THE INFLUENCE OF GRADED LEVELS OF ATMOSPHERIC AMMONIA ON CHICKENS

I. EFFECTS ON RESPIRATION AND ON THE PERFORMANCE OF BROILERS AND REPLACEMENT GROWING STOCK

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Received for publication 19th October 1965

SYNOPSIS

Experiments have been carried out to study the influence of high concentrations of ammonia on the performance of chickens. One hundred parts per million by volume of ammonia caused reductions in the respiration rate of adult hens of between 7 and 24 per cent. Carbon dioxide production and respiratory depth were also reduced.

Broiler chickens reared in atmospheres containing high concentrations of ammonia from 28 days of age tended to eat less food than broilers reared in ammonia-free atmospheres and at 100 p.p.m. of ammonia their growth rate was significantly reduced.

Replacement laying pullets reared in atmospheres containing ammonia from 11 to 18 weeks of age, ate less food than similar birds reared in ammonia-free atmospheres. When the ammonia level was 78 p.p.m. by volume, food consumption was significantly reduced in the period from 15 to 30 weeks of age. This lowered food intake was associated with significantly less live-weight gain up to 22 weeks of age, and pullets reared in atmospheres with high ammonia concentrations matured up to 2 weeks later than pullets reared in ammonia-free atmospheres.

INTRODUCTION

Originally the ventilation requirements of intensive poultry houses were calculated on the basis of the provision of oxygen and the removal of carbon dioxide. While Davies (1951) suggested that carbon dioxide levels should be kept below 0·2 per cent by volume, Mitchell and Kelley (1933) had found only 0·11 per cent of carbon dioxide, and Osbaldiston and Sainsbury (1963) only 0·06 per cent of carbon dioxide in poultry houses ventilated at low rates. The importance of these two gases is self-evident, but it would appear that it is unlikely for a poultry building to be inadequately ventilated in these respects.

In recent years therefore, attention has been focused more on ventilation rates computed on the basis of temperature and humidity control. The subtraction of structural heat losses through the building materials from the sensible heat production of the stock housed therein indicates the quantity of heat available for heating and ventilating the building in cold weather when ventilation rates are at a minimum (Sainsbury, 1959; Longhouse, Ota and Ashby, 1960).

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The thermal concept of ventilation has been successfully applied for several years though it makes no provision for the control of air pollution by dust, micro-organisms and gaseous pollutants arising from the excreta and litter. Bullis, Snoeyenbos and Van Roekel (1950) associated an increase in the incidence of kerato-conjunctivitis with the swing to intensive management and the associated build-up of atmospheric ammonia concentrations. The same symptoms were produced experimentally by Carnaghan (1958). In experiments described by Valentine (1964), the growth rate of broilers was depressed by ammonia at concentrations of 60-70 p.p.m. by volume as measured by colorimetry. This level caused both kerato-conjunctivitis and tracheitis. Much of the pathological syndrome may have been due to secondary infections after primary predisposition by ammonia. Anderson, Beard and Hanson (1964) showed that chickens vaccinated against Newcastle disease were more susceptible to that infection if exposed to moderate concentrations of ammonia than those housed in ammonia-free atmospheres.

A series of studies was initiated into the effects of ammonia on respiratory turnover, since numerous authors (*e.g.* Joules, 1954; Valentine, 1964), have attributed pollutant-induced debility to respiratory effects, especially to damage of the tracheobronchial tree. In addition, the prolonged effects of graded concentrations of ammonia on the performance of chickens housed in environmental cabinets were studied, where in order to avoid the possible complications of other pollutants, the separate effects of ammonia was investigated by its introduction as the sole variable to chickens housed in otherwise standardised environments. Both Scarborough (1957) and Valentine (1964) had observed ammonia concentrations of the order of 60-70 p.p.m. under commercial conditions. It was decided however to study the effects of ammonia concentrations as high as 100 p.p.m. on poultry performance although in field observations connected with the studies reported in this paper, 70 p.p.m. of ammonia by volume was confirmed to be the maximum concentration.

MATERIALS AND METHODS

The respiration rate of chickens of various ages was studied by several methods. Attempts were made to measure the respiration rate of chickens of 10 or more days of age by placing them in a closed circuit apparatus normally used for the measurement of basal metabolic rate. The carbon dioxide produced by the chick was absorbed by sodium hydroxide, and water was run into the equilibration chamber to compensate for the oxygen consumed, so that the internal pressure (as measured by a manometer) remained constant. Ammonia was applied qualitatively by the addition of ammonium hydroxide solution. Respiration was recorded by a tambour connected to a kymograph.

Various methods were attempted for the measurement of the effects of ammonia on the respiration rate of adult stock, and these methods took into account that known ammonia concentrations could not be satisfactorily incorporated in the closed circuit system. Therefore, a pneumograph was constructed to fit around the thorax of an adult chicken. A hen wearing the pneumograph was allowed to rest for 30 min. in a darkened control cabinet without ammonia and then a series of recordings of breathing rate was taken. The same hen was then transferred to an ammonia cabinet, allowed to rest in darkness, and a further series of readings was taken. Thus in this technique each hen was used as its own control. Inducing a hen to rest quietly wearing a pneumograph was an incredibly tedious process and, in addition, data were required from hens which had become acclimatised to ammonia. It was observed that breathir

movements of a sedentary hen were proportionally reflected in the movement of the feathers around the vent. The hens used in these experiments were quiescent in blue light, therefore data on their sedentary respiration rates were obtained by the use of blue illumination within the environmental chambers. The crissal feathers of these white hens were painted black to facilitate visual perception of their movements. Observations of respiration rate were made via small apertures in the cabinet doors. Depth of respiration was estimated by the juxtaposition of a sheet of graph paper against the marked feathers so that a magnitude of the body movements due to respiration could be noted.

The influence of ammonia on respiration was also measured by indirect calorimetry. A small respiration chamber was constructed of inert materials. The chamber was essentially an airtight box approximately 0·22 m.^{3} in volume, in which two adult chickens could be placed. The chamber was ventilated at a constant low rate equivalent to 0·17 m.^{3}/adult hen/hr, and the carbon dioxide level within the chamber was measured using a direct reading carbon dioxide analyser (Shandon Scientific Company, London). A preliminary series of determinations was made without chickens in the chamber to ascertain that ammonia *per se* did not affect the analyser. Thereafter the carbon dioxide production of two White Leghorn hens housed together in the chamber in clean air was compared with that of the same hens in ammoniated air.

The effects of exposure to ammonia on blood *p*H were also determined. In order to achieve the reliability of *in vivo* measurements without the associated difficulties, a technique was evolved whereby freshly shed blood could be passed over a microelectrode system. A puncture was made in the brachial vein and the blood allowed to form a gradually expanding drop. A microelectrode system (W. G. Pye & Company, Limited, Cambridge) coupled to a *p*H meter was inserted into the drop. The readings diminished in magnitude asymptotically, and the recording in all cases was taken at the asymptote. Each bird was used as its own control, *p*H readings being taken after 15 min. in a control cabinet and again after 15 min. in a polluted cabinet (75 p.p.m. ammonia by volume) from the opposite brachial vein.

For the growth experiments a total of eleven chambers were constructed. For experiments A1 and A2, two chambers each of 120 × 65 × 85 cm. were constructed of hardboard and each was fitted with a wire floor above a droppings pit lined with removable polythene sheeting. Each chamber contained a food bowl and water trough and was constantly illuminated using one 15 watt red electric bulb. Each chamber was ventilated by one 7·62 cm. diameter 2400 r.p.m. centrifugal fan which removed 60 m.^{3}/hr of air. Incoming air was drawn from a controlled temperature building in which the chambers were contained. In the study on the effects of ammonia on the performance of broiler-type chickens (experiments A1 and A2) ten 4-week-old White Rock × Light Sussex cockerels[1] were housed in each chamber. Synthetic anhydrous ammonia (Imperial Chemical Industries Ltd, Billingham, Co. Durham) was drawn from cylinders and metered into one of the chambers via a gap flowmeter and a liquid paraffin manometer. This manometer was calibrated by chemical titrations in which ammonia was passed into known volumes of standard sulphuric acid and the time required for neutralisation, using methyl red as indicator, was recorded. In experiment A1 ammonia was added for a period of 19 days at approximately 100 p.p.m. by volume, after which time recovery was

[1] Supplied by Midland Livestock Producers Ltd., Worksop, Notts.

studied for a further 18 days in an ammonia-free atmosphere. In experiment A2 ammonia was added at approximately 50 p.p.m. by volume for the duration of a 33-day experiment. In these experiments data for food consumption, growth rate and for environmental temperature and humidity were recorded.

For experiment A3 a further nine cabinets were constructed, similar in principle to the two already described, but structurally modified to accommodate three cages each containing three hens. Each chamber was 150 × 65 × 80 cm. in size and each vertical block of three chambers was ventilated by a 10·80 cm. diameter, 2400 r.p.m. centrifugal fan, which removed 60 m.³/hr of air from each chamber. Each chamber was lit by two 15 watt red electric light bulbs controlled by a 24-hr time clock. Synthetic anhydrous ammonia was introduced into six of the chambers as previously described. In experiment A3 three 11-week-old White Leghorn pullets (W. D. Evans Ltd., Kibworth, Leics.) were housed in each of the three cages in every chamber to give three treatments each consisting of 27 pullets. The three treatments were 0, 53 and 78 p.p.m. by volume of ammonia administered from 11-18 weeks of age inclusive, thereby allowing the effects of ammonia on growing pullets, and on their subsequent laying performance to be studied. An 8-hr day was imposed to 20 weeks of age, thereafter the lighting pattern of Bowman and Jones (1963) was employed, reaching a maximum of 17 hr of light per day.

The superimposition of ammonia produced from faeces on the calculated concentrations was minimised by the daily removal of the droppings and by the absence of litter. Losses of ammonia by leakage out of the chambers were unlikely as static pressure measurements and smoke tests showed that the internal pressure in the chambers was lower than atmospheric so that all air movement was from the outside inwards. However, comparative analyses of food samples before and after 2 days in the ammonia-polluted air showed appreciable absorption of ammonia by the food. Approximately 4 per cent of the ammonia released into the chambers was found to be lost in this way and since the rate of absorption was probably as variable as the amount of food present, it was concluded that the estimated ammonia concentrations could not be regarded as absolute. In experiment A3, measurements were made of food intake, live-weight change, egg production, egg weight, internal egg quality and environmental temperature and humidity.

The diets used in experiments A1, A2 and A3 are listed in Table 1. Diet D1 was offered in experiments A1 and A2, and diet D2 was offered throughout experiment A3. In experiment A3, no separate growers diet

TABLE 1

Diets used in experiments A1, A2 and A3 (percentage inclusion rate)

	Diet D1 (used in expts. A1 & A2)	Diet D2 (used throughout expt. A3)
Wheat meal	20	—
Yellow maize meal	24	45
Milo meal	24	20
Maize gluten meal (44 per cent protein)	8	—
Soyabean meal (44 per cent protein)	12	20
White fishmeal (65 per cent protein)	5	—
Dried whey	2	—
Supplement	5[1]	5[2]
Limestone	—	5
Fat[3]	—	5
Proximate analyses		
Crude protein (per cent)	20·3	15·8
Metabolisable energy (kcal./kg.)[4]	2870	3075
Calcium (per cent)	1·2	3·2
Methionine (per cent)	0·46	0·34

[1] Described by Payne and Lewis (1963).
[2] Described by Smith and Lewis (1964).
[3] H.E.F. No. 1 (Proctor & Gamble Ltd., Newcastle-on-Tyne) and described by Lewis and Payne (1963).
[4] Calculated from the data of Lewis and Morgan (1963).

TABLE 2

The effects of ammonia on respiratory activity. The methods used are as described in the text; ammonia was at uncontrolled levels for the closed circuit method, and at 100 p.p.m. by volume for all other records. The means include figures for standard deviation

	Observation	Method	Age of stock	Number of readings per treatment	Recordings		Percentage reduction due to ammonia
					Control	In ammonia 100 p.p.m.	
Respiration rate (respirations per minute)	1	Closed circuit	10 days	7	96·9± 7·7	93·0± 4·2	4·0
	2	Closed circuit	10 days	20	71·4±13·0	63·5±10·4	11·1
	3	Pneumographic	8 months	20	54·5± 1·2	41·5± 2·6	23·9***
	4	Crissal, no acclimatisation	9 months	20	15·3± 0·5	13·7± 0·7	10·5***
	5	Crissall after 3 weeks' acclimatisation to ammonia	10 months	9	17·8± 1·6	16·6± 1·6	6·9
	6	Crissall after 4 weeks' acclimatisation to ammonia	10 months	9	19·4± 2·4	15·8± 1·7	18·4***
Respiration depth (mm. of crissal feather movement)	7	Crissall feather movement	10 months	100	6·2± 0·59	5·93± 0·77	5·57
	8	Crissall feather movement	10 months	100	5·04± 0·22	4·41± 0·46	12·50***
	9	Crissall feather movement	10 months	100	5·12± 0·16	4·37± 0·37	14·65***
Carbon dioxide concentration (per cent in respiration chamber)	10	Indirect calorimetry	11 months	35	0·78± 1·10	0·75± 0·10	3·7
	11	Indirect calorimetry	11 months	30	0·95± 0·07	0·73± 0·13	23·1*

* Significant difference $P \leqq 0.05$.
*** Significant difference $P \leqq 0.001$.

was used thereby avoiding the problem of the timing of the changeover from the growers to a layers diet.

RESULTS

Data on the effects of ammonia on respiratory activity are given in Table 2. Except for observations 1 and 2, ammonia was supplied at 100 p.p.m. by volume. In all instances the addition of ammonia reduced respiratory activity and this occurred even when adult hens had become accustomed to a polluted atmosphere.

The effects of ammonia on blood *p*H, as measured by the bleeding vein method, indicated that a slight but statistically non-significant rise in blood *p*H occurred after 15 min. exposure to 75 p.p.m. by volume of ammonia. Fifteen hens were used and prior to exposure their blood *p*H averaged 6·447 with a standard deviation of 0·470, and after exposure the blood *p*H was 6·563±0·072.

A summary of the environmental dat obtained during the growth experiments A1 and A2 is given in Table 3. The experiments were conducted at approximately 21° C. and 18° C. respectively, with relative humidities of 50 per cent. The data for food consumption, live-weight gain and efficiency of food utilisation obtained in these experiments are given in Tables 4 and 5. In experiment A1 (see Table 4) 106 p.p.m. of ammonia by volume, administered from 28 to 47 days of age, caused a reduction of voluntary food intake of 14·5 per cent. Food consumption did not return to the normal level until 12 days after the cessation of ammonia flow. Significant growth rate differences between the birds of the two treatments were due to these differences in

TABLE 3

A summary of environmental data obtained from experiments A1 and A2. Data were recorded twice daily and the means include figures of standard deviation

	Experiment A1		Experiment A2	
	Control chamber	Experimental chamber	Control chamber	Experimental chamber
Mean temperature (°C.)	20·8±3·8	20·8± 3·8	18·4±3·4	18·4± 3·5
Mean relative humidity (per cent)	50·1±8·1	50·1± 8·5	50·9±5·0	50·8± 5·2
Mean added ammonia concentration (p.p.m. by volume)	Nil	106·0±15·8	Nil	51·3±18·8

TABLE 4

Results obtained from experiment A1. Each treatment consisted of 10 individually weighed White Rock × Light Sussex cockerels; 28 days of age at the commencement of the experiment. Ammonia (106·0 p.p.m. by volume) was added for the period 28-47 days of age as described in Table 3 and in the text

Age of chicks days	Food consumption (g./bird/day)		Live-weight gains (g./bird/day)		Food conversion (g. food/g. live-weight gain)	
	Controls	Experimental	Controls	Experimental	Controls	Experimental
28-34	63·5	59·1	27·0	25·4	2·35	2·33
35-40	77·9	72·8	33·4	30·6	2·33	2·38
40-47	85·2	66·2	37·1	22·0*	2·30	3·01
48-53	92·6	71·5	33·6	20·6*	2·76	3·46
54-59	100·2	88·5	35·2	33·9	2·85	2·61
60-65	103·4	104·7	31·7	38·9	3·26	2·69

* Significant difference from the controls $P \leqq 0{\cdot}05$.

food consumption. In experiment A2 (see Table 5) the effect of 51 p.p.m. of ammonia by volume was far less marked and it was only after the 12th day of the experiment that the food consumption of the ammonia-exposed chicks fell below that of the controls. No significant differences in growth rate occurred in this experiment.

exposed to 52·6 p.p.m. ammonia and the controls was not significant, from the 15th to 38th week of age the level of voluntary food intake was always intermediate between the controls and the birds exposed to 78·2 p.p.m. ammonia. The live-weight gain figures for the control group and for the 52·6 p.p.m. ammonia treatment were very similar.

TABLE 5

Results obtained from experiment A2. Each treatment consisted of 10 individually weighed White Rock × Light Sussex cockerels: 28 days of age at the commencement of the experiment. Ammonia (51·3 p.p.m. by volume) was added for the duration of the experiment as described in Table 3 and in the text

Age of chicks days	Food consumption (g./bird/day)		Live-weight gain (g./bird/day)		Food conversion (g. food/g. live-weight gain)	
	Controls	Experimental	Controls	Experimental	Controls	Experimental
28-33	54·5	61·3	22·0	27·4	2·47	2·23
34-39	79·7	79·7	32·4	30·8	2·46	2·59
40-45	96·5	90·1	39·7	35·8	2·43	2·52
46-51	103·6	100·1	42·4	39·0	2·44	2·57
52-60	111·1	103·5	35·7	31·9	3·09	3·24

TABLE 6

A summary of environmental data obtained from experiment A3. Each treatment consisted of 27 White Leghorn pullets housed at 11 weeks of age, three birds per cage and three cages per environmental chamber. Ammonia at the levels indicated was added for the period 11-18 weeks of age inclusive. The temperature and humidity data refer to the whole of the experiment from 11-46 weeks of age. The data include figures of standard deviation

Environmental factor	No added ammonia	54 p.p.m. by volume added ammonia	78 p.p.m. by volume added ammonia
Temperature °C.	19·4±3·9	19·8± 3·7	19·6± 4·0
Relative humidity (per cent)	62·6±9·7	64·7± 7·3	64·0± 8·6
Added ammonia	Nil	52·6±19·6	78·2±27·2

A summary of the environmental data obtained during experiment A3 is given in Table 6. The experiment was conducted at approximately 20° C., with a relative humidity of approximately 64 per cent. During the period 11 to 18 weeks of age ammonia was added at either 0, 52·6 or 78·2 p.p.m. by volume. The effects of these different levels of ammonia on food consumption and on live-weight change are given in Table 7. From 15 to 30 weeks of age, food consumption of the birds treated with 78·2 p.p.m. ammonia was significantly less than the controls. Although the differences in food consumption between the birds

However, the birds exposed to 78·2 p.p.m. ammonia grew significantly more slowly during the 15 to 22 weeks of age period, and it was not until after the 38th week of age that the live-weight of the 78·2 p.p.m. ammonia treated birds attained the level of those exposed to the other two treatments.

The egg production and egg weight records obtained in experiment A3 are given in Table 8. The stock reared in the ammoniated atmospheres from 11 to 18 weeks of age came into lay later and also tended to lay slightly larger eggs. Table 9 shows that the average delay in sexual maturity was approximately 1 week for the birds treated

TABLE 7

Experiment A3. Results for food consumption and live-weight gain. Twenty-seven pullets per treatment, housed three birds per laying cage. Initial live-weight 1250 g./bird. Environmental factors as described in Table 6 and in the text

Weeks of age	Food consumption g./bird/day			Live-weight change g./bird/day			Live-weight overall gain (g./bird) from 11 weeks of age, measured at the termination of each 4-week period		
	No ammonia	52·6 p.p.m. ammonia	78·2 p.p.m. ammonia	No ammonia	52·6 p.p.m. ammonia	78·2 p.p.m. ammonia	No ammonia	52·6 p.p.m. ammonia	78·2 p.p.m. ammonia
11-14	70·2	70·9	69·2	4·89	5·82	4·60	137	163	129
15-18	67·4	64·7	60·8*	4·64	3·21	1·00**	267	253	157[1]
19-22	76·8	73·3	67·8*	8·11	8·17	6·39	494	482	336[2]
23-26	106·8	97·1	88·7**	4·71	6·07	6·68	626	652	523
27-30	125·5	119·0	111·0**	1·11	2·14	2·57	657	712	595
31-34	130·2	120·4	122·2	1·39	1·32	1·39	696	749	634
35-38	126·2	120·2	119·8	0·57	0·18	0·32	680	754	643
39-42	117·1	112·3	113·1	1·07	1·00	4·43	710	792	778
43-46	114·7	117·3	114·9	−1·50	−1·14	−1·07	668	760	748

* Significantly less than the controls $P \leqq 0{\cdot}05$.
** Significantly less than the controls $P \leqq 0{\cdot}01$.
[1] Significantly different $P \leqq 0{\cdot}01$ from other treatments.
[2] Significantly different $P \leqq 0{\cdot}05$ from other treatments.

with 52·6 p.p.m. ammonia and about 2 weeks for those treated with 78·2 p.p.m. ammonia. The differences given in Table 9 in total egg production to 322 days of age were partly a reflection of the differences in age of sexual maturity and also were due partly to the differences in mortality.

TABLE 8

Experiment A3. Results for egg production and egg weight. Twenty-seven pullets per treatment. The ammonia was supplied to the cages during the weeks 11-18 inclusive. Environmental factors as described in Table 6, and in the text

Weeks of age	Egg production—eggs per 100 hens per day			Mean egg weight g.		
	No ammonia	52·6 p.p.m.	78·3 p.p.m.	No ammonia	52·6 p.p.m.	78·3 p.p.m.
19-22	17·7	11·5	8·0	50·4	51·5	53·6
23-26	70·2	51·5*	42·2**	57·7	59·4	58·1
27-30	90·7	85·7	73·9**	60·5	61·1	61·4
31-34	90·2	88·5	83·2	61·8	62·0	62·5
35-38	90·9	86·7	83·8	61·6	63·2	62·5
39-42	87·2	82·1	83·6	63·3	63·4	63·8
43-46	83·2	81·4	84·1	64·3	65·5	65·0

* Significantly different from the controls $P \leqq 0{\cdot}05$.
** Significantly different from the controls $P \leqq 0{\cdot}01$.

TABLE 9

Experiment A3. Results for age at sexual maturity, mortality, peak egg production and total egg production per hen to 322 days of age. The ammonia levels were administered from 11-18 weeks of age inclusive. Each group of birds consisted of 27 White Leghorn pullets

Treatment	No ammonia	52·6 p.p.m. added ammonia	78·3 p.p.m. added ammonia
Age at 30 per cent production (days)	150	156*	163**
Age at 50 per cent production (days)	158	172**	177***
Age at 75 per cent production (days)	172	182**	193***
Mortality (nil prior to 133 days) 133-322 days	Nil	2	3
Peak production (no. eggs/100 hen days) on a weekly basis	93·7	90·7	87·5
Production—number of eggs/hen to 322 days per hen housed	149	130**	118***
Production—number of eggs/hen to 322 days calculated on a hen day basis	149	137**	126***

* Significantly different from the controls $P \leqq 0{\cdot}05$.
** Significantly different from the controls $P \leqq 0{\cdot}01$.
*** Significantly different from the controls $P \leqq 0{\cdot}001$.

DISCUSSION

Minimal ventilation rates in intensive poultry houses are associated with several problems of air pollution, including high ammonia concentrations. This problem has not been studied in the past. For reliable estimations of the effects of individual pollutants, they must be investigated as the sole variable in otherwise standardised environments, rather than one of several variables in experiments involving low ventilation rates.

The experiments described in this paper indicate that one fundamental cause of the

chickens including resistance to infection with Newcastle Disease virus. *Avian Dis.*, 8: 375.

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