for both Windle & Barcroft (1937) and Visschedijk (1962a) have shown that the embryo is at least twice as sensitive to carbon dioxide as it is to oxygen.

Calculations based on the data of Romijn (1950a, 1954a) show that the permeability of the shell is such that it is in no way involved in the production of the critical partial pressure of carbon dioxide in the blood. The normal partial pressure gradients existing between the egg and the atmosphere are sufficient for all the carbon dioxide produced by the embryo to diffuse away (see table 53). The findings of Noyons & de Hesselle (1939) and Romijn (1948) give support to this conclusion, for they found that the total destruction of the shell of the air space did not prevent the completion of embryonic development. It seems, therefore, that the chorio-allantois must become either less efficient or that its capacity for gaseous diffusion is exceeded as incubation proceeds. The observation that the chorio-allantoic blood vessels constrict when exposed to a high partial pressure of carbon dioxide (Hammond & Zoll, 1937) seems to

support the former conclusion. Furthermore, the gradual atrophy of the vessels in the air space region during the latter phase of incubation can only aggravate this condition.

Waxing the air space shell - approximately 30% of the total shell surface - results in about 13% of the chorio-allantoic membrane being rendered useless for gaseous exchange. Visschedijk (1962a) has shown that within half an hour the new gas equilibrium is set up in the air space: the partial pressure of carbon dioxide rising to 50 mm of mercury, the oxygen tension falling to 46 mm of mercury. Moderate hypercapnia would be expected to develop rapidly under these conditions, resulting in the earlier initiation of pulmonary respiration. However, the advance was found to be one of only 5.3 hours, some 19 hours after waxing had been carried out. The long period between the establishment of the new gaseous equilibrium and the onset of breathing might be due to the tissues being unable to react, or that the changes in the air space were only slightly paralleled by changes in the blood of the embryo, or that another factor, such as the uptake of the amniotic fluid, was preventing the onset of pulmonary respiration.

Since the embryo is capable of respiratory movements as early as the thirteenth day of incubation (Kuo & Shen, 1937; Windle & Barcroft, 1938) the first explanation cannot be valid. Either of the latter two explanations seem more likely, and may even be complementary to one another. At the time that pulmonary respiration is initiated about 30% of the oxygen required by the embryo is obtained via the

air space whilst about 27% of the carbon dioxide produced is lost by the same route (Visschedijk, 1962a). Visschedijk has also shown that approximately 66% of the carbon dioxide previously lost through the air space is lost through the allantois when the air space shell is covered with paraffin wax. This demonstrates that the gaseous changes in the waxed air space of the embryo are only partially reflected in the blood.

Since the overall partial pressure gradients between the embryo and the atmosphere are not affected by perforating the air space shell, the loss of carbon dioxide from this part of the chorio-allantois ought not to be seriously affected. The onset of pulmonary respiration was only delayed by 1.4 hours by such treatments and therefore confirms this conclusion.

The uptake of the amniotic fluid is an active process (Wislocki, 1921; Vrbitch, 1924; Taylor & Saenz, 1949), the control of which is unknown. Pulmonary respiration certainly cannot begin until the major portion of the fluid has been absorbed, as Byerly & Olsen (1931) and Kuo & Shen (1937) have pointed out. Kuo & Shen (1937) showed that breathing could be depressed by injecting isotonic saline into the amniotic cavity. Thus the removal of the amniotic fluid may be a prerequisite for the establishment of pulmonary respiration. Once this has been substantially completed the gaseous stimulus, carbon dioxide, is able to act.

It may be concluded that the stimulus for the onset of pulmonary respiration is carbon dioxide, and is mediated through the respiratory

centres in the medulla oblongata. The removal of the amniotic fluid is probably a prerequisite for the onset of breathing but the control of the removal of this fluid is unknown. Until it has been removed the carbon dioxide stimulus is possibly inhibited.

Vascularization of the pulmonary system resulting in the falling partial pressure of oxygen and the rising partial pressure of carbon dioxide within the air space of the normal embryo leads to increased hypercapnia and possibly a lowered oxygen uptake (fig. 8, page 49). This appears to initiate spasms of the cervical muscles. Finally these spasms cause the fracture of the shell at one point, i.e. the shell is pipped. There will then follow a rapid loss of carbon dioxide from the air space and from the blood of the parafoetus and also an increase in the partial pressure of the oxygen in the air space, with the subsequent loss of anoxia. With the removal of the noxious stimuli the chick enters the quiescent period.

It is not surprising, therefore, that waxing the air space shell led to an advance of pipping by 14 hours whilst pipping was delayed by perforating or aerating the air space. Furthermore these results demonstrate the importance of the air space in realizing the conditions necessary for pipping. However, it was found that in some aerated embryos the phenomenon was entirely abolished, and together with the evidence of Noyons & de Hesselle (1939) and Romijn (1948), that the air space is not necessary for the completion of embryonic development, shows that pipping is also not essential. Rather it is a chance, non-essential phenomenon in the hatching

process.

During the quiescent period the vessels of the chorio-allantois are constricted gradually causing the lungs to become the sole site of gaseous exchange. During the parafoetal period at least, it appears that the carbon dioxide concentration of the environment should not fall below a minimum percentage. High mortality (35%) occurred after pipping when the eggs were kept in the respirometers for many hours. The carbon dioxide concentration in these was almost zero. It seems likely that continual stimulation of the respiratory centres is necessary to ensure the ventilation of the lungs. Under practical conditions the lower limit of carbon dioxide concentration is probably about 0.5%. Once the lungs have taken over completely, the dependence upon atmospheric carbon dioxide is less apparent, and may even be lost, since atmospheres containing no carbon dioxide do not adversely affect the respiration of the hatched chick.

The quiescent period is terminated with the onset of active hatching. It was observed that the metabolic rate of the chick began to rise before any increase in its physical activity could be detected. The hypothesis was therefore advanced that an increase in the rate of thyroid hormone secretion was responsible for the initiation of active hatching. Experiments to test this hypothesis have been fairly conclusive. Although other workers have investigated the effects of thyroxine on the embryo, the drug has usually been given at an early stage of incubation and has consequently

| | n | I' ug/100ml |
|---|---|---|
| Full-term embryo | 2 | 6.3 |
| Parafoetus (5hr)* | 2 | 10.9 |
| Hatched chick (1hr)* | 2 | 13.2 |

* Age.

Table 54. The protein bound iodine concentrations in the serum of the hatching chick embryo.

affected the whole course of embryonic development. Here the drugs were administered at as late a stage as possible. Both thyroxine and triiodothyronine advanced hatching, and it is noteworthy that they had a very similar potency in the chick embryo. This is in accordance with the published data for the hatched chicken (Shellabarger, 1955; Newcomer, 1957; Tata & Shellabarger, 1959) and forms an interesting comparison with mammalian response (Gross & Pitt-Rivers, 1952, 1953). Large doses of thyrotrophic hormone (1.0 i.u.) had to be employed to elicit significant responses. The large molecular size of this substance might be a factor in the smallness of the response (the drug was merely injected into the air space and therefore uptake by the blood might be affected) and the lack of large histological changes in the thyroid (fig. 22) suggests that uptake was impaired.

In pilot experiments on the measurement of the protein bound iodine (PBI) of the blood serum, it was found that there was approximately a 100% increase in the concentration of PBI between the commencement and termination of the hatching process (table 54). This indicates that the thyroid is very active at hatching confirming the work of Sun (1932) and, together with the other results, suggests that active hatching is controlled by the thyroid gland.

Why an increase in the rate of secretion of the thyroid hormones should cause the chick to escape from its shell is not known. There is limited evidence to suggest that it may be a behavioural response. It has been demonstrated that large doses of thyroxine cause increases in the metabolic rate, body temperature and physical

activity of the euthyroidal chicken (Martin, 1929). Assuming that the chick at the end of the parafoetal period is the euthyroidal animal, then similar responses might be expected if the amount of thyroid hormones in the blood were to double within a short time. Such an increase in thyroid hormones does occur, and the metabolic rate and physical activity increase markedly. The hypothesis is therefore advanced that the increased rate of thyroid hormone secretion leads to a behavioural response by the chick, which is manifested by the breakdown of the shell and the escape from it.

IV.1.ii The hatched bird

Studies on the gaseous metabolism of the hatched bird have generally been carried out upon the starving animal. In many instances, however, the supposition that the basal metabolic rate was measured was incorrect, since it has been shown that the period of starvation employed was only sufficient to bring the fowl to a post-absorptive state (Mitchell & Haines, 1927b; Dukes, 1937). Investigations on the normally fed bird are few and the work in this thesis has therefore been confined to this neglected field. In such experiments standardization and control of the experimental conditions are of great importance. Thus the temperatures at which the determinations of oxygen consumption were carried out were controlled to within $\pm$ 0.1°C; a day length of 14 hours was established; food and water were available *ad libitum* and the characteristics of the diets were known.

One of the most important factors affecting metabolism is the environmental temperature. Although there is general agreement about the broad limits of the temperature range at which the metabolic rate of the adult fowl is lowest, the detailed movements of these zones of thermal neutrality, particularly during the first month after hatching, were not known. In the study on the effects of temperature during the first month it became clear that the results did not agree with published work. The only factor that appeared to be different was that the birds were fully fed and therefore it seemed likely that the nutritional status of the fowl was affecting

| | Age (days) 1 | 9 | 18 | Ref. |
|---|---|---|---|---|
| Rearing temperature | 35 | 31 | 27 | 1-3 |
| Zone of thermal neutrality | 35 | 34-35 | 31-32 | 4 |
| Zone of thermal neutrality | 35 | 35 | 35 | 5 |

1. Barott & Pringle (1947). 2. Barott & Pringle (1949). 3. Barott & Pringle (1950). 4. Present work. 5. Barott & Pringle (1946).

Table 55. A comparison of the zones of thermal neutrality and the optimum rearing temperatures as determined by Barott and Pringle and the zones of thermal neutrality as found in the present work.

the extent of the zone.

Barott & Pringle (1947, 1949, 1950) determined the environmental temperatures at which birds could be reared most successfully. These studies involved fully fed birds and therefore it may be expected that the recommended environmental temperatures should show a better correlation with the zones of thermal neutrality as determined here. Reference to table 55 shows that this is so. The results obtained by Blaxter (1962) with sheep add support to this view. He found that increased food intake led to a movement of the zone to lower temperatures, although there did not appear to be any effect on the actual range of the zone.

Oxygen consumption during the first week was found to increase at a greater rate than the body weight ($b > 1.0$), confirming the findings of Beattie & Freeman (1962). Comparison of groups C and D, and F, G and H shows that the diet had no effect on the oxygen requirements or on the metabolic rate during this time. Where large groups of chicks were used, it was found that the body temperature and the metabolic rate increased in parallel during the 6 or 7 days after hatching, and although the body temperature then became relatively constant, the metabolic rate often continued to rise for a few more days.

The increase in the body temperature appears to be a result of the progressive replacement of the yolk by actively metabolising tissue, resulting in an increase in the active mass of the bird without a corresponding increase in the surface area. Indeed it was found

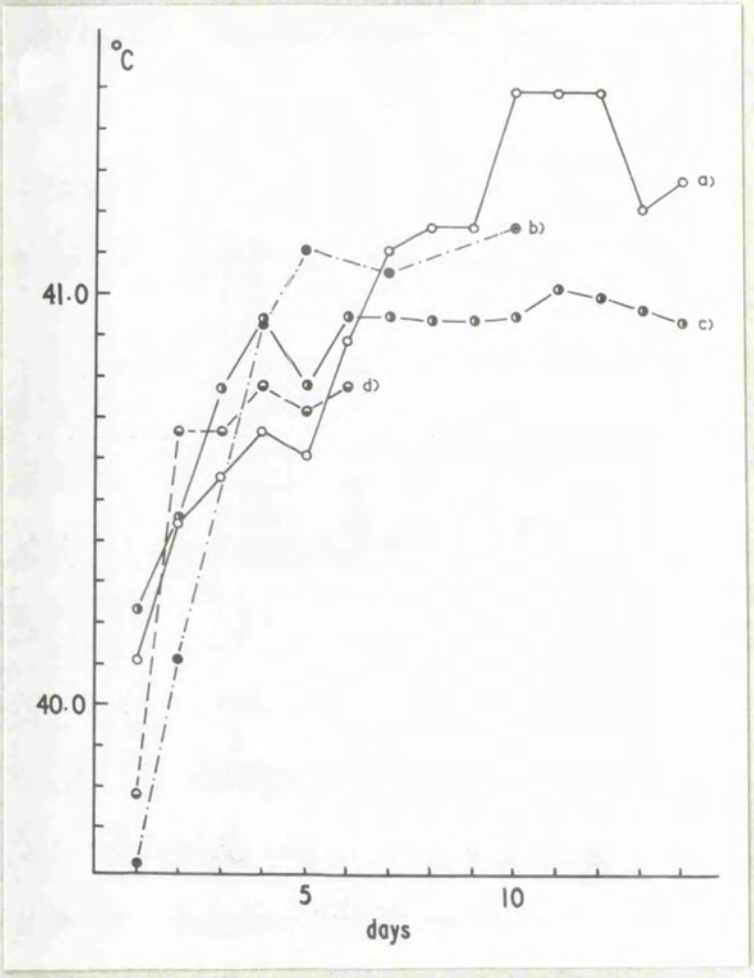



Fig. 50. The body temperature curve of the chick during the first fortnight of post-embryonic life: a comparison with published results.

a) King (1956); b) Lamoreux & Hutt (1939); c) present work; d) Hutt & Crawford (1960).

that the body weight and the surface area during this time increased proportionately. Thus the suggestion of Kendeigh & Baldwin (1928), Baldwin & Kendeigh (1932) and Randall (1943) that body weight *per se* increases at a greater rate than surface area and is therefore responsible for the increase in body temperature, is not strictly correct. Since the metabolic rate continued to rise to the eighth day at least and was not abolished by lecithectomy, it would appear to be real.

That the body temperature should become constant on about the sixth day after hatching is in agreement with the findings of Lamoreux & Hutt (1939) and Hutt & Crawford (1960), although at variance with those of King (1956). The values for the body temperature, however, are of the same order as those found by these authors (see fig. 50).

The significant fall in body temperature and the fall in metabolic rate on the fifth day after hatching coincide with the atrophy of the yolk sac, and the two events are probably related. Although this embryonic relic supplies the chick with food material, it certainly does not appear to be essential to the newly hatched fowl, since its removal did not affect oxygen uptake, the metabolic pattern or the growth rate.

Examination of fig. 51 shows that the "metabolic status" of a strain is not necessarily determined at hatching, but more usually at the end of the first metabolic phase. Thus although strain C had a greater metabolic rate than strain A at hatching, their metabolic rates became almost identical at the end of the first phase.

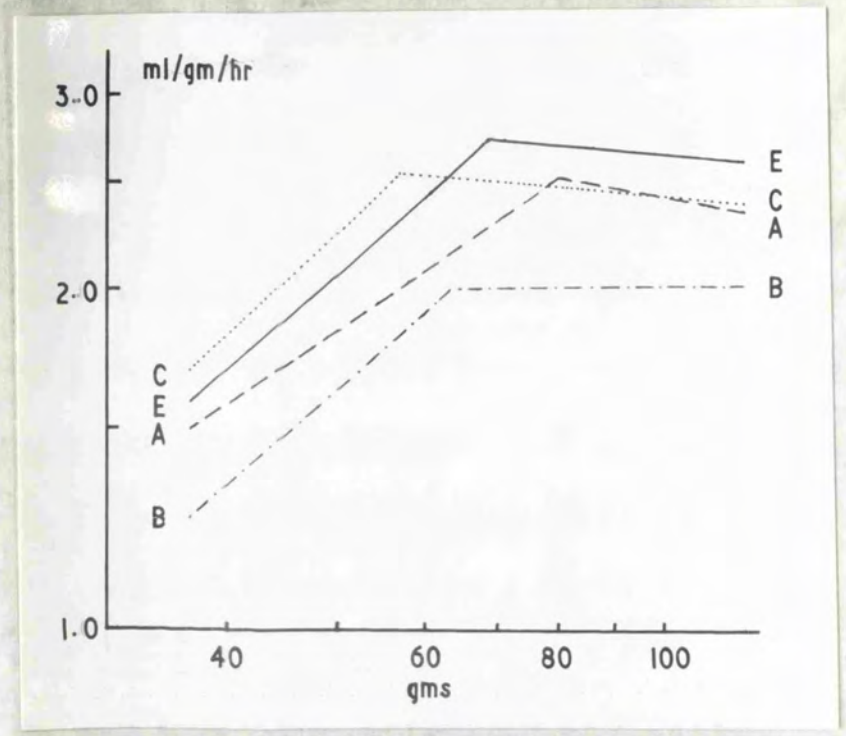



Fig. 51. A comparison of the metabolic rates of groups A-C during the first fortnight. A = RIR x LS; B = a broiler strain; C = a laying strain; E = group B broilers of Beattie & Freeman (1962).

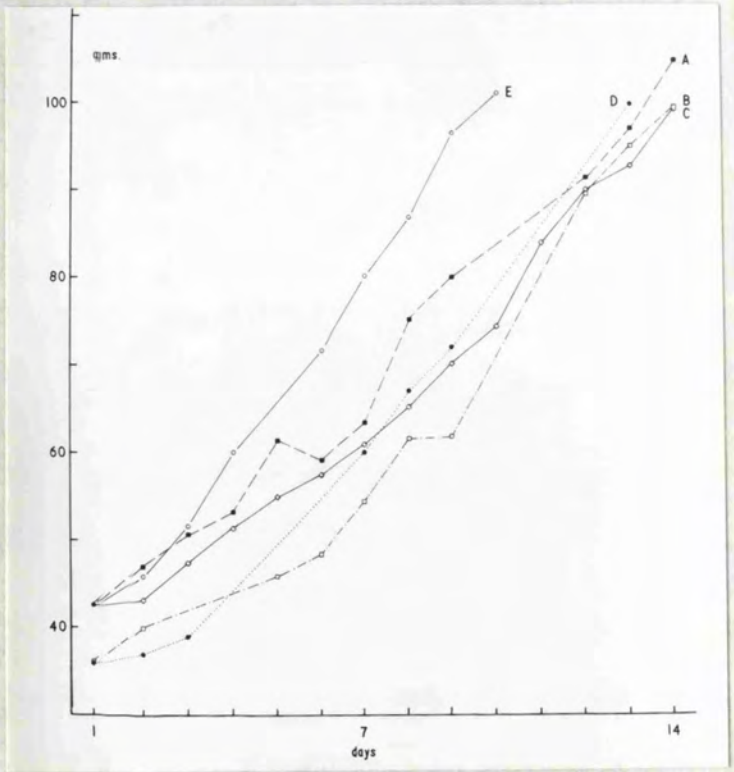



Fig. 52. Growth rates of 3 strains together with a broiler strain examined by Beattie & Freeman (1962). Symbols as above.

The rate of the increase in metabolic rate is perhaps not so important in determining the metabolic status of a strain but rather the time at which the second phase is initiated.

There is a definite tendency for the groups with the highest metabolic rates to have the highest growth rates (e.g. groups A and C, as determined at the beginning of the second metabolic phase). Group B with the lowest metabolic rate had the lowest growth rate, whilst group B of Beattie & Freeman (1962) had the highest rates (figs. 51 and 52). Undoubtedly the metabolic rate is closely correlated with the thyroid secretion rate. Mixner & Upp (1947), Glazener, Shaffner & Jull (1949), Smyth & Fox (1951) and Biellier & Turner (1957) have shown that the thyroid secretion rate and the growth rate of chicks are positively correlated. Hoffmann (1950) and Smyth & Fox (1951) found the same for turkeys and ducks. Thus it may be possible to predict growth rates from the metabolic rates measured during the first fortnight after hatching. This might be of use in breeding for higher growth rates, since observations could be completed in a relatively short time.

The second metabolic phase, which is characterized by a direct relationship between the rate of increase in the oxygen uptake and growth ($b < 1.0$), is common to all strains and does not appear to be influenced by the diet. That the regression coefficient is generally slightly less than 1.0 during this period suggests that the feathers are becoming more efficient as an insulating material.

Subsequent oxygen uptake was influenced by the diet and must be

discussed in relation to this factor therefore. That there was no such influence prior to 1 month of age suggests that the embryonic nutrient, yolk, continues to exert an effect upon metabolism long after its exhaustion. Such a mechanism appears to exist at a slightly later age. The metabolic pattern of group H was essentially the same as that of group F and the absolute oxygen requirements assumed a more intermediate position between those of groups F and G although the broiler diet was fed for only the first two weeks after hatching.

Only the protein content of the diet was found to have any significant difference upon the metabolic pattern. In all groups examined there was a characteristic levelling out in the rate of oxygen uptake giving rise to the third metabolic phase. Only where a high protein diet had been fed was there any indication that this plateau in oxygen consumption would be abolished. However, the results of the high protein high energy diet (group K) do not confirm those obtained for a similar diet fed to group F. It may be due to the breed difference for the former was a RIR x LS cross and the latter a commercial laying strain.

It was not until the eighth week that the metabolisable content of the diet had any significant effect on the oxygen requirements. This is in contrast to the results of Singh & Shaffner (1950) and Mellen et al. (1954), but in view of the criticisms of March & Biely (1957) on the probable inadequacy of the low energy diets used in these studies further comparisons are of little value. Fat and

protein also had little effect on the weekly metabolic rate, although it was noticeable that the rate was more variable for birds fed the high fat diet than for any of the other groups.

Brody et al.(1932) found that the protein content of a diet affected oxygen uptake, but again the lower levels of protein were quite inadequate for the growing chick. Their highest level of protein was 18.9% as compared with 21.7% in the normal diet used in these experiments. That dietary fat should have little effect on oxygen uptake is in agreement with the results of March & Biely (1957).

Protein was the only dietary factor that was found to have a really significant effect upon the weights of the thyroid and adrenal glands. This was in contrast to the findings of March & Biely (1957) and Treat et al. (1960) who reported that fat depressed the weights of these glands. The significantly heavier glands of the birds fed the high protein diet were consistent with the better growth rate of these birds at 5 weeks of age, and may have been simply a result of this growth rate since all the differences were lost by the eighth week, when both groups had a similar body weight.

The growth rate was unaffected by the diets fed to groups I, J and K. The high protein diet led to an improved growth rate between the fourth and seventh week only. In contrast group F, fed the broiler diet, had a much better growth rate than the birds fed the standard diet (group G) whilst group H again had an intermediate growth rate (see fig. 43).

The metabolic patterns exhibited by the birds of group G

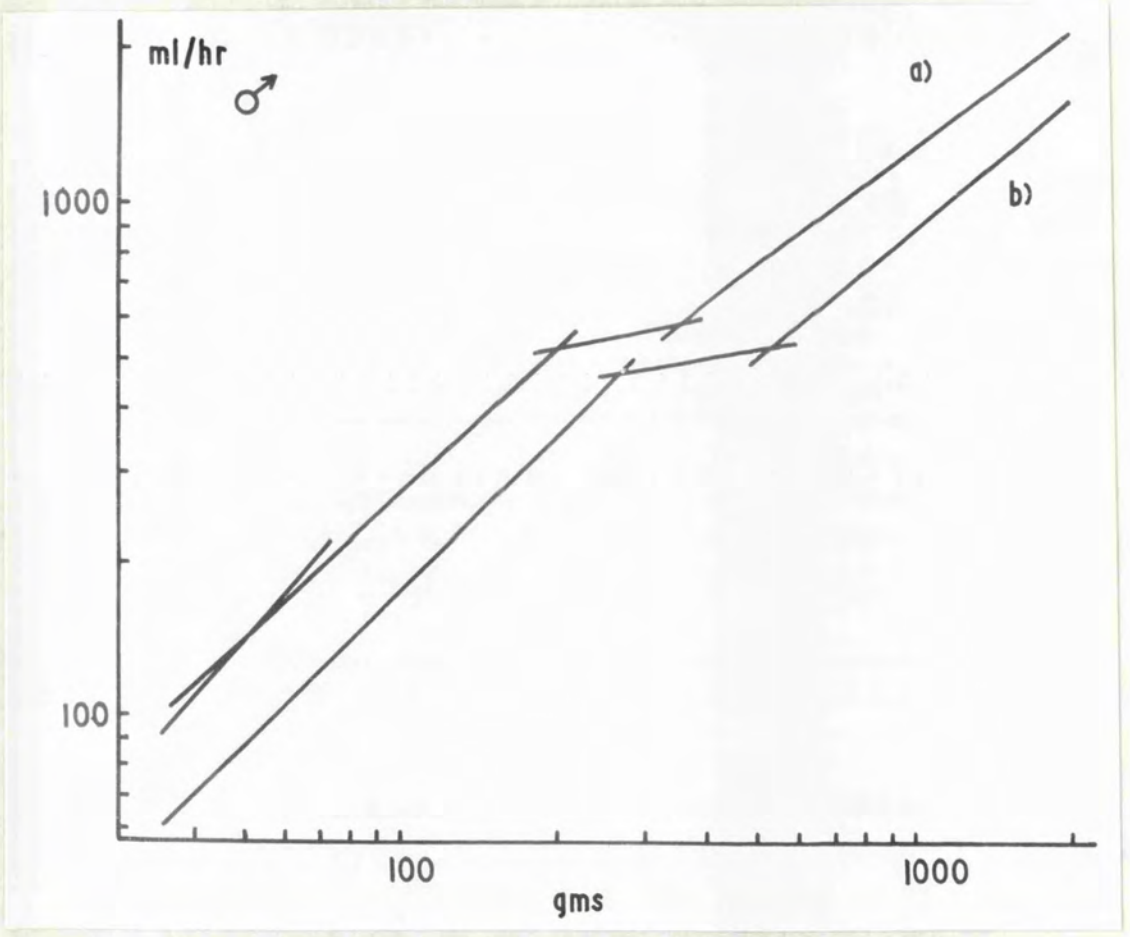



Fig. 53. A comparison of the metabolic pattern of a laying strain (group G males) with that of RIR males (Kibler & Brody, 1944). Note the great similarity between the two curves; the difference in the absolute oxygen requirements may be due to the difference in breed.

a = laying strain; b = RIR.

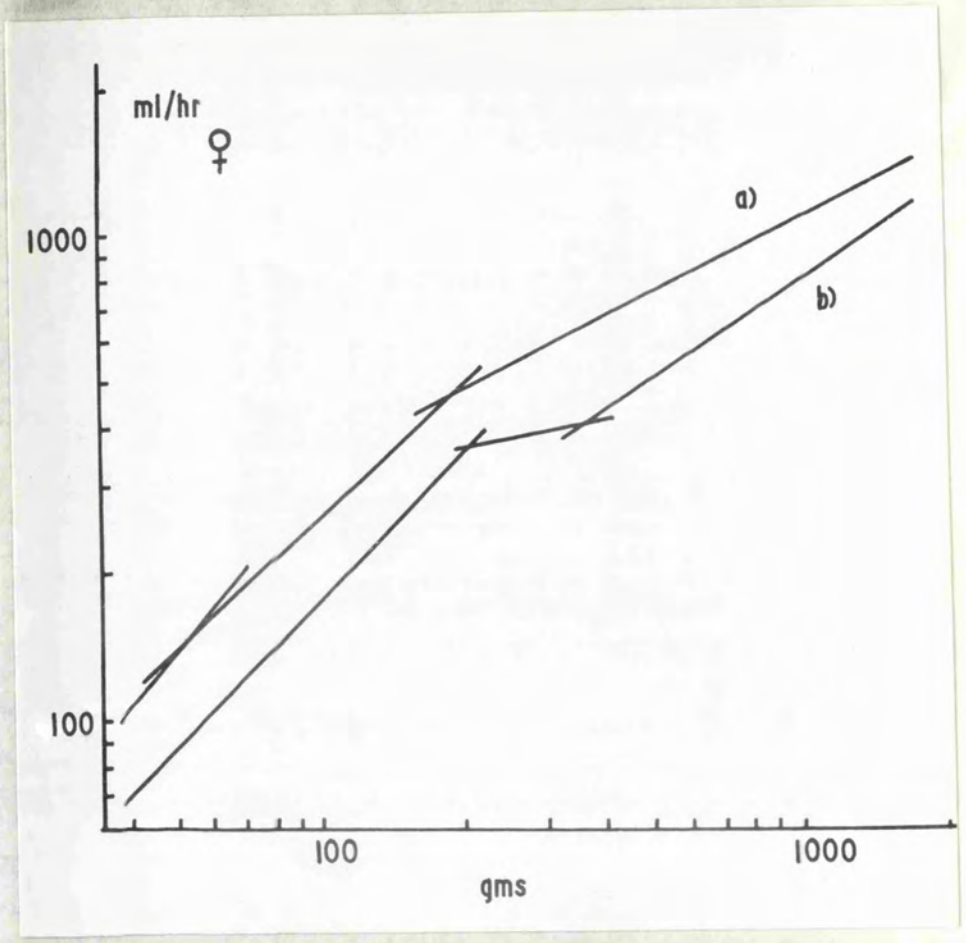



Fig. 54. A comparison of the metabolic patterns of laying strain females and RIR females. The data for the latter breed are taken from Kibler & Brody (1944). The laying strain was fed the standard ration as were the males (fig. 53). a = laying strain; b = RIR.

(males), I, J and K (normal protein) are essentially the same as that obtained by Kibler & Brody (1944). Probably the diets used in those two investigations had similar levels of metabolisable energy, protein and fat, whereas birds fed the commercial broiler diet with its substantially different composition, gave an almost "atypical" pattern. Broiler diets are the product of the last decade or so and were formulated specifically for strains of fowl which had extremely high growth rates. They are characterised by elevated protein and metabolizable energy contents. The similarity of the results for the males of group G (fed the standard ration) to those of Kibler & Brody (1944) is illustrated in fig. 53. However, the pattern for the females of group G was quite different after the second phase and shows that the diet may affect the sexes differently (fig. 54). Kennelly & Maynard (1953) and Mellen et al. (1954) have also noted such differences.

Kibler & Brody (1944) have suggested that there is no sexual difference in oxygen requirements. However, a higher metabolic rate was characteristic of males above a body weight of about 200 gm thus normally preceding any detectable weight differences. The higher metabolic rate of the males was evident both on a weight basis (table 36) and on an age basis (fig. 55) and must occur if the growth rate is to be greater than that of the female. If no sexual divergence occurred then the male would have to possess more efficient oxidative processes.

It has already been noted that the diet does not appear to

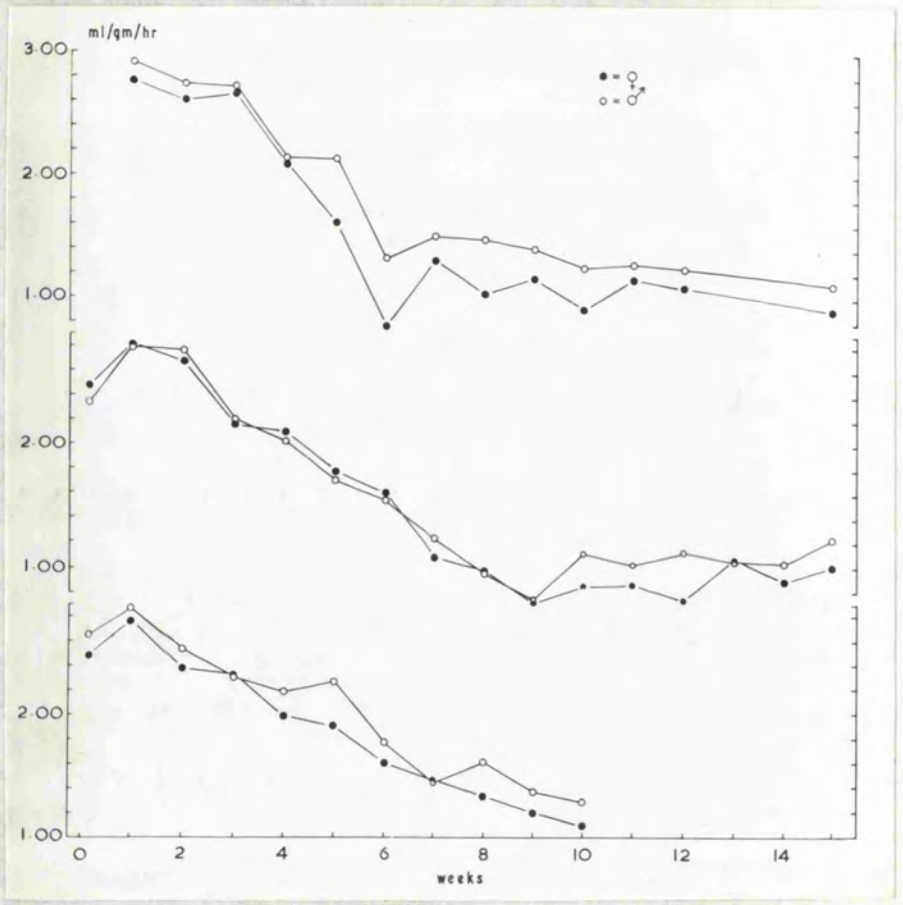



Fig. 55. The influence of age upon the metabolic rates of males and females of the laying strain. (a) Birds fed on the broiler diet; (b) Birds fed on the standard diet; (c) Birds fed the broiler diet for 2 weeks and then the standard diet.

affect either the oxygen requirements or the metabolic pattern until the fowl is about 1 month old. It is interesting to note that there is one significant change in the metabolic rate that appears to be fairly independent of diet but related to the age of the bird. A very significant fall occurred between the third and fourth weeks in groups F, I, J and K. This is paralleled by a fall in the concentrations of α-ketoglutaric acid and pyruvic acid in the blood at about this time (Chubb & Freeman, 1963), falls in the concentrations of blood sugar, creatinine and blood amino-acids, an increase in the total proteins in the serum (though no change in the ratio of albumins to globulins), a slight change in the levels of chloresterol and uric acid in the blood (Tournut, Lacaze & Montlaur-Ferradou, 1963), a fall in adrenal and blood ascorbic acid concentrations (Freeman & Chubb, unpublished) and a fall in adrenal gland weight (mg/100 gm body weight)(Breneman, 1941). These changes have led Tournut et al. (1963) to suggest that there is "une crise physiologique" at 1 month of age. There must indeed be a major, fundamental physiological change in the fowl at this time. Tournut et al. (1963) also suggest that there is a similar change at 8 weeks of age.

The causes of both changes are as yet unknown. The thyroid does not appear to be involved for Schultze & Turner (1944; 1945) found that the thyroid secretion rate was constant between 1 and 12 weeks after hatching. Tournut et al. (1963) have compared the change at 4 weeks of age with weaning in mammals. It has been shown that the diet of the fowl has little or no effect on the metabolism during

the first fortnight at least. Furthermore, it has also been suggested that the yolk continues to exert an effect on the metabolism for some time after its absorption and utilization, possibly in the same way that the broiler diet appears to have influenced the metabolic pattern of group H and therefore may be analagous to weaning. Nutritional disorders do not normally become apparent until 4 weeks of age. It is therefore quite possible that a nutritional factor is involved in this metabolic change. Other factors could be the onset of the secretion of gonadotrophic or corticosteroid hormones.

Further work, both of a biochemical and physiological nature, on the month old bird might be very rewarding.

## IV.2 Summary

1. The oxygen consumption of the hatching chick embryo has been determined. Two distinct rises were differentiated: the first appears to be directed, at least in part, to the provision of energy for the ventilation of the lungs and the other partly directed towards the initiation of active hatching and partly to maintaining the body temperature of the chick at its pre-hatching level.

2. Transient homeothermy was detected in the full-term and hatching embryo, but sustained responses to environmental temperature changes did not appear until the moment of the escape from the shell membranes.

3. Evidence was presented which showed that pulmonary respiration is initiated by a high partial pressure of carbon dioxide within the blood, although an unknown mechanism for the active uptake of the amniotic fluid pervading the respiratory tract is probably also involved.

4. It is suggested that it is not the air space but the chorio-allantois that is involved in the realisation of the critical partial pressure of carbon dioxide necessary to stimulate breathing. However, the air space is concerned in providing the necessary gaseous mixture to stimulate pipping.

5. Pipping is a non-essential phenomenon in the hatching sequence.

6. Active hatching is probably mediated through the thyroid gland by an increase in the secretion of the thyroid hormones. It

is further suggested that the escape from the shell is a behavioural response to an increased metabolic rate.

7. The body temperature of the hatched chick rises for about six days and is probably a result of the progressive replacement of the non-metabolizing yolk with active metabolizing tissues, thereby causing a greater rate of increase in the active mass as compared with the surface area of the chick.

8. The metabolic rate rises for about eight days after hatching; part of the rise may not be real, i.e. there may be no change at the cellular level, but that part from 6 to 8 days does appear to be a real increase in metabolic rate since it is not abolished by lecithectomy.

9. Both absolute oxygen requirements and the metabolic pattern are affected by the diet but not below a body weight of about 200 gm (3 weeks of age). Birds fed normal levels of protein, high or normal levels of fat or carbohydrate exhibit a period of fairly constant oxygen uptake over a range of 150-200 gm. High levels of protein tended to abolish this severe depression in metabolic rate.

10. A highly significant fall in the metabolic rate at about one month of age was consistently noted. This appeared to be largely independent of the diet and was accompanied by several other physiological changes within the bird. This "physiological crisis" is not understood.

## PART V

### Date

## V.1 Data on the oxygen consumption of the hatching embryo

### Table I. Oxygen consumption of three White Leghorn embryos.

The double line (===) gives the time of hatching.

| Hrs. before and after pipping | 12 ml/hr | 12 ml/gm/hr | 13 ml/hr | 13 ml/gm/hr | 14 ml/hr | 14 ml/gm/hr |
|---|---|---|---|---|---|---|
| P - 10 | — | — | 26.8 | 0.69 | — | — |
| - 9 | — | — | 27.3 | 0.70 | — | — |
| - 8 | 24.6 | 0.51 | — | — | 24.5 | 0.56 |
| - 7 | 26.3 | 0.53 | — | — | 24.9 | 0.57 |
| - 6 | 25.6 | 0.53 | 28.0 | 0.72 | 25.9 | 0.59 |
| - 5 | 26.1 | 0.54 | 28.1 | 0.72 | 24.1 | 0.55 |
| - 4 | 25.9 | 0.54 | 28.0 | 0.72 | 24.1 | 0.55 |
| - 3 | — | — | 30.5 | 0.78 | 23.8 | 0.54 |
| - 2 | — | — | 26.7 | 0.69 | — | — |
| - 1 | — | — | 25.6 | 0.66 | — | — |
| P + 1 | 31.7 | 0.66 | 27.6 | 0.71 | 29.4 | 0.67 |
| + 2 | 29.9 | 0.62 | — | — | — | — |
| + 3 | 29.8 | 0.62 | — | — | 32.0 | 0.73 |
| + 4 | 32.4 | 0.68 | 33.6 | 0.86 | 31.7 | 0.73 |
| + 5 | 34.5 | 0.72 | — | — | 33.0 | 0.76 |
| + 6 | 35.0 | 0.73 | 35.3 | 0.91 | 33.9 | 0.78 |
| + 7 | 35.9 | 0.75 | 33.6 | 0.92 | 34.9 | 0.80 |
| + 9 | — | — | 35.5 | 0.91 | — | — |
| + 10 | 37.3 | 0.78 | 41.8 | 1.07 | 37.6 | 0.86 |
| + 11 | — | — | 45.5 | 1.17 | — | — |
| + 12 | 34.1 | 0.71 | — | — | 38.3 | 0.88 |
| + 13 | 40.4 | 0.84 | — | — | 36.8 | 0.84 |
| + 15 | 43.4 | 0.90 | — | — | 39.1 | 0.90 |
| + 16 | 43.5 | 0.91 | — | — | ========== | |
| + 17 | 44.8 | 0.93 | — | — | 50.8 | 1.16 |
| | ========== | | | | | |

Table I cont'd.

| Hrs. before and after pipping | 12 ml/hr | 12 ml/gm/hr | 13 ml/hr | 13 ml/gm/hr | 14 ml/hr | 14 ml/gm/hr |
|---|---|---|---|---|---|---|
| P + 18 | 56.8 | 1.18 | — | — | — | — |
| + 33 | — | — | 44.0 | 1.24 | — | — |
| + 34 | — | — | 41.2 | 1.16 | — | — |
| + 36 | 54.4 | 1.25 | — | — | 50.4 | 1.26 |
| + 37 | 52.8 | 1.21 | — | — | 55.7 | 1.39 |
| + 56 | — | — | 52.0 | 1.65 | — | — |
| + 57 | — | — | 48.3 | 1.53 | 48.6 | 1.34 |
| + 60 | 55.1 | 1.41 | — | — | 59.3 | 1.64 |
| + 61 | 62.6 | 1.60 | — | — | — | — |

Table II. Oxygen consumption of a typical RIR x LS embryo

The double line (===) gives the time of hatching.

| Hrs. before and after pipping | ml/hr | ml/gm/hr |
|---|---|---|
| P - 2 | 21.3 | 0.49 |
| - 1 | 20.8 | 0.47 |
| + 2 | 28.4 | 0.65 |
| + 4 | 33.7 | 0.77 |
| + 12 | 36.7 | 0.84 |
| + 13 | 35.9 | 0.82 |
| + 14 | 35.1 | 0.80 |
| + 15 | 40.9 | 0.93 |
| + 16 | 39.3 | 0.90 |
| + 17 | 39.7 | 0.91 |
| + 18 | 33.3 | 0.76 |
| + 22 | 64.5 | 1.47 |
| + 23 | 59.1 | 1.35 |
| + 52 | 56.0 | 1.40 |

Table III. Oxygen consumption during the first six hours after hatching in RIR x LS chicks. Results of four chicks.

H = Hatched

| Age (hrs) | 23T/20/2/62 ml/hr | ml/gm/hr | 15W/28/2/62 ml/hr | ml/gm/hr |
|---|---|---|---|---|
| H + 0.5 | — | — | 32.1 | 0.68 |
| + 1.0 | — | — | 45.1 | 0.96 |
| + 1.5 | 40.7 | 0.93 | 50.7 | 1.08 |
| + 2.0 | 49.1 | 1.12 | 52.5 | 1.12 |
| + 2.5 | 52.2 | 1.19 | 56.2 | 1.20 |
| + 3.0 | 54.8 | 1.24 | 52.4 | 1.12 |
| + 3.5 | 54.8 | 1.24 | — | — |
| + 4.0 | 57.9 | 1.32 | — | — |
| + 4.5 | 57.6 | 1.31 | — | — |
| + 5.0 | 53.6 | 1.22 | — | — |
| + 5.5 | 57.9 | 1.32 | — | — |
| + 6.0 | 57.6 | 1.31 | — | — |
| | 14/3/62/1 | | 23T/20/3/62 | |
| H + 0.3 | 34.5 | 0.74 | 34.2 | 0.86 |
| + 0.6 | 43.8 | 0.94 | 39.5 | 1.00 |
| + 1.0 | 50.4 | 1.07 | 37.6 | 0.95 |
| + 1.3 | — | — | 48.7 | 1.23 |
| + 1.6 | — | — | 52.2 | 1.32 |
| + 2.0 | 48.9 | 1.05 | 46.9 | 1.18 |
| + 2.3 | 51.5 | 1.11 | 49.3 | 1.25 |
| + 2.6 | 54.4 | 1.17 | 47.9 | 1.21 |
| + 3.0 | 60.2 | 1.29 | 44.2 | 1.12 |
| + 3.3 | 57.4 | 1.23 | 43.2 | 1.09 |
| + 3.6 | 61.0 | 1.31 | 43.7 | 1.10 |
| + 4.0 | — | — | 46.6 | 1.18 |
| + 4.3 | — | — | 46.1 | 1.16 |
| + 4.6 | — | — | 46.9 | 1.18 |

## V.2 Data on oxygen consumption of the hatched bird

Table IV. The oxygen consumption of the White Leghorn chick: standard diet.

| Wt.(gm) | ml/hr | Wt.(gm) | ml/hr |
|---|---|---|---|
| 32.4 | 56.1 | 41.1 | 89.0 |
| 33.7 | 53.9 | 41.9 | 65.3 |
| 33.7 | 45.1 | 49.2 | 114.0 |
| 36.0 | 58.9 | 52.7 | 148.0 |
| 36.2 | 46.6 | 53.4 | 146.0 |
| 36.2 | 60.9 | 54.0 | 156.1 |
| 38.0 | 51.7 | 54.4 | 174.5 |
| 38.1 | 88.2 | 55 | 113.6 |
| 38.1 | 56.0 | 55 | 134.9 |
| 38.2 | 73.4 | 56 | 108.5 |
| 38.8 | 64.8 | 57 | 139.0 |
| 39.1 | 85.0 | 58 | 188.6 |
| 39.2 | 58.1 | 58 | 156.6 |
| 39.4 | 56.9 | 58 | 174.3 |
| 39.4 | 89.0 | | |

**Table V.** Oxygen requirements of the RIR x LS strain, standard diet; group A.

| Wt. | ml/hr | Wt. | ml/hr |
|---|---|---|---|
| 39 | 61.6 | 60 | 114.6 |
| 39 | 68.6 | 64 | 106.8 |
| 44 | 89.0 | 64 | 139.1 |
| 45 | 84.8 | 66 | 112.9 |
| 46 | 68.3 | 66 | 126.0 |
| 47 | 87.2 | 66 | 148.1 |
| 47 | 89.7 | 70 | 146.3 |
| 48 | 87.6 | 71 | 134.1 |
| 49 | 72.1 | 71 | 175.2 |
| 50 | 92.5 | 74 | 192.5 |
| 50 | 110.2 | 74 | 154.5 |
| 50 | 116.6 | 75 | 162.3 |
| 51 | 94.3 | 79 | 200.0 |
| 51 | 114.9 | 80 | 178.6 |
| 53 | 96.0 | 83 | 216.7 |
| 54 | 106.0 | 88 | 229.7 |
| 54 | 125.9 | 91 | 208.0 |
| 54 | 123.0 | 94 | 226.2 |
| 55 | 98.3 | 100 | 246.1 |
| 57 | 103.4 | 101 | 261.0 |
| 57 | 114.2 | 103 | 247.8 |
| 57 | 106.8 | 104 | 253.0 |
| 59 | 124.0 | 110 | 265.6 |
| 60 | 115.5 | | |

**Table VI.** Oxygen consumption of a broiler strain fed the standard diet: group B.

| Wt.(gm) | n | ml/hr | Wt.(gm) | n | ml/hr |
|---|---|---|---|---|---|
| 40 | 2 | 52.4 | 68 | 1 | 117.6 |
| 41 | 3 | 55.8 | 69 | 3 | 140.1 |
| 42 | 4 | 58.8 | 70 | 1 | 163.8 |
| 43 | 2 | 62.3 | 72 | 5 | 143.3 |
| 44 | 5 | 62.5 | 73 | 3 | 134.3 |
| 45 | 3 | 69.8 | 75 | 2 | 132.7 |
| 46 | 4 | 70.8 | 76 | 1 | 164.9 |
| 47 | 4 | 71.9 | 77 | 2 | 171.7 |
| 48 | 5 | 74.4 | 78 | 2 | 152.1 |
| 49 | 4 | 73.0 | 80 | 2 | 183.2 |
| 50 | 4 | 80.5 | 81 | 1 | 143.4 |
| 51 | 3 | 86.2 | 82 | 1 | 149.2 |
| 52 | 4 | 83.7 | 83 | 1 | 162.7 |
| 53 | 3 | 85.3 | 84 | 4 | 174.7 |
| 54 | 2 | 89.6 | 85 | 1 | 190.4 |
| 55 | 4 | 102.8 | 89 | 1 | 188.7 |
| 56 | 3 | 100.2 | 90 | 2 | 191.7 |
| 57 | 5 | 98.6 | 91 | 2 | 175.6 |
| 58 | 5 | 106.1 | 92 | 3 | 207.0 |
| 59 | 3 | 116.8 | 93 | 1 | 170.2 |
| 60 | 6 | 117.6 | 94 | 2 | 194.6 |
| 61 | 4 | 111.0 | 95 | 1 | 174.8 |
| 62 | 3 | 127.1 | 96 | 1 | 200.6 |
| 63 | 3 | 123.5 | 97 | 1 | 167.8 |
| 64 | 5 | 126.1 | 98 | 2 | 201.9 |
| 65 | 3 | 140.4 | 99 | 1 | 183.2 |
| 66 | 3 | 136.6 | 100 | 2 | 224.0 |
| 67 | 2 | 124.0 | | | |

Table VII. Oxygen consumption of laying strain chicks during the first fortnight after hatching; standard diet: group C.

| Wt.(gm) | n | ml/hr | Wt.(gm) | n | ml/hr |
|---|---|---|---|---|---|
| 30 | 1 | 46.7 | 55 | 2 | 130.3 |
| 33 | 1 | 39.2 | 56 | 1 | 147.0 |
| 34 | 1 | 44.8 | 56 | 1 | 136.0 |
| 35 | 2 | 64.1 | 57 | 1 | 150.6 |
| 36 | 4 | 55.9 | 57 | 1 | 126.4 |
| 36 | 1 | 46.6 | 59 | 2 | 163.1 |
| 37 | 3 | 67.3 | 59 | 1 | 172.7 |
| 37 | 1 | 79.0 | 60 | 2 | 149.5 |
| 39 | 4 | 58.9 | 61 | 1 | 148.1 |
| 40 | 1 | 70.9 | 61 | 1 | 151.8 |
| 40 | 2 | 86.0 | 62 | 2 | 155.3 |
| 41 | 2 | 74.2 | 62 | 2 | 146.1 |
| 42 | 5 | 77.5 | 63 | 1 | 188.7 |
| 45 | 2 | 90.6 | 63 | 1 | 184.4 |
| 46 | 1 | 108.9 | 64 | 1 | 144.2 |
| 46 | 2 | 100.0 | 65 | 1 | 141.4 |
| 47 | 3 | 113.7 | 65 | 2 | 161.5 |
| 48 | 4 | 98.5 | 66 | 1 | 151.7 |
| 49 | 2 | 102.0 | 69 | 2 | 170.1 |
| 50 | 1 | 118.4 | 71 | 1 | 191.6 |
| 50 | 3 | 115.4 | 73 | 1 | 190.3 |
| 51 | 3 | 112.4 | 74 | 1 | 193.1 |
| 51 | 4 | 117.7 | 75 | 1 | 192.3 |
| 52 | 4 | 124.2 | 76 | 1 | 217.4 |
| 52 | 2 | 120.1 | 77 | 3 | 187.6 |
| 53 | 2 | 126.8 | 79 | 1 | 237.3 |
| 54 | 4 | 132.8 | 80 | 2 | 224.6 |
| 54 | 1 | 123.0 | 81 | 3 | 207.3 |

Table VII cont'd.

| Wt.(gm) | n | ml/hr | Wt.(gm) | n | ml/hr |
|---|---|---|---|---|---|
| 82 | 3 | 188.9 | 92 | 1 | 216.9 |
| 83 | 1 | 200.8 | 93 | 2 | 205.0 |
| 84 | 1 | 209.5 | 94 | 1 | 213.2 |
| 85 | 1 | 258.3 | 95 | 1 | 211.6 |
| 86 | 1 | 182.3 | 96 | 2 | 256.8 |
| 86 | 1 | 181.2 | 98 | 1 | 223.9 |
| 88 | 1 | 248.4 | 99 | 1 | 255.9 |
| 89 | 4 | 211.7 | 99 | 1 | 264.2 |
| 90 | 1 | 197.4 | 100 | 1 | 247.6 |
| 92 | 4 | 247.1 | | | |

Table VIII. Oxygen requirements of the laying strain. Broiler diet fed throughout the period: group D.

| Wt.(gm) | n | ml/hr | Wt.(gm) | n | ml/hr |
|---|---|---|---|---|---|
| 32 | 1 | 56.1 | 68 | 2 | 178.3 |
| 34 | 2 | 49.5 | 69 | 3 | 191.9 |
| 36 | 3 | 55.5 | 70 | 1 | 163.1 |
| 38 | 4 | 67.3 | 71 | 5 | 186.2 |
| 39 | 5 | 70.8 | 72 | 5 | 188.9 |
| 41 | 1 | 89.0 | 73 | 3 | 201.0 |
| 42 | 1 | 65.3 | 75 | 2 | 190.7 |
| 49 | 1 | 114.0 | 76 | 3 | 204.5 |
| 53 | 2 | 147.0 | 77 | 2 | 206.2 |
| 54 | 3 | 143.9 | 79 | 1 | 198.8 |
| 55 | 3 | 119.7 | 80 | 1 | 204.1 |
| 56 | 2 | 122.4 | 83 | 2 | 229.5 |
| 57 | 2 | 125.1 | 85 | 1 | 198.5 |
| 58 | 3 | 173.2 | 92 | 2 | 212.4 |
| 62 | 1 | 173.6 | 98 | 1 | 257.1 |
| 65 | 1 | 154.4 | 99 | 1 | 279.7 |
| 66 | 2 | 172.6 | 100 | 1 | 232.1 |
| 67 | 3 | 177.9 | | | |

Table IX. The oxygen requirements of lecithectomised and normal chicks, RIR x LS. The standard diet was fed throughout the period: group E.

| | Lecithectomized | | | | Normal | | | |
|---|---|---|---|---|---|---|---|---|
| | ♂♂ | | ♀♀ | | ♂♂ | | ♀♀ | |
| Wt.(gm) | n | ml/hr | n | ml/hr | n | ml/hr | n | ml/hr |
| 36 | | | 2 | 71.7 | | | | |
| 39 | 1 | 76.0 | | | | | | |
| 40 | 1 | 81.7 | | | | | | |
| 41 | 2 | 77.5 | | | | | 1 | 100.8 |
| 42 | | | | | | | 1 | 92.1 |
| 43 | 1 | 73.5 | 1 | 96.2 | | | | |
| 44 | | | 1 | 81.7 | | | | |
| 46 | 1 | 112.8 | | | 1 | 91.3 | | |
| 47 | | | 1 | 114.4 | | | | |
| 48 | 2 | 101.5 | 1 | 110.6 | | | 2 | 125.4 |
| 49 | 4 | 102.5 | 1 | 117.6 | | | | |
| 50 | 2 | 120.4 | | | | | 2 | 119.5 |
| 51 | | | 1 | 121.5 | | | | |
| 52 | 2 | 121.8 | 2 | 136.7 | | | | |
| 53 | 1 | 95.6 | 2 | 114.5 | 1 | 139.9 | | |
| 54 | 1 | 108.9 | 4 | 128.3 | 2 | 111.6 | 1 | 137.9 |
| 55 | 2 | 107.7 | 4 | 115.3 | | | | |
| 56 | 3 | 122.9 | 2 | 139.4 | 1 | 95.9 | | |
| 57 | 2 | 136.5 | 2 | 151.6 | | | 2 | 143.9 |
| 58 | 4 | 133.8 | 3 | 156.0 | 2 | 120.4 | | |
| 59 | 3 | 144.6 | 2 | 129.1 | 1 | 98.3 | 2 | 142.3 |
| 60 | 2 | 151.1 | 1 | 142.4 | 1 | 129.3 | 1 | 122.8 |
| 61 | 2 | 148.0 | 1 | 119.6 | | | 2 | 156.7 |
| 62 | | | 4 | 154.8 | | | | |
| 63 | 2 | 153.3 | 1 | 154.2 | 2 | 137.8 | 1 | 125.9 |
| 64 | 1 | 161.6 | 2 | 153.6 | 2 | 150.3 | 1 | 137.7 |

Table IX cont'd.

| | Lecithectomised | | | | Normal | | | |
|---|---|---|---|---|---|---|---|---|
| | ♂♂ | | ♀♀ | | ♂♂ | | ♀♀ | |
| Wt.(gm) | n | ml/hr | n | ml/hr | n | ml/hr | n | ml/hr |
| 65 | | | 1 | 164.3 | | | 1 | 159.0 |
| 66 | 1 | 124.4 | 2 | 122.5 | | | 2 | 161.7 |
| 67 | 3 | 153.9 | 2 | 147.0 | | | 2 | 140.9 |
| 68 | 3 | 138.8 | 1 | 123.1 | 1 | 133.4 | | |
| 69 | 2 | 168.6 | 2 | 177.0 | | | 1 | 136.1 |
| 70 | 1 | 163.4 | | | | | | |
| 71 | | | 1 | 165.5 | | | 1 | 182.9 |
| 72 | 2 | 158.8 | | | | | | |
| 73 | | | | | | | 3 | 146.7 |
| 74 | 1 | 189.0 | | | 2 | 155.7 | 1 | 129.0 |
| 75 | | | | | | | 1 | 161.8 |
| 76 | | | 1 | 181.7 | | | | |
| 77 | 2 | 188.6 | | | 1 | 155.7 | 1 | 168.8 |
| 79 | 1 | 208.4 | 1 | 203.6 | | | | |
| 80 | 1 | 174.7 | | | | | 1 | 165.5 |
| 81 | | | 2 | 224.1 | | | | |
| 82 | | | 1 | 267.7 | | | 1 | 219.7 |
| 83 | | | 1 | 196.1 | | | | |
| 84 | 1 | 210.9 | | | | | | |
| 85 | | | 1 | 241.4 | 1 | 137.3 | | |
| 87 | 2 | 217.4 | 1 | 199.1 | 1 | 183.6 | | |
| 88 | 1 | 213.8 | 1 | 255.1 | | | | |
| 90 | 1 | 214.8 | | | | | | |
| 92 | 1 | 229.2 | 1 | 213.7 | | | 1 | 209.0 |
| 93 | 1 | 227.8 | | | | | | |
| 94 | 1 | 247.7 | | | | | 1 | 263.1 |
| 95 | | | 3 | 237.1 | | | | |

Table IX cont'd.

| | Lecithectomized | | | | Normal | | | |
|---|---|---|---|---|---|---|---|---|
| | ♂♂ | | ♀♀ | | ♂♂ | | ♀♀ | |
| Wt.(gm) | n | ml/hr | n | ml/hr | n | ml/hr | n | ml/hr |
| 96 | 2 | 229.7 | 2 | 238.6 | | | 1 | 242.9 |
| 97 | 1 | 254.5 | | | | | 1 | 247.2 |
| 98 | 3 | 244.6 | | | | | 1 | 235.2 |
| 99 | 1 | 242.6 | 2 | 257.2 | 1 | 233.6 | 1 | 216.3 |
| 100 | | | | | | | 1 | 267.5 |
| 102 | 1 | 307.5 | 1 | 293.4 | | | | |
| 103 | | | | | | | 1 | 275.4 |
| 104 | | | 2 | 277.3 | | | | |
| 105 | 2 | 244.0 | 1 | 265.2 | | | | |
| 106 | | | | | | | 1 | 278.7 |
| 107 | 2 | 281.1 | | | | | 1 | 275.4 |
| 108 | | | | | | | 1 | 249.7 |
| 109 | 1 | 297.9 | 2 | 310.2 | | | | |
| 110 | | | | | | | 1 | 281.4 |
| 111 | 1 | 293.6 | | | | | | |
| 113 | | | 1 | 305.4 | | | 1 | 291.7 |
| 114 | | | | | | | 1 | 278.2 |
| 115 | | | | | 1 | 224.4 | | |
| 116 | 1 | 289.7 | 1 | 242.8 | | | | |
| 118 | 1 | 308.0 | 1 | 354.7 | | | | |
| 119 | 1 | 260.8 | | | | | | |
| 120 | 1 | 284.8 | | | | | | |
| 121 | | | 2 | 297.4 | | | | |
| 123 | 1 | 239.5 | | | | | | |
| 127 | 1 | 381.5 | | | | | | |
| 128 | 1 | 339.4 | | | | | | |
| 129 | | | | | 1 | 249.7 | | |

Table IX cont'd.

| | Lecithectomized | | | | Normal | | | |
|---|---|---|---|---|---|---|---|---|
| | ♂♂ | | ♀♀ | | ♂♂ | | ♀♀ | |
| Wt.(gm) | n | ml/hr | n | ml/hr | n | ml/hr | n | ml/hr |
| 131 | 1 | 309.7 | | | | | | |
| 132 | | | 1 | 354.0 | | | | |
| 133 | | | 1 | 335.8 | | | | |
| 136 | 2 | 339.3 | | | | | | |
| 138 | 1 | 387.7 | | | | | | |
| 144 | | | 1 | 353.8 | | | | |
| 145 | 1 | 383.2 | | | | | | |
| 146 | | | 1 | 357.8 | | | | |
| 150 | 1 | 365.6 | 1 | 400.8 | | | | |
| 151 | | | 1 | 335.1 | | | | |
| 152 | | | | | | | 1 | 330.9 |
| 153 | | | | | 1 | 280.7 | | |
| 154 | | | | | | | 1 | 349.9 |
| 155 | | | | | | | 1 | 423.5 |
| 156 | | | | | | | 1 | 369.5 |
| 157 | 1 | 411.2 | 1 | 413.3 | | | | |
| 158 | | | | | | | 1 | 449.9 |
| 159 | 2 | 385.3 | | | | | 1 | 381.6 |
| 160 | 2 | 421.7 | 1 | 457.5 | | | | |
| 161 | 1 | 383.3 | | | | | | |
| 162 | 1 | 408.3 | | | | | | |
| 163 | 1 | 421.0 | | | 1 | 453.9 | 1 | 370.9 |
| 164 | | | 1 | 362.4 | | | 2 | 450.3 |
| 165 | | | 1 | 368.3 | | | | |
| 167 | 2 | 377.8 | | | | | | |
| 168 | | | | | | | 1 | 401.4 |
| 169 | | | 1 | 493.5 | | | 1 | 418.2 |

Table IX cont'd.

| | Lecithectomized | | | | Normal | | | |
|---|---|---|---|---|---|---|---|---|
| | ♂♂ | | ♀♀ | | ♂♂ | | ♀♀ | |
| Wt.(gm) | n | ml/hr | n | ml/hr | n | ml/hr | n | ml/hr |
| 172 | 1 | 415.7 | 1 | 471.6 | | | | |
| 173 | 1 | 453.9 | | | | | 1 | 387.9 |
| 174 | 1 | 461.6 | | | | | 1 | 398.3 |
| 175 | 2 | 399.9 | 1 | 441.2 | | | | |
| 176 | | | 2 | 457.5 | | | | |
| 177 | 1 | 524.8 | 2 | 384.6 | | | 1 | 341.5 |
| 178 | 1 | 406.8 | | | | | 2 | 420.9 |
| 179 | 2 | 382.2 | 2 | 421.5 | | | | |
| 182 | | | | | | | 1 | 418.4 |
| 183 | | | | | 2 | 432.8 | 1 | 534.0 |
| 184 | | | | | | | 1 | 457.8 |
| 186 | 1 | 449.9 | | | | | | |
| 187 | | | 1 | 418.8 | | | | |
| 188 | | | 1 | 448.0 | 1 | 456.2 | | |
| 189 | 1 | 479.0 | 2 | 407.6 | | | | |
| 190 | 2 | 464.5 | 1 | 448.5 | 1 | 526.9 | 2 | 429.8 |
| 191 | 1 | 423.1 | | | | | | |
| 192 | | | | | | | 1 | 499.4 |
| 193 | 1 | 457.6 | 1 | 419.2 | | | 1 | 509.8 |
| 195 | 1 | 457.6 | 1 | 509.9 | | | 2 | 443.5 |
| 196 | 1 | 425.4 | | | | | | |
| 197 | 1 | 443.1 | | | 1 | 417.6 | | |
| 198 | | | 1 | 444.1 | 1 | 415.7 | | |
| 199 | 1 | 418.3 | 3 | 455.6 | | | 2 | 453.2 |
| 200 | 1 | 471.3 | | | | | | |
| 201 | | | | | | | 1 | 465.0 |
| 202 | 1 | 522.9 | | | | | | |

Table IX cont'd.

| | Lecithectomized | | | | Normal | | | |
|---|---|---|---|---|---|---|---|---|
| | ♂♂ | | ♀♀ | | ♂♂ | | ♀♀ | |
| Wt.(gm) | n | ml/hr | n | ml/hr | n | ml/hr | n | ml/hr |
| 204 | | | 1 | 457.7 | | | | |
| 207 | 1 | 497.6 | | | | | | |
| 208 | 1 | 458.8 | 1 | 510.4 | | | | |
| 210 | 1 | 547.9 | | | | | | |
| 212 | | | 1 | 513.1 | | | | |
| 213 | 1 | 545.0 | 1 | 467.2 | | | | |
| 216 | | | 1 | 489.6 | | | 1 | 573.3 |
| 218 | | | | | 1 | 451.0 | | |
| 219 | 1 | 527.3 | | | | | | |
| 220 | 1 | 480.0 | | | | | | |
| 221 | | | 1 | 541.7 | | | | |
| 223 | 1 | 554.6 | | | | | | |
| 224 | | | 1 | 489.8 | | | | |
| 225 | 1 | 486.4 | | | | | | |
| 226 | | | 1 | 492.7 | | | | |
| 229 | 1 | 560.7 | | | | | 1 | 481.0 |
| 233 | | | | | | | 1 | 572.6 |
| 234 | | | 1 | 571.9 | | | 1 | 515.7 |
| 236 | | | | | | | 1 | 551.1 |
| 238 | | | | | 1 | 466.1 | | |
| 239 | 1 | 557.5 | 2 | 528.0 | | | | |
| 240 | 1 | 563.6 | | | 1 | 515.0 | 1 | 562.4 |
| 244 | 1 | 652.3 | | | 1 | 568.4 | | |
| 245 | | | 1 | 572.0 | | | | |
| 247 | | | 1 | 573.7 | | | | |
| 250 | 1 | 586.9 | | | | | | |
| 251 | | | 1 | 484.9 | | | | |

Table IX cont'd.

| | Lecithectomized | | | | Normal | | | |
|---|---|---|---|---|---|---|---|---|
| | ♂♂ | | ♀♀ | | ♂♂ | | ♀♀ | |
| Wt.(gm) | n | ml/hr | n | ml/hr | n | ml/hr | n | ml/hr |
| 255 | | | | | | | 1 | 531.9 |
| 260 | | | | | | | 1 | 612.4 |
| 270 | 2 | 614.4 | | | | | 1 | 610.3 |
| 277 | 1 | 539.9 | | | | | | |
| 286 | 1 | 507.7 | | | | | | |
| 290 | 1 | 563.0 | | | | | | |

Table X. Oxygen consumption of a commercial laying strain, broiler ration: group F.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 36 | | | 1 | 108.3 | 76 | 2 | 200.9 | 4 | 198.8 |
| 38 | 1 | 102.1 | 1 | 93.6 | 77 | 1 | 210.7 | 4 | 205.0 |
| 39 | 2 | 86.7 | 1 | 114.1 | 78 | | | 1 | 216.2 |
| 40 | 2 | 103.6 | 1 | 111.0 | 79 | | | 2 | 190.3 |
| 41 | 1 | 110.5 | 1 | 101.3 | 80 | 2 | 211.9 | 4 | 208.4 |
| 46 | | | 2 | 118.9 | 81 | 2 | 221.4 | | |
| 47 | 1 | 131.2 | | | 82 | 2 | 238.9 | 3 | 220.8 |
| 50 | 3 | 147.2 | 1 | 170.6 | 83 | 2 | 248.5 | 1 | 239.2 |
| 53 | 1 | 116.9 | 2 | 161.9 | 84 | 1 | 206.4 | | |
| 54 | 2 | 138.9 | | | 85 | | | 3 | 235.5 |
| 55 | 1 | 157.3 | 1 | 157.3 | 86 | 2 | 224.5 | 1 | 236.0 |
| 56 | 1 | 167.9 | | | 87 | 1 | 238.8 | | |
| 57 | | | 1 | 222.4 | 88 | | | 1 | 193.4 |
| 59 | 1 | 162.2 | 3 | 154.7 | 89 | 3 | 231.4 | 1 | 208.6 |
| 60 | 1 | 213.1 | | | 90 | | | 3 | 216.1 |
| 61 | 3 | 186.6 | | | 91 | 4 | 256.0 | | |
| 62 | 1 | 177.3 | 1 | 177.1 | 92 | | | 1 | 224.1 |
| 64 | 2 | 190.0 | 3 | 191.0 | 93 | 1 | 231.4 | 1 | 242.5 |
| 65 | 1 | 188.9 | 4 | 188.4 | 94 | 2 | 259.3 | | |
| 66 | 2 | 216.0 | 1 | 188.1 | 95 | 1 | 240.0 | | |
| 67 | 1 | 187.4 | 2 | 185.5 | 97 | 1 | 317.0 | | |
| 68 | 2 | 195.8 | 4 | 175.4 | 101 | 1 | 264.6 | | |
| 69 | 1 | 219.0 | 2 | 209.5 | 102 | 1 | 291.7 | | |
| 70 | 1 | 206.3 | 1 | 233.7 | 103 | | | 1 | 315.4 |
| 71 | 2 | 200.0 | 3 | 198.4 | 104 | 1 | 337.5 | | |
| 72 | 5 | 208.5 | 5 | 189.4 | 105 | 1 | 290.3 | 2 | 280.3 |
| 73 | 2 | 222.3 | | | 109 | | | 1 | 299.0 |
| 74 | 2 | 213.9 | 1 | 234.2 | 110 | 1 | 273.1 | | |
| 75 | 3 | 217.3 | 3 | 191.9 | 111 | 1 | 290.6 | | |

Table X cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 113 | 1 | 317.0 | | | 153 | 1 | 408.3 | | |
| 114 | | | 1 | 258.8 | 154 | 1 | 441.3 | | |
| 115 | | | 1 | 340.3 | 155 | 1 | 450.7 | | |
| 116 | 1 | 260.5 | 2 | 280.6 | 156 | 2 | 396.9 | | |
| 119 | | | 1 | 329.8 | 157 | | | 1 | 405.0 |
| 120 | | | 1 | 259.9 | 159 | | | 1 | 377.8 |
| 121 | | | 4 | 276.9 | 160 | 2 | 385.1 | | |
| 123 | 1 | 332.2 | 3 | 293.4 | 165 | 1 | 376.3 | | |
| 124 | | | 1 | 317.7 | 166 | | | 1 | 476.5 |
| 126 | | | 2 | 311.0 | 167 | 1 | 448.1 | | |
| 128 | | | 1 | 302.3 | 168 | 1 | 416.7 | 1 | 390.7 |
| 129 | 1 | 297.8 | 2 | 277.0 | 169 | 2 | 419.1 | | |
| 130 | 1 | 309.7 | 3 | 306.2 | 171 | | | 1 | 413.7 |
| 131 | 1 | 296.7 | 3 | 297.3 | 172 | | | 1 | 442.1 |
| 132 | 1 | 288.9 | 1 | 300.1 | 175 | 1 | 441.4 | | |
| 133 | | | 2 | 316.4 | 181 | | | 1 | 391.7 |
| 134 | | | 1 | 313.2 | 182 | | | 1 | 417.8 |
| 135 | | | 1 | 298.8 | 183 | 1 | 456.4 | | |
| 137 | 3 | 328.0 | 3 | 316.5 | 184 | 1 | 413.6 | 1 | 329.4 |
| 138 | 1 | 283.6 | 1 | 395.3 | 186 | 1 | 467.6 | 2 | 415.2 |
| 139 | 1 | 391.0 | | | 187 | | | 1 | 456.8 |
| 141 | 2 | 380.1 | 3 | 301.4 | 189 | 1 | 475.2 | 2 | 441.9 |
| 142 | 2 | 342.3 | 3 | 306.3 | 191 | 1 | 425.9 | | |
| 143 | 1 | 316.4 | | | 192 | | | 1 | 439.9 |
| 146 | 1 | 338.6 | 2 | 323.9 | 193 | 1 | 437.7 | 1 | 439.2 |
| 147 | | | 2 | 330.9 | 195 | 1 | 591.0 | | |
| 149 | | | 2 | 361.9 | 198 | | | 1 | 366.4 |
| 150 | 4 | 367.5 | 4 | 343.3 | 199 | | | 3 | 482.2 |

Table X cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 202 | | | 1 | 496.3 | 240 | | | 3 | 478.4 |
| 204 | 1 | 461.1 | | | 242 | 1 | 546.0 | | |
| 205 | | | 1 | 512.3 | 246 | | | 1 | 546.6 |
| 206 | | | 2 | 503.6 | 251 | 1 | 495.1 | 1 | 464.0 |
| 207 | | | 1 | 463.1 | 253 | 1 | 626.5 | | |
| 210 | 1 | 454.6 | 2 | 424.3 | 257 | 2 | 556.3 | | |
| 211 | 1 | 469.9 | | | 258 | | | 2 | 517.5 |
| 213 | 1 | 488.6 | | | 260 | 1 | 525.0 | 2 | 535.6 |
| 214 | 1 | 508.6 | 2 | 435.6 | 262 | 1 | 492.0 | | |
| 216 | | | 1 | 433.2 | 264 | 2 | 581.0 | | |
| 217 | | | 1 | 459.8 | 266 | 1 | 601.9 | 1 | 545.5 |
| 218 | | | 1 | 485.7 | 267 | | | 1 | 578.9 |
| 219 | | | 2 | 488.1 | 269 | 2 | 517.9 | | |
| 220 | 2 | 487.0 | | | 271 | | | 1 | 561.0 |
| 221 | | | 1 | 460.1 | 276 | | | 1 | 568.0 |
| 222 | 1 | 534.8 | 1 | 476.3 | 279 | 1 | 488.7 | | |
| 223 | 1 | 599.7 | | | 283 | 1 | 550.3 | 1 | 465.8 |
| 224 | | | 1 | 443.2 | 284 | 1 | 631.3 | 1 | 530.0 |
| 225 | | | 1 | 496.3 | 285 | | | 1 | 631.5 |
| 228 | 2 | 503.4 | | | 287 | | | 1 | 569.9 |
| 231 | | | 1 | 488.5 | 294 | | | 2 | 622.0 |
| 232 | 1 | 550.2 | | | 295 | | | 1 | 559.9 |
| 233 | 1 | 505.1 | | | 296 | | | 1 | 557.3 |
| 234 | 1 | 513.7 | 1 | 516.7 | 297 | | | 1 | 586.6 |
| 235 | 1 | 649.0 | | | 298 | 1 | 525.7 | 1 | 542.0 |
| 237 | | | 2 | 484.6 | 404 | | | 1 | 708.0 |
| 238 | | | 1 | 500.1 | 414 | 1 | 933.3 | 1 | 765.3 |
| 239 | 1 | 477.4 | 2 | 514.1 | 418 | 1 | 992.3 | | |

Table X cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 420 | | | 1 | 992.5 | 620 | 1 | 1221.5 | 1 | 941.8 |
| 422 | | | 1 | 718.4 | 625 | | | 1 | 979.5 |
| 427 | | | 1 | 780.3 | 628 | 1 | | 1 | 975.3 |
| 439 | | | 1 | 844.4 | 634 | | | 1 | 812.8 |
| 446 | | | 1 | 851.2 | 651 | | | 1 | 816.7 |
| 447 | | | 1 | 857.5 | 655 | | | 1 | 832.0 |
| 448 | | | 1 | 765.6 | 660 | 1 | 1259.9 | | |
| 458 | | | 1 | 777.6 | 675 | | | 1 | 988.1 |
| 471 | 1 | 1044.4 | | | 690 | | | 1 | 877.4 |
| 473 | 1 | 1069.7 | | | 707 | | | 1 | 999.1 |
| 485 | 1 | 1063.2 | | | 712 | 1 | 1036.7 | | |
| 487 | 1 | 1139.2 | | | 723 | | | 1 | 934.1 |
| 494 | 1 | 1095.3 | | | 731 | | | 1 | 878.5 |
| 495 | | | 1 | 864.1 | 744 | 1 | 1259.1 | | |
| 500 | | | 1 | 905.5 | 745 | | | 1 | 953.2 |
| 504 | 1 | 1154.9 | | | 747 | | | 1 | 967.9 |
| 512 | | | 1 | 896.2 | 760 | | | 1 | 804.4 |
| 514 | 1 | 1127.6 | | | 765 | | | 1 | 856.4 |
| 516 | 1 | 1044.5 | | | 766 | | | 1 | 957.3 |
| 525 | 1 | 1085.6 | | | 781 | 1 | 1163.4 | | |
| 526 | | | 2 | 773.7 | 784 | | | 2 | 946.7 |
| 533 | | | 1 | 768.7 | 795 | 1 | 1261.3 | | |
| 534 | 1 | 1234.2 | | | 812 | 1 | 1457.9 | | |
| 546 | | | 1 | 844.7 | 820 | | | 1 | 1050.0 |
| 553 | 1 | 1011.2 | | | 823 | | | 1 | 985.9 |
| 580 | | | 1 | 974.2 | 831 | 1 | 1096.2 | | |
| 589 | | | 1 | 874.5 | 834 | 1 | 1452.7 | | |
| 617 | 1 | 967.8 | | | 846 | | | 1 | 925.4 |

Table X cont'd.

| | ♂♂ | | ♀♀ | |
|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr |
| 856 | 1 | 1173.4 | | |
| 858 | | | 1 | 920.4 |
| 865 | | | 1 | 1132.7 |
| 867 | 1 | 1510.2 | | |
| 870 | | | 1 | 939.5 |
| 871 | | | 1 | 1019.0 |
| 872 | | | 2 | 1056.6 |
| 874 | | | 1 | 872.5 |
| 875 | | | 1 | 1037.8 |
| 881 | 1 | 1395.8 | | |
| 885 | | | 1 | 865.9 |
| 886 | | | 1 | 1062.5 |
| 891 | | | 1 | 922.9 |
| 898 | 1 | 1475.5 | | |
| 900 | | | 1 | 1035.5 |
| 917 | | | 1 | 864.5 |
| 923 | 1 | 1378.3 | | |
| 929 | | | 1 | 994.3 |
| 939 | 1 | 1317.8 | | |
| 941 | 1 | 1453.8 | | |
| 944 | | | 2 | 1034.1 |
| 949 | | | 1 | 1024.4 |
| 953 | 1 | | 1 | 1100.7 |
| 954 | | | 1 | 1075.0 |
| 955 | 1 | 1479.3 | | |
| 971 | | | 1 | 858.2 |
| 975 | 1 | 1521.8 | | |
| 977 | | | 1 | 933.1 |
| 989 | 1 | 1668.1 | | |
| 998 | 1 | 1577.8 | | |
| 1007 | 1 | 1497.0 | | |
| 1009 | | | 1 | 1114.1 |
| 1017 | | | 1 | 1131.0 |
| 1066 | 1 | 1341.4 | | |
| 1084 | 1 | 1509.2 | | |
| 1110 | 1 | 1750.5 | | |
| 1115 | 1 | 1407.6 | | |
| 1122 | 1 | 1561.0 | | |
| 1123 | 1 | 1539.0 | | |
| 1132 | 1 | 1728.3 | | |
| 1142 | 1 | 1407.6 | | |
| 1158 | 1 | 1168.8 | | |
| 1178 | 1 | 1402.5 | | |
| 1188 | 1 | 1818.8 | | |
| 1204 | 1 | 1465.3 | | |
| 1209 | 1 | 1293.9 | | |
| 1210 | 1 | 1756.6 | | |
| 1225 | 1 | 1578.6 | | |
| 1240 | 1 | 1581.4 | | |
| 1257 | 1 | 1256.8 | | |
| 1259 | 1 | 1481.4 | | |
| 1271 | 1 | 1554.0 | | |
| 1380 | 1 | 1831.1 | | |

Table XI. Oxygen consumption of the laying strain fed the standard ration: group G.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 35 | 2 | 93.5 | | | 92 | | | 1 | 267.3 |
| 36 | 2 | 96.5 | | | 94 | 1 | 249.5 | | |
| 37 | | | 1 | 108.4 | 95 | 3 | 260.7 | 1 | 269.7 |
| 41 | 1 | 93.5 | | | 96 | 1 | 252.8 | | |
| 43 | 1 | 107.0 | | | 97 | 1 | 263.6 | | |
| 44 | 2 | 137.1 | | | 98 | | | 2 | 245.7 |
| 45 | 3 | 119.4 | | | 99 | 1 | 269.1 | 1 | 259.3 |
| 47 | | | 1 | 148.1 | 101 | 1 | 278.0 | | |
| 48 | 1 | 139.1 | | | 102 | 1 | 297.6 | 1 | 254.4 |
| 49 | 2 | 148.3 | 2 | 147.9 | 103 | 1 | 300.8 | 1 | 294.1 |
| 50 | 4 | 142.0 | | | 104 | 2 | 287.3 | 1 | 260.1 |
| 51 | 1 | 162.7 | 3 | 132.3 | 105 | 1 | 275.3 | | |
| 52 | | | 3 | 117.1 | 106 | 1 | 341.9 | | |
| 53 | 4 | 150.6 | 3 | 131.4 | 107 | | | 1 | 259.6 |
| 54 | 1 | 147.3 | 1 | 150.1 | 109 | 1 | 319.6 | 2 | 281.4 |
| 55 | 3 | 164.1 | 1 | 146.8 | 110 | 1 | 329.4 | | |
| 56 | 1 | 185.9 | | | 111 | 1 | 290.0 | 1 | 307.9 |
| 57 | 1 | 184.3 | | | 112 | 1 | 288.0 | 1 | 278.3 |
| 58 | | | 3 | 173.6 | 113 | 2 | 296.3 | | |
| 59 | 1 | 128.9 | 3 | 178.9 | 115 | | | 1 | 271.6 |
| 60 | 2 | 186.1 | | | 116 | 1 | 304.6 | 1 | 302.5 |
| 61 | 3 | 178.4 | 1 | 182.7 | 118 | | | 1 | 304.3 |
| 62 | 1 | 162.5 | | | 119 | | | 2 | 347.9 |
| 63 | | | 3 | 175.9 | 121 | 1 | 342.5 | | |
| 65 | 2 | 206.3 | 1 | 184.4 | 124 | 1 | 301.1 | | |
| 68 | | | 2 | 181.7 | 125 | | | 1 | 312.0 |
| 86 | 3 | 216.3 | | | 128 | 1 | 313.1 | | |
| 91 | | | 1 | 229.9 | 138 | 1 | 409.0 | | |

Table XI cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 140 | 1 | 387.7 | | | 204 | 1 | 523.6 | | |
| 141 | | | 1 | 355.2 | 208 | 1 | 562.8 | | |
| 142 | 1 | 375.7 | | | 217 | 1 | 478.7 | 1 | 449.0 |
| 145 | | | 1 | 346.6 | 220 | 1 | 588.1 | | |
| 146 | 1 | 372.4 | | | 222 | | | 1 | 455.8 |
| 147 | | | 1 | 398.5 | 225 | | | 2 | 496.2 |
| 149 | 1 | 413.6 | | | 228 | | | 1 | 479.8 |
| 150 | 1 | 368.1 | | | 229 | 2 | 513.9 | | |
| 158 | 1 | 381.6 | | | 232 | | | 1 | 508.3 |
| 167 | 1 | 496.8 | | | 234 | 1 | 524.8 | | |
| 168 | | | 1 | 473.0 | 235 | | | 1 | 491.9 |
| 169 | 1 | 488.2 | | | 236 | | | 1 | 467.3 |
| 171 | 1 | 455.5 | | | 237 | 1 | 520.5 | | |
| 175 | | | 1 | 444.9 | 238 | 1 | 521.6 | 2 | 521.6 |
| 178 | 1 | 508.7 | | | 239 | 1 | 513.3 | | |
| 179 | | | 2 | 419.1 | 240 | 1 | 545.7 | | |
| 180 | 1 | 496.8 | | | 243 | | | 1 | 462.9 |
| 182 | 3 | 469.7 | | | 245 | 1 | 540.3 | | |
| 183 | 1 | 483.8 | | | 246 | 1 | 527.0 | | |
| 184 | | | 1 | 497.9 | 247 | | | 1 | 485.3 |
| 185 | | | 1 | 494.6 | 249 | | | 1 | 506.7 |
| 186 | | | 1 | 478.8 | 250 | 1 | 549.4 | | |
| 190 | 1 | 515.2 | | | 251 | | | 1 | 534.7 |
| 191 | | | 1 | 461.3 | 252 | 1 | 536.8 | | |
| 193 | 1 | 484.9 | 1 | 481.0 | 255 | 2 | 573.8 | | |
| 194 | 1 | 490.8 | | | 256 | | | 1 | 545.0 |
| 197 | | | 1 | 506.2 | 260 | 2 | 571.9 | 1 | 496.9 |
| 202 | | | 1 | 504.0 | 263 | | | 1 | 555.7 |

Table XI cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 264 | 1 | 552.7 | 1 | 411.6 | 636 | | | 1 | 787.4 |
| 265 | 1 | 553.8 | | | 637 | 1 | 808.5 | | |
| 268 | | | 1 | 568.0 | 648 | 1 | 1042.3 | | |
| 269 | 2 | 572.2 | | | 698 | 1 | 849.1 | | |
| 270 | 1 | 553.0 | | | 725 | | | 1 | 772.3 |
| 271 | | | 1 | 606.3 | 735 | 1 | 1077.2 | | |
| 275 | 2 | 573.7 | | | 745 | | | 1 | 873.8 |
| 276 | 1 | 513.5 | | | 746 | 1 | 965.3 | | |
| 277 | | | 1 | 544.8 | 773 | 1 | 1102.1 | | |
| 279 | 1 | 615.1 | | | 803 | | | 1 | 980.6 |
| 282 | 1 | 540.3 | 1 | 629.8 | 831 | 1 | 939.1 | | |
| 283 | 2 | 574.2 | 1 | 573.5 | 837 | 1 | 941.0 | | |
| 289 | | | 1 | 523.7 | 838 | 1 | 1011.1 | | |
| 293 | 1 | 597.1 | | | 842 | | | 1 | 819.9 |
| 295 | 1 | 542.7 | 1 | 533.4 | 850 | | | 1 | 1100.9 |
| 299 | 1 | 601.9 | | | 877 | | | 1 | 1045.7 |
| 339 | | | 1 | 725.9 | 885 | | | 1 | 1023.4 |
| 364 | 1 | 781.4 | | | 887 | | | 1 | 975.5 |
| 379 | 1 | 857.6 | | | 892 | 1 | 1113.6 | | |
| 383 | | | 1 | 694.2 | 894 | 1 | 1223.7 | | |
| 446 | 1 | 796.3 | | | 907 | | | 1 | 959.0 |
| 503 | 1 | 866.2 | | | 938 | | | 1 | 1054.7 |
| 523 | 1 | 674.4 | | | 958 | | | 1 | 1058.0 |
| 534 | 1 | 629.9 | | | 969 | | | 1 | 1053.6 |
| 556 | 1 | 977.7 | | | 991 | | | 1 | 1159.4 |
| 562 | | | 1 | 728.3 | 993 | 1 | 1456.1 | | |
| 577 | | | 1 | 788.2 | 1012 | 1 | 1752.6 | | |

Table XI cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 1070 | 1 | 1269.7 | | | 1322 | 1 | 1450.1 | | |
| 1073 | 1 | 1504.6 | | | 1349 | | | 1 | 1254.4 |
| 1079 | 1 | 1243.7 | | | 1516 | 1 | 1965.0 | | |
| 1160 | 1 | 1866.6 | | | 1534 | 1 | 1573.0 | | |
| 1174 | 1 | 1540.5 | | | 1542 | 1 | 1864.1 | | |
| 1190 | | | 1 | 1324.2 | 1547 | 1 | 2352.9 | | |
| 1223 | | | 1 | 1288.9 | 1612 | 1 | 1799.3 | | |
| 1235 | | | 1 | 1371.5 | 1660 | 1 | 1573.6 | | |
| 1237 | 1 | 1499.0 | | | 1667 | 1 | 1533.8 | | |
| 1241 | | | 1 | 1165.8 | 1669 | 1 | 1796.9 | | |
| 1244 | | | 1 | 1201.0 | 1681 | 1 | 1536.3 | | |
| 1245 | | | 1 | 1349.2 | 1708 | 1 | 1672.4 | | |
| 1250 | | | 3 | 1104.3 | 1760 | 1 | 1915.9 | | |
| 1251 | | | 1 | 1190.6 | 1770 | 1 | 1606.2 | | |
| 1257 | | | 1 | 1036.7 | 1791 | 1 | 1709.4 | | |
| 1274 | | | 1 | 1451.2 | | | | | |

Table XII. Oxygen consumption of the laying strain fed the broiler and standard diet: group H.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 36 | | | 1 | 108.3 | 72 | 5 | 208.5 | 4 | 194.3 |
| 38 | 2 | 101.3 | 2 | 79.4 | 73 | 2 | 222.4 | | |
| 39 | 1 | 72.9 | 1 | 114.1 | 74 | 2 | 213.9 | 1 | 234.2 |
| 40 | 1 | 104.7 | 2 | 106.8 | 75 | 3 | 217.3 | 6 | 197.8 |
| 41 | 1 | 110.5 | 1 | 101.3 | 76 | 2 | 200.8 | 2 | 219.9 |
| 44 | | | 1 | 106.2 | 77 | | | 5 | 195.2 |
| 46 | | | 2 | 118.9 | 78 | | | 1 | 216.2 |
| 47 | 1 | 131.2 | | | 79 | 1 | 229.6 | 3 | 195.3 |
| 49 | 1 | 121.6 | | | 80 | 2 | 211.9 | 4 | 208.4 |
| 50 | 2 | 149.9 | 1 | 170.6 | 81 | 2 | 220.5 | | |
| 53 | 1 | 116.9 | | | 82 | 2 | 238.9 | 3 | 220.8 |
| 54 | 2 | 138.9 | 1 | 174.0 | 83 | 2 | 248.9 | 1 | 239.2 |
| 55 | | | 1 | 157.3 | 84 | 1 | 206.4 | | |
| 56 | 1 | 167.9 | | | 85 | | | 4 | 234.4 |
| 57 | | | 1 | 222.4 | 86 | 2 | 224.5 | 1 | 236.0 |
| 59 | 1 | 162.2 | 2 | 146.3 | 87 | 1 | 238.8 | 1 | 191.8 |
| 60 | 1 | 213.1 | 1 | 171.6 | 88 | 1 | 169.6 | 1 | 193.4 |
| 61 | 2 | 174.3 | | | 89 | 2 | 233.4 | 1 | 208.6 |
| 62 | 1 | 177.3 | 1 | 177.1 | 90 | 1 | 227.5 | 3 | 216.1 |
| 64 | 2 | 190.1 | 4 | 185.5 | 91 | 4 | 256.1 | | |
| 65 | | | 5 | 185.7 | 92 | 1 | 231.4 | 2 | 221.1 |
| 66 | 2 | 205.5 | 1 | 200.3 | 93 | | | 1 | 242.5 |
| 67 | | | 2 | 185.5 | 94 | 2 | 259.4 | | |
| 68 | 3 | 182.9 | 3 | 181.5 | 95 | 1 | 240.0 | | |
| 69 | 1 | 219.0 | 2 | 209.5 | 97 | 1 | 317.0 | | |
| 70 | | | 1 | 233.7 | 101 | 1 | 264.6 | | |
| 71 | 2 | 200.0 | 3 | 198.4 | 102 | 1 | 291.7 | | |

Table XII cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 103 | | | 1 | 315.4 | 143 | 1 | 316.4 | 1 | 318.2 |
| 105 | 2 | 313.9 | 1 | 280.3 | 145 | 1 | 338.6 | | |
| 108 | 1 | 265.1 | | | 146 | | | 2 | 323.9 |
| 109 | | | 1 | 299.0 | 147 | | | 2 | 330.9 |
| 110 | 1 | 273.1 | | | 149 | | | 2 | 361.9 |
| 113 | 1 | 317.0 | | | 150 | 2 | 320.7 | 4 | 343.3 |
| 114 | | | 1 | 258.8 | 152 | | | 2 | 380.9 |
| 115 | | | 1 | 340.3 | 153 | 1 | 408.3 | | |
| 116 | 1 | 260.5 | 2 | 280.6 | 154 | 1 | 441.3 | | |
| 119 | | | 1 | 329.8 | 155 | 1 | 450.7 | | |
| 120 | | | 1 | 259.9 | 156 | 2 | 396.9 | | |
| 121 | | | 4 | 276.8 | 157 | | | 1 | 405.0 |
| 123 | 1 | 332.2 | 3 | 293.4 | 158 | | | 1 | 377.8 |
| 124 | | | 1 | 317.7 | 160 | 2 | 385.1 | | |
| 126 | | | 2 | 310.9 | 161 | | | 2 | 400.5 |
| 128 | | | 1 | 302.3 | 163 | 1 | 341.0 | | |
| 129 | 1 | 297.8 | 2 | 276.9 | 165 | 1 | 376.3 | | |
| 130 | 1 | 309.7 | 4 | 308.4 | 166 | | | 1 | 476.3 |
| 131 | 1 | 296.7 | 4 | 304.7 | 167 | 1 | 448.1 | | |
| 132 | 1 | 288.9 | 1 | 300.6 | 168 | 1 | 416.7 | 1 | 390.7 |
| 133 | | | 3 | 304.7 | 169 | 2 | 419.2 | | |
| 135 | | | 1 | 298.8 | 171 | | | 1 | 413.7 |
| 137 | 3 | 328.0 | 3 | 316.5 | 175 | 1 | 441.4 | | |
| 138 | 1 | 283.6 | 1 | 395.3 | 181 | | | 1 | 391.7 |
| 139 | 1 | 391.0 | | | 182 | 1 | 522.4 | | |
| 141 | 1 | 408.3 | 5 | 323.6 | 184 | | | 1 | 329.4 |
| 142 | 2 | 342.4 | 3 | 306.3 | 186 | 1 | 467.6 | 1 | 411.5 |

Table XII cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 188 | | | 1 | 456.8 | 364 | | | 1 | 693.4 |
| 193 | | | 1 | 439.2 | 365 | 1 | 794.7 | | |
| 196 | 1 | 517.4 | | | 366 | 1 | 812.7 | | |
| 198 | | | 1 | 366.4 | 378 | 2 | 775.8 | | |
| 199 | | | 1 | 506.2 | 384 | | | 1 | 578.2 |
| 202 | | | 1 | 496.3 | 391 | 1 | 812.9 | | |
| 204 | 1 | 461.1 | | | 392 | | | 1 | 748.2 |
| 211 | 1 | 469.9 | | | 394 | 1 | 783.7 | | |
| 213 | 1 | 488.6 | | | 404 | | | 3 | 739.6 |
| 220 | 1 | 484.2 | | | 406 | | | 1 | 695.3 |
| 223 | 1 | 534.8 | | | 414 | | | 2 | 746.9 |
| 228 | 1 | 509.5 | | | 419 | | | 1 | 727.0 |
| 232 | 1 | 550.2 | | | 432 | 1 | 964.3 | | |
| 239 | | | 1 | 521.6 | 437 | | | 1 | 832.1 |
| 302 | | | 2 | 650.6 | 441 | 1 | 891.7 | | |
| 311 | | | 1 | 696.7 | 442 | | | 1 | 790.7 |
| 312 | | | 1 | 689.3 | 457 | | | 1 | 787.2 |
| 318 | | | 2 | 697.5 | 467 | 1 | 871.2 | 1 | 699.7 |
| 323 | | | 1 | 693.8 | 472 | | | 1 | 681.5 |
| 326 | | | 1 | 647.2 | 473 | | | 1 | 821.2 |
| 329 | 1 | 834.3 | | | 475 | | | 1 | 705.1 |
| 333 | | | 1 | 858.8 | 481 | 1 | 865.6 | | |
| 337 | 1 | 786.0 | | | 487 | | | 1 | 693.7 |
| 341 | | | 1 | 712.9 | 491 | 1 | 874.6 | | |
| 355 | 1 | 775.8 | | | 495 | | | 1 | 848.2 |
| 360 | 1 | 731.6 | | | 500 | | | 1 | 907.7 |
| 361 | | | 1 | 751.1 | 505 | 1 | 846.6 | | |
| 362 | 1 | 659.9 | | | 506 | 1 | 856.7 | | |

Table XII cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 520 | 1 | 983.6 | | | 744 | 1 | 1034.5 | | |
| 521 | | | 1 | 994.7 | 761 | | | 1 | 769.2 |
| 526 | 1 | 930.3 | | | 765 | | | 1 | 840.3 |
| 533 | 1 | 1061.4 | | | 789 | 2 | 900.2 | | |
| 537 | | | 1 | 985.8 | 802 | | | 1 | 724.4 |
| 538 | 1 | 904.8 | 1 | 729.1 | 806 | 1 | 901.9 | | |
| 541 | | | 1 | 738.9 | 818 | 1 | 923.6 | | |
| 551 | 1 | 1021.1 | | | 820 | 1 | 1061.2 | | |
| 559 | | | 1 | 743.7 | 835 | 1 | 1193.1 | | |
| 560 | | | 1 | 868.5 | 844 | 1 | 1131.3 | | |
| 562 | | | 1 | 935.8 | 845 | | | 1 | 711.5 |
| 590 | | | 1 | 700.8 | 849 | | | 1 | 850.9 |
| 611 | 1 | 970.5 | | | 851 | 1 | 957.8 | | |
| 612 | | | 1 | 1123.4 | 878 | | | 1 | 1099.2 |
| 629 | | | 1 | 857.0 | 889 | 1 | 1232.5 | | |
| 636 | | | 1 | 930.5 | 906 | | | 1 | 920.0 |
| 644 | 1 | 903.9 | 1 | 736.9 | 933 | 1 | 1060.1 | | |
| 647 | 1 | 904.8 | | | 951 | 1 | 993.4 | | |
| 657 | | | 1 | 805.9 | 985 | | | 1 | 928.2 |
| 661 | 1 | 979.2 | 1 | 804.1 | 992 | | | 1 | 867.3 |
| 662 | 1 | 993.5 | | | 1021 | | | 1 | 870.6 |
| 666 | | | 1 | 740.2 | 1026 | | | 1 | 944.0 |
| 672 | 1 | 1068.6 | | | 1034 | | | 1 | 751.6 |
| 679 | | | 1 | 700.4 | 1036 | | | 1 | 1082.6 |
| 685 | 1 | 975.6 | | | 1066 | | | 1 | 787.3 |
| 694 | 1 | 1197.4 | | | 1133 | | | 1 | 836.3 |
| 708 | | | 1 | 1127.8 | 1134 | 1 | 1173.2 | | |
| 712 | 1 | 903.4 | 1 | 776.2 | 1165 | | | 1 | 822.6 |

Table XII cont'd.

| | ♂♂ | | ♀♀ | | | ♂♂ | | ♀♀ | |
|---|---|---|---|---|---|---|---|---|---|
| Wt. | n | ml/hr | n | ml/hr | Wt. | n | ml/hr | n | ml/hr |
| 1179 | | | 1 | 1203.4 | 1435 | 1 | 1393.4 | | |
| 1193 | | | 1 | 1049.1 | 1443 | 1 | 1834.2 | | |
| 1196 | | | 1 | 826.4 | 1460 | 1 | 1862.4 | | |
| 1205 | 1 | 1115.5 | | | 1469 | 1 | 1351.6 | | |
| 1250 | | | 1 | 1496.1 | 1487 | 1 | 1517.8 | | |
| 1256 | 1 | 1292.9 | | | 1600 | 1 | 1774.8 | | |
| 1291 | 1 | 1251.5 | | | 1644 | 1 | 1679.8 | | |
| 1298 | | | 1 | 1174.3 | 1696 | 1 | 1761.6 | | |
| 1330 | | | 1 | 1395.7 | 1715 | 1 | 1710.1 | | |
| 1384 | | | 1 | 1327.6 | 1756 | 1 | 1934.8 | | |
| 1424 | 1 | 1540.1 | | | 1759 | 1 | 1929.8 | | |

Table XIII. Oxygen consumption of the fowl as affected by the calorific content of the diet. R x S ♂♂: group I. H.E. = high energy; N.E. = normal energy

| Wt. (gm) | H.E.diet ml/hr | N.E.diet ml/hr | Wt. (gm) | H.E.diet ml/hr | N.E.diet ml/hr |
|---|---|---|---|---|---|
| 41 | | 46.8 | 137 | | 281.9 |
| 44 | 52.7 | | 141 | 346.0 | |
| 48 | | 83.3 | 144 | 253.4 | |
| 48 | | 83.9 | 145 | 274.0 | |
| 50 | 86.9 | | 147 | | 271.0 |
| 50 | 86.8 | | 150 | | 268.1 |
| 51 | | 88.0 | 152 | 245.0 | |
| 53 | 111.5 | | 157 | 300.0 | |
| 56 | 102.2 | | 158 | | 325.9 |
| 57 | | 124.2 | 163 | 276.2 | |
| 58 | | 112.3 | 164 | | 334.0 |
| 62 | 122.1 | | 165 | 295.3 | |
| 63 | 123.3 | | 167 | 263.2 | |
| 73 | | 133.4 | 171 | | 319.9 |
| 80 | 157.6 | 147.1 | 173 | 286.9 | |
| 83 | | 128.3 | 177 | | 250.8 |
| 84 | 164.6 | | 181 | 331.2 | |
| 85 | 151.5 | | 188 | | 323.1 |
| 88 | | 207.2 | 198 | 329.2 | 309.8 |
| 93 | | 191.1 | 207 | 331.1 | |
| 93 | | 195.3 | 209 | | 366.4 |
| 94 | 208.9 | | 212 | 346.0 | |
| 100 | 164.1 | | 218 | 354.3 | 332.2 |
| 101 | 172.8 | 218.0 | 228 | | 354.1 |
| 104 | | 235.3 | 232 | | 258.9 |
| 105 | 183.6 | 189.1 | 251 | 405.3 | |
| 113 | 234.1 | | 253 | | 320.8 |
| 129 | | 226.1 | 254 | | 437.0 |

Table XIII cont'd.

| Wt. (gm) | H.E.diet ml/hr | N.E.diet ml/hr | Wt. (gm) | H.E.diet ml/hr | N.E.diet ml/hr |
|---|---|---|---|---|---|
| 261 | | 382.0 | 426 | | 649.9 |
| 266 | | 375.8 | 430 | | 575.9 |
| 281 | 347.1 | 366.0 | 435 | | 684.0 |
| 287 | 415.3 | | 439 | 508.1 | |
| 288 | | 369.9 | 443 | 561.3 | |
| 290 | 336.6 | | 446 | 757.9 | |
| 290 | 413.5 | | 448 | 563.9 | |
| 292 | | 481.8 | 456 | 507.9 | |
| 302 | 422.5 | | 460 | | 803.3 |
| 304 | 412.9 | | 461 | 685.0 | 686.7 |
| 306 | 453.1 | | 463 | | 667.9 |
| 307 | | 378.9 | 469 | 612.0 | |
| 313 | | 338.5 | 471 | 699.3 | |
| 320 | 367.2 | 514.0 | 478 | | 817.9 |
| 330 | 406.2 | | 479 | 568.5 | |
| 332 | 587.9 | | 503 | | 583.5 |
| 339 | 517.5 | | 521 | 696.6 | |
| 341 | 602.8 | 551.9 | 525 | 662.0 | 662.0 |
| 352 | | 524.9 | 531 | | 654.8 |
| 354 | 492.3 | | 556 | | 588.5 |
| 362 | 411.0 | | 587 | 785.5 | |
| 382 | | 359.0 | 590 | 658.4 | |
| 400 | | 407.9 | 595 | | 699.4 |
| 401 | 491.0 | | 596 | | 854.6 |
| 410 | 546.8 | | 598 | | 781.2 |
| 414 | | 725.7 | 601 | 756.5 | |
| 422 | 510.8 | | 607 | 686.5 | |
| 426 | | 486.1 | 617 | 880.5 | |

Table XIII cont'd.

| Wt. (gm) | H.E.diet ml/hr | N.E.diet ml/hr | Wt. (gm) | H.E.diet ml/hr | N.E.diet ml/hr |
|---|---|---|---|---|---|
| 623 | | 947.2 | 849 | 743.2 | |
| 626 | 629.2 | | 856 | 728.5 | |
| 628 | 714.0 | | 858 | 740.1 | |
| 631 | | 823.5 | 862 | 1074.5 | |
| 644 | 884.9 | | 876 | | 868.4 |
| 645 | 646.8 | | 897 | | 956.8 |
| 667 | 664.1 | 805.9 | 911 | 887.3 | |
| 685 | 820.0 | | 926 | | 939.9 |
| 686 | 850.4 | | 939 | | 895.9 |
| 691 | 874.3 | | 944 | | 906.5 |
| 694 | | 836.6 | 949 | | 769.5 |
| 702 | 728.0 | | 967 | 973.8 | |
| 706 | | 888.3 | 985 | 848.6 | |
| 707 | | 925.3 | 993 | 983.2 | |
| 714 | | 741.8 | 994 | | 905.1 |
| 745 | | 753.7 | 997 | | 709.8 |
| 750 | 808.5 | | 1016 | 1091.9 | |
| 751 | | 904.2 | 1016 | 916.3 | |
| 751 | | 765.2 | 1026 | | 890.0 |
| 779 | | 877.5 | 1028 | | 942.0 |
| 780 | 841.9 | | 1033 | 1058.0 | |
| 781 | 728.6 | | 1034 | 1045.9 | |
| 801 | 924.3 | | 1071 | | 949.2 |
| 801 | 718.5 | | 1072 | 1205.0 | |
| 816 | | 847.5 | 1080 | 952.0 | |
| 823 | | 1042.9 | 1082 | 1025.9 | |
| 828 | | 930.1 | 1092 | | 1066.1 |
| 830 | | 906.5 | 1093 | | 899.0 |
| 830 | | 856.4 | 1109 | 1131.8 | |

Table XIII cont'd.

| Wt. (gm) | H.E.diet ml/hr | N.E.diet ml/hr | Wt. (gm) | H.E.diet ml/hr | N.E.diet ml/hr |
|---|---|---|---|---|---|
| 1117 | | 1114.9 | 1126 | | 1050.8 |

Table XIV. The effect of dietary fat on the oxygen consumption of the male RIR x LS fowl

| Wt. (gm) | High fat ml/hr | Normal fat ml/hr | Wt. (gm) | High fat ml/hr | Normal fat ml/hr |
|---|---|---|---|---|---|
| 40 | 57.1 | 73.2 | 61 | 156.1 | 156.5 |
| 44 | 70.0 | 73.1 | 62 | | 127.1 |
| 44 | 93.4 | 73.2 | 63 | | 99.1 |
| 45 | | 88.9 | 64 | 159.8 | |
| 46 | 86.8 | | 67 | | 147.6 |
| 47 | 89.1 | | 70 | 217.5 | 141.3 |
| 48 | | 79.4 | 71 | 147.7 | |
| 48 | | 66.6 | 73 | 215.3 | |
| 48 | | 69.1 | 76 | 177.7 | 154.5 |
| 48 | | 105.9 | 81 | 97.2 | |
| 49 | 71.1 | | 81 | 170.2 | |
| 50 | 110.1 | | 81 | 176.3 | |
| 50 | 78.2 | | 82 | | 164.2 |
| 50 | 80.2 | | 82 | | 178.3 |
| 51 | 89.6 | | 83 | 169.8 | |
| 51 | 75.0 | | 84 | 191.2 | 217.9 |
| 52 | | 77.0 | 85 | 160.9 | |
| 52 | | 90.7 | 88 | | 181.5 |
| 52 | | 96.6 | 88 | | 206.2 |
| 53 | 87.9 | 111.2 | 89 | 198.7 | 175.0 |
| 54 | 81.0 | 100.0 | 89 | 175.0 | |
| 54 | 107.8 | 98.4 | 91 | 203.8 | |
| 55 | 117.0 | | 92 | 201.7 | 151.4 |
| 56 | 103.4 | 93.1 | 94 | 224.6 | 194.9 |
| 57 | 140.2 | | 95 | 192.8 | |
| 59 | 115.8 | 109.9 | 97 | | 188.5 |
| 59 | 130.3 | | 99 | | 222.0 |
| 60 | 161.8 | | 100 | | 202.7 |

Table XIV cont'd.

| Wt. (gm) | High fat ml/hr | Normal fat ml/hr | Wt. (gm) | High fat ml/hr | Normal fat ml/hr |
|---|---|---|---|---|---|
| 100 | | 206.0 | 170 | | 240.0 |
| 102 | 255.2 | 196.8 | 174 | 361.8 | |
| 103 | 254.0 | 190.6 | 176 | | 306.4 |
| 107 | 195.7 | | 179 | 361.3 | |
| 109 | | 209.3 | 184 | 332.3 | 460.2 |
| 109 | | 208.7 | 187 | | 346.6 |
| 110 | 226.0 | | 191 | | 335.4 |
| 114 | 218.0 | | 192 | 321.8 | |
| 116 | | 251.7 | 193 | 411.6 | |
| 119 | | 297.2 | 194 | 357.0 | |
| 120 | 314.0 | | 198 | | 386.0 |
| 125 | | 238.4 | 207 | 329.1 | |
| 126 | | 280.5 | 218 | 457.0 | |
| 128 | 274.4 | 274.5 | 219 | | 374.2 |
| 128 | 298.4 | | 222 | 415.2 | |
| 129 | | 304.0 | 223 | 458.0 | |
| 130 | | 272.6 | 226 | | 465.7 |
| 135 | | 269.4 | 227 | | 398.0 |
| 137 | 264.2 | | 235 | | 372.7 |
| 137 | 267.3 | | 253 | | 369.9 |
| 143 | | 303.3 | 253 | | 473.0 |
| 149 | | 390.3 | 259 | | 411.5 |
| 150 | 313.7 | | 264 | | 418.5 |
| 154 | 323.0 | | 266 | | 377.6 |
| 155 | | 364.0 | 267 | | 324.3 |
| 156 | | 378.2 | 268 | 363.6 | |
| 159 | | 290.8 | 269 | | 328.7 |
| 161 | 381.6 | | 270 | 317.7 | |
| 169 | | 355.2 | 271 | 379.0 | |

Table XIV cont'd.

| Wt. (gm) | High fat ml/hr | Normal fat ml/hr | Wt. (gm) | High fat ml/hr | Normal fat ml/hr |
|---|---|---|---|---|---|
| 273 | 489.2 | | 448 | 1006.7 | |
| 280 | | 544.4 | 451 | | 524.0 |
| 282 | 411.5 | | 452 | 773.6 | |
| 296 | | 323.2 | 454 | 1204.7 | |
| 304 | 563.4 | | 455 | 663.9 | |
| 315 | 601.6 | | 457 | | 736.7 |
| 319 | 769.8 | | 463 | | 775.9 |
| 335 | | 685.5 | 471 | | 754.6 |
| 337 | 510.1 | | 493 | 1022.3 | |
| 341 | | 171.5 | 500 | 1072.6 | |
| 351 | 754.2 | | 523 | | 789.4 |
| 364 | | 708.5 | 529 | 1109.5 | |
| 366 | | 569.2 | 530 | | 930.2 |
| 367 | | 782.4 | 531 | 835.9 | |
| 371 | | 758.2 | 538 | | 1169.1 |
| 373 | | 835.4 | 549 | 862.2 | 833.8 |
| 385 | 927.9 | | 552 | | 1323.7 |
| 387 | 944.4 | | 557 | 1077.0 | 986.9 |
| 389 | | 612.9 | 558 | | 1013.5 |
| 397 | 793.4 | | 559 | | 1034.2 |
| 402 | | 1016.7 | 562 | | 889.8 |
| 404 | 785.7 | | 566 | | 951.3 |
| 405 | | 998.9 | 570 | | 1190.9 |
| 412 | | 845.3 | 572 | 1265.1 | |
| 419 | | 763.6 | 572 | 1035.6 | |
| 422 | 718.0 | | 586 | | 1071.1 |
| 424 | 956.1 | | 589 | 1023.4 | |
| 430 | 927.0 | 944.9 | 594 | 942.4 | |

Table XIV cont'd.

| Wt. (gm) | High fat ml/hr | Normal fat ml/hr | Wt. (gm) | High fat ml/hr | Normal fat ml/hr |
|---|---|---|---|---|---|
| 597 | 1063.1 | | 769 | | 1266.0 |
| 600 | | 971.5 | 773 | | 1284.0 |
| 604 | 828.3 | | 781 | | 1111.1 |
| 615 | 974.0 | | 795 | | 1219.7 |
| 621 | 973.4 | 1036.7 | 803 | | 1204.0 |
| 624 | 989.2 | | 806 | 785.0 | |
| 633 | 1438.5 | 913.0 | 810 | 1402.8 | |
| 633 | | 1118.9 | 817 | 1071.6 | |
| 646 | 962.4 | | 823 | | 981.2 |
| 654 | | 1014.5 | 827 | 957.1 | |
| 655 | 1177.7 | | 829 | | 1241.0 |
| 656 | | 1238.8 | 829 | | 994.0 |
| 663 | 970.1 | 1046.7 | 830 | 1248.9 | |
| 666 | 997.0 | | 842 | | 819.1 |
| 669 | 1390.8 | | 853 | | 897.6 |
| 681 | 1082.9 | | 878 | 1068.0 | |
| 690 | 987.0 | | 896 | 1222.8 | |
| 708 | 1068.2 | | 907 | 1175.0 | |
| 713 | | 1153.0 | 919 | | 1338.6 |
| 728 | | 1355.3 | 924 | 1156.2 | |
| 730 | 1002.0 | | 930 | | 1256.2 |
| 734 | | 1010.7 | 939 | 1205.0 | |
| 735 | | 1455.5 | 952 | | 1295.0 |
| 743 | 1049.0 | | 972 | 1004.5 | |
| 747 | 1188.0 | | 991 | | 1329.3 |
| 755 | 1176.5 | 919.3 | 992 | 1095.7 | |
| 766 | 1051.2 | | 997 | | 1379.4 |
| 766 | 877.4 | | 1007 | | 1108.7 |

Table XIV cont'd.

| Wt. (gm) | High fat ml/hr | Normal fat ml/hr | Wt. (gm) | High fat ml/hr | Normal fat ml/hr |
|---|---|---|---|---|---|
| 1020 | | 1355.3 | 1036 | 1100.8 | |
| 1021 | | 1300.6 | 1041 | | 1422.7 |
| 1025 | 1058.6 | | 1069 | 1111.8 | |
| 1035 | | 1310.0 | | | |

Table XV. The effect of the protein content of the diet on the oxygen consumption of the RIRx IS male chicken

| Wt. (gm) | High protein ml/hr | Normal protein ml/hr | Wt. (gm) | High protein ml/hr | Normal protein ml/hr |
|---|---|---|---|---|---|
| 37 | 54.5 | | 73 | 148.7 | |
| 40 | 83.7 | | 74 | | 117.6 |
| 42 | 72.5 | | 75 | 120.7 | |
| 43 | 88.2 | | 77 | 147.9 | |
| 43 | 69.4 | | 78 | 139.5 | |
| 44 | 63.4 | | 81 | 209.7 | |
| 50 | | 65.5 | 83 | | 185.3 |
| 53 | 97.7 | 92.0 | 85 | | 192.2 |
| 55 | 114.7 | | 86 | | 185.5 |
| 55 | 104.1 | | 88 | 218.6 | |
| 56 | 134.9 | | 88 | 214.4 | |
| 57 | 105.6 | | 89 | | 208.7 |
| 59 | 128.1 | 111.4 | 91 | 183.6 | 203.1 |
| 59 | 106.0 | | 91 | 283.9 | 221.9 |
| 60 | 124.7 | 90.1 | 92 | | 205.3 |
| 61 | | 95.9 | 92 | | 217.6 |
| 62 | 116.5 | | 93 | 216.5 | |
| 63 | | 101.7 | 94 | 252.9 | |
| 64 | 153.8 | 107.3 | 94 | 234.7 | |
| 65 | | 93.6 | 97 | 250.0 | 216.1 |
| 67 | 126.2 | 135.8 | 99 | | 227.7 |
| 68 | 133.3 | | 101 | 271.4 | 195.6 |
| 69 | 163.8 | | 102 | | 239.5 |
| 69 | 112.9 | | 102 | | 236.8 |
| 72 | | 130.1 | 106 | 204.7 | 255.6 |
| 72 | | 128.1 | 107 | | 211.7 |
| 72 | | 207.3 | 114 | 228.0 | 217.3 |

Table XV cont'd.

| Wt. (gm) | High protein ml/hr | Normal protein ml/hr | Wt. (gm) | High protein ml/hr | Normal protein ml/hr |
|---|---|---|---|---|---|
| 115 | 280.6 | | 164 | 377.4 | |
| 118 | 287.9 | | 168 | | 401.0 |
| 120 | 276.3 | | 173 | | 466.1 |
| 121 | 250.8 | 313.6 | 179 | 341.9 | |
| 122 | 281.9 | 315.7 | 183 | 396.5 | |
| 122 | | 265.8 | 185 | | 352.6 |
| 123 | 309.3 | 270.0 | 186 | 461.3 | 367.7 |
| 123 | 294.7 | | 187 | 355.9 | 397.6 |
| 124 | 286.0 | | 188 | 423.9 | |
| 126 | 257.0 | 262.3 | 200 | | 485.0 |
| 126 | | 230.0 | 201 | 453.1 | |
| 127 | 308.8 | | 205 | 463.7 | 396.5 |
| 128 | | 275.3 | 205 | 430.4 | |
| 129 | | 287.9 | 206 | 445.8 | |
| 131 | | 267.7 | 208 | 488.5 | |
| 132 | 278.5 | | 211 | 475.1 | 395.1 |
| 137 | 310.7 | | 211 | | 454.1 |
| 139 | 289.1 | | 217 | 526.9 | |
| 141 | | 271.1 | 218 | | 560.8 |
| 141 | | 327.5 | 219 | 474.5 | |
| 145 | | 287.9 | 220 | 536.3 | 441.2 |
| 148 | | 298.9 | 220 | 468.4 | |
| 149 | | 260.2 | 221 | 468.4 | |
| 152 | 307.5 | | 223 | 454.1 | |
| 154 | | 340.8 | 223 | 621.3 | |
| 156 | | 324.7 | 227 | 454.4 | |
| 157 | 329.0 | 358.0 | 228 | 459.5 | 481.4 |

Table XV cont'd.

| Wt. (gm) | High protein ml/hr | Normal protein ml/hr | Wt. (gm) | High protein ml/hr | Normal protein ml/hr |
|---|---|---|---|---|---|
| 229 | | 437.2 | 315 | | 476.8 |
| 234 | 495.3 | | 316 | | 416.8 |
| 236 | | 421.3 | 319 | 672.6 | |
| 239 | 582.3 | | 322 | 590.1 | |
| 240 | | 535.8 | 329 | 435.3 | |
| 242 | 554.0 | | 332 | 482.8 | |
| 245 | 531.0 | 439.8 | 333 | 376.9 | 697.1 |
| 245 | 513.2 | | 334 | | 425.9 |
| 246 | | 518.6 | 337 | 401.3 | |
| 252 | 527.4 | | 338 | 390.6 | |
| 253 | | 490.3 | 339 | | 541.3 |
| 257 | 522.0 | | 342 | 544.6 | |
| 258 | | 549.0 | 346 | | 536.9 |
| 259 | 509.8 | | 348 | 467.4 | |
| 262 | | 509.4 | 348 | 496.9 | |
| 265 | | 592.2 | 349 | | 481.1 |
| 266 | 540.0 | | 351 | 616.5 | |
| 268 | 581.4 | | 359 | 448.2 | |
| 269 | | 495.3 | 361 | 569.5 | 646.1 |
| 273 | | 594.0 | 363 | 455.0 | |
| 276 | | 600.1 | 365 | | 582.2 |
| 282 | 511.2 | | 366 | 469.6 | |
| 289 | 425.9 | | 371 | 597.0 | |
| 290 | | 550.7 | 373 | | 670.0 |
| 308 | | 485.9 | 374 | | 502.4 |
| 308 | | 479.1 | 378 | | 493.1 |
| 309 | | 453.6 | 380 | 772.1 | |

Table XV cont'd.

| Wt. (gm) | High protein ml/hr | Normal protein ml/hr | Wt. (gm) | High protein ml/hr | Normal protein ml/hr |
|---|---|---|---|---|---|
| 382 | 460.9 | | 473 | 612.7 | |
| 386 | 766.2 | | 478 | 776.5 | |
| 389 | 693.4 | 487.1 | 479 | 670.0 | |
| 393 | 558.7 | | 479 | 756.2 | |
| 398 | 551.4 | | 486 | 631.1 | |
| 400 | | 710.2 | 488 | | 743.4 |
| 404 | | 808.8 | 489 | | 607.9 |
| 407 | | 526.2 | 489 | | 850.0 |
| 409 | | 586.0 | 490 | | 643.7 |
| 410 | 686.3 | | 492 | 856.5 | |
| 411 | | 541.5 | 497 | | 850.0 |
| 428 | 467.7 | | 506 | 771.0 | |
| 429 | 632.7 | | 507 | | 646.7 |
| 430 | 647.4 | 752.6 | 510 | | 679.7 |
| 432 | | 724.4 | 512 | | 685.2 |
| 433 | | 602.3 | 514 | | 710.2 |
| 435 | | 715.2 | 517 | 574.7 | 772.8 |
| 436 | | 604.2 | 520 | 548.9 | |
| 443 | 645.8 | | 522 | 670.5 | 827.0 |
| 444 | | 739.1 | 527 | | 779.2 |
| 448 | 764.8 | | 534 | 688.9 | |
| 448 | 567.8 | | 545 | 773.6 | |
| 450 | 662.1 | | 547 | | 679.7 |
| 455 | 739.7 | | 550 | 740.3 | |
| 458 | 704.7 | | 552 | 655.0 | |
| 460 | 586.9 | 804.1 | 554 | | 810.5 |
| 465 | 746.0 | | 563 | | 654.0 |
| 473 | 631.8 | | 568 | | 757.5 |

Table XV cont'd.

| Wt. (gm) | High protein ml/hr | Normal protein ml/hr | Wt. (gm) | High protein ml/hr | Normal protein ml/hr |
|---|---|---|---|---|---|
| 572 | 977.9 | | 650 | | 1044.5 |
| 581 | 839.0 | | 653 | | 822.4 |
| 581 | 672.3 | | 662 | 1207.8 | |
| 582 | 622.6 | | 686 | 837.2 | |
| 596 | 1053.4 | | 690 | | 1254.0 |
| 600 | | 638.5 | 693 | 812.3 | |
| 609 | 873.9 | | 722 | 981.0 | |
| 617 | | 723.6 | 742 | | 1084.3 |
| 619 | | 1022.2 | 744 | | 1031.0 |
| 620 | | 996.4 | 782 | 1051.1 | |
| 626 | | 749.3 | 1026 | 1248.6 | |
| 632 | 959.3 | | 1044 | | 1360.0 |
| 634 | 704.7 | | 1055 | | 1206.0 |
| 637 | 957.5 | | 1100 | | 1365.6 |
| 645 | 1052.9 | | 1111 | 1324.7 | |
| 649 | 682.6 | 717.6 | 1147 | 1191.0 | |

# PART VI

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