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The development of ophthalmacrosis in the domestic chicken

Robert Allen Voitle

To the Graduate Council:

I am submitting herewith a dissertation written by Robert Allen Voitle entitled "The development of ophthalmacrosis in the domestic chicken." I have examined the final electronic copy of this dissertation for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy, with a major in Animal Science.

H.V. Shirley Jr, Major Professor

We have read this dissertation and recommend its acceptance:

O.E. Goff, R.C. Fraser, R.L. Murphee

Accepted for the Council:

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Vice Provost and Dean of the Graduate School

(Original signatures are on file with official student records.)

May 22, 1969

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and recommend its acceptance:

Ronald C. Fraser

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THE DEVELOPMENT OF OPHTHALMACROSIS IN THE DOMESTIC CHICKEN

A Dissertation
Presented to
the Graduate Council of
The University of Tennessee

In Partial Fulfillment
of the Requirements for the Degree
Doctor of Philosophy

by
Robert Allen Voitle

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ABSTRACT

A series of 12 experiments was conducted to study factors involved in the development of ophthalmacrosis in chickens.

It was found that exposure of chicks to continuous light, regardless of color or intensity, caused the eyeballs to enlarge, but more strikingly that continuous darkness caused the enlargement to an even greater extent. Breaking the day into long, short, or superimposed photoperiods resulted in less apparent eye enlargement than was found in birds exposed to a continuous lighting regime. When the birds were exposed to a diurnal light cycle the eyes appeared to be unaffected.

Chicks hatched from eggs incubated in continuous darkness appeared to have ophthalmacrosis at hatching time; however, when the chicks were subsequently placed under a diurnal light cycle they returned to normal. When these chicks were placed under a continuous lighting regime upon hatching the condition continued to worsen and permanently impaired vision resulted.

Regardless of whether the birds were placed under a continuous lighting regime from 0-8 weeks or from 8-22 weeks of age they developed ophthalmacrosis; however, the symptoms appeared more slowly in the 8-22 week period. When birds with ophthalmacrosis were switched to a diurnal light cycle from 8-22 weeks all returned to normal but with varying degrees of retinal damage. Switching birds with normal eyes to a continuous lighting regime from 8-22 weeks of age resulted in varying degrees of ophthalmacrosis and retinal damage. In all cases the degree of retinal

damage was positively related to the degree of ophthalmacrosis.

The development of ophthalmacrosis was found to be related to the accumulation of excess amounts of sodium and water in the eye.

The effects of pinealectomy, estrogen implantation, and the feeding of thiouracil and protamone on the development of ophthalmacrosis were studied. The relationships of pituitary, bursa, and thyroid weights to ophthalmacrosis were determined.

A hypothesis, involving the hypothalmic-pituitary-adrenal complex, to explain the development of ophthalmacrosis is presented.

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I. INTRODUCTION

In 1956 Shirley reported a peculiar eyeball enlargement in chickens reared on The University of Tennessee Poultry Farm that seemed to be restricted to those birds exposed to continuous light. The hypothesis that light was a factor in the development of this abnormality was supported in a paper by Jensen and Matson the following year. Since that time a few reports have appeared in the literature which also indicate that the exposure of chickens to continuous light will cause their eyeballs to enlarge (Lauber et al., 1961; Shutze et al., 1961; McCluskey and Arscott, 1967).

In order to differentiate this specific type of eye enlargement from others of a similar nature, the term ophthalmacrosis will be used to refer only to the type of eye enlargement that is characterized by the following: reduction in the depth of the anterior chamber, flattening of the cornea, increase in the size of the posterior chamber and a peculiar "slant eye" appearance of the individual. This condition had previously been referred to as buphthalmia; however, since this latter term is generally reserved for use in relation to the disease of congenital glaucoma, it does not seem to be the most appropriate.

It is important to establish the etiology of ophthalmacrosis both from the standpoint of the environmental factors responsible as well as the basic physiological changes which are manifested in enlarged eyes. If ophthalmacrosis in chickens is related to the disease of glaucoma in humans, the results of studies conducted on this problem might lead to improvements in the treatment of this disease.

Objectives

The primary objectives of these studies were to establish the relationships between exposure to various photoperiods, intensities of light and ophthalmacrosis, a peculiar eye enlargement in poultry, in addition to determining the basic mechanism involved in the development of this abnormal eye condition.

II. REVIEW OF LITERATURE

Sturkie (1965) pointed out that the avian eye has all of the structural components of the reptilian eye and primarily differs from the mamalian eye in that it has a pecten and relatively fewer rods. These components include the eyeball, optic nerve, and such accessory structures as eyelids, conjunctiva, lachrymal glands, and ocular muscles. The avian eyeball consists of the cornea, lens, the anterior chamber containing the aqueous humor, the posterior chamber containing the vitreous body, the iris, pupil, ciliary body, pecten, sclera, choroid and retina with its connection to the optic nerve.

Adler (1950) stated that the aqueous humor, contains sugars and amino acids, transports oxygen and carries away waste products such as carbon dioxide. This fluid arises in large part behind the iris in the ciliary body, fills the posterior chamber, flows between the iris and lens into the anterior chamber and leaves the eye at the canal of Schlemm.

White *et al*., (1959) stated that the aqueous humor was responsible for maintaining the intraocular pressure desirable for optical purposes in addition to providing nutrients to the avascular cornea and lens. Adler (1950) described a normal diurnal rhythm in the intraocular pressure with a low at five to seven in the evening, rising during the night and reaching a peak in the early morning hours.

Adler (1950) also found that placing an individual with a narrow filtration angle in the dark for one hour may increase the intraocular pressure by preventing fluid outflow as a result of the blockage of the canal of Schlemm with the dilated pupil. Adler further reported that the overstretching of the corneal stroma causes irregular refraction and reflection in addition to making the cornea less transparent. There is at the same time a tendency for the cornea to become edematous, which increases the cloudiness. All of these factors interfere with normal vision.

Coulombre (1956) demonstrated that intraocular pressure is of prime importance in the normal increase of eye size in the developing chick embryo.

Langham (1958) suggested that the fundamental process of secretion of the aqueous humor may be the result of an exchange of hydrogen ions derived from the cells for the sodium ions from the blood with carbonic anhydrase functioning to facilitate neutralization of the residual hydroxyl ion in the cell by hydration of carbon dioxide. This explains in part the phenomenon of reduction of the rate of aqueous humor formation with the administration of a carbonic anhydrase inhibitor such as Diamox (acetazolamide). Seaman and Storm (1963) reporting on the effects of intravenous injections of Diamox reported a 60 percent drop in intraocular pressure within 60-90 minutes in year old chickens, although some birds were found to be refractory to this inhibitor. Becker (1954) also reported a drop in intraocular pressure 60-90 minutes after administration of Diamox. He further stated it reached a minimum in three to five hours and returned to the control level in eight to twelve hours. He concluded from the data that the decreased pressure was due to a decrease in the rate of production fo aqueous humor. In a subsequent paper Becker and Constant (1955) reaffirmed this hypothesis by reporting that Diamox appeared to lower intraocular pressure by directly inhibiting the secretion of aqueous humor from the ciliary body.

It is accepted that the proper balance of pH, salts and endocrines in the body fluids are of prime importance in the maintenance of normal intraocular pressure and ultimately eye shape. Any factor disturbing this balance is capable of producing a pathological condition. White et al. (1959) reported that increased aqueous humor secretion raises the intraocular pressure giving rise to glaucoma, a disease of the human eye. Duke-Elder (1941) stated that glaucoma is characterized by an increase in intraocular pressure and concluded that this condition could be aggravated by "stress."

According to Bunning (1964) humans and animals are not able to adjust physiologically when light and dark periods deviate too much from a normal diurnal cycle. This places the individual in a state of general physiological stress. Selye (1951) reported that animals respond in a typical manner to any general systemic stress. Stress causes the release of adrenocorticotropic hormone (ACTH) from the anterior pituitary. The adrenocorticotropic hormone then moves through the general circulation and acts on the adrenal cortex to produce predominantly gluco-corticoids and to a lesser extent mineralocorticoids which enter the general circulation and ultimately effect their specific target organs. This response is accompanied by a release of the adrenal medullary hormones; however, this latter process is independent of the secretion of the adrenocorticotropic hormone.

The most outstanding manifestations of the general-adaptation-syndrome (stress response) were outlined by Selye (1951) as: adreno-cortical hypertrophy, thymico-lymphatic involution, eosin-openia, lymphopenia, polynucleosis and gastrointestinal ulcers. Bernhard (1966) reporting on the human stated that the adrenal cortical hormones; control kidney function, promote retention of sodium and chloride ions and the excretion of potassium ion, regulates fat, protein and carbohydrate metabolism and affects the sex organs. They have an additional effect on the membranes in the joints, the eyes and the skin as a consequence of cortical control over water, ion and carbohydrate metabolism. He also stated that the secretion of these hormones normally follow a circadia rhythm.

Clayton _et al_. (1957) indicated the presence of an additional factor in the hypothalamic region of the brain that triggers the release of the adrenocorticotropic hormone to set this entire chain of events in motion. McCann and Haberland (1959) referred to this factor as corticotropin-releasing-factor (CRF). These reports are supported by the finding of

Brizzee and Eik-Nes (1961) in the dog and by isolation and partial purification of a "corticotropin influencing factor" (CIF) from human spinal fluid by Eik-Nes et al. (1961).

The importance of this general-adaptation-syndrome (stress response) to eye enlargement was indicated in papers by Armaly (1963), Becker and Mills (1963), and Nichols (1964) who found that topically administered adrenocortcosteroids cause an increase in intraocular pressure. This increase in pressure on the growing eye could theoretically be the ultimate cause of the enlargement.

Ophthalmacrosis or eye enlargement in poultry in response to photoperiod modification was first reported in a paper by Jensen and Matson (1957). They found that the eyes of birds subjected to continuous light were 38 percent greater in weight than those of birds exposed to only 12 hours of artificial light per day. This increase in weight was attributed to increased fluid content in the vitreous body. Shutze et al. (1961) reported that 60 percent of the pullets grown under continuous light showed signs of blindness or bulging of the eye which they also attributed to accumulation of fluid in the posterior chamber of the eye. One third of these pullets were found to be blind in one or both eyes. The iris was damaged and the fundus of the eye could be readily observed without the aid of instruments. The affected birds moved cautiously and were extremely docile. The birds exposed to continuous light from 0-8 weeks or from 8-20 weeks showed moderate signs of the condition at 20 weeks, but they returned to normal appearance after being placed on a diurnal light cycle in the laying house. Lauber et al. (1961) found that chicks grown under continuous light developed elongated eyelids or a peculiar "slant-eye" condition. The cornea appeared to be less convex than those birds exposed to a diurnal light cycle with the iris more closely associated with the inner surface of the cornea. Upon removal and disection of the eye it was evident that there was a flattening of the cornea and a reduction in the thickness of the eye wall. The most striking difference was in the size of the vitreous body. Histological sections showed a distinct difference in the thickness of the eye wall. The enlarged eye showed a thinner choroid with flattened and elongated vessels as well as a much thinner retina with the layer of rods and cones appearing much reduced while the pigment layer and the inner nuclear layer appeared to be slightly thicker. No differences were noted in the optic nerve. Lillie and Denton (1965) have reported similar findings. McCluskey and Arscott (1967) found this particular enlarged eye condition in chicks continuously exposed to red infrared, white infrared or incandescent light.

Hogg (1967) found that exposure of chickens to a continuous lighting regime resulted in the development of enlarged eyes. The administration of Diamox reduced the severity of this condition while corticosterone injection failed to appreciably affect eye size. It was further reported that adrenal function was positively related to the degree of ophthalmacrosis, and to the sodium and potassium concentrations of the ocular fluid as well as the osmotic pressure of the ocular fluid, while adrenal weight was found to be negatively related. Thyroid weights were greater in those birds suffering from ophthalmacrosis and histological sections of this tissue revealed signs of hyperplasia, squamous cells and colloid deficiency.

III. PROCEDURE

General Procedure

One-day-old male Leghorn chicks were randomized, wing banded and vaccinated with a combination Newcastle-infectious bronchitis vaccine. Groups of chicks were then randomly assigned to windowless insulated and fan-ventilated rooms each of which was approximately five feet square. Each room contained a single waterer and feeder and approximately four inches of wood shavings as litter. The ventilating fans were operated for two out of every ten minutes with an overriding thermostat set at 80° F. The brooder facilities were such that no light was emitted from them and the rooms were sealed so that no outside light could enter. Feed and water were kept before the birds at all times. Incandescent bulbs were used to furnish the light in all of the experiments. The formula for the diet used in all experiments is given in Table I.

The one notable exception to these procedures was in the tenth experiment of this series which dealt only with the effects of light upon chicks during incubation.

The lighting regimes in Experiments III, V and VIII are characterized with a standard notation i.e., 12L-12D; under this system of notation the number preceding the L indicates the hours of light within the photoperiod while the number preceding the D indicates the hours of darkness within the

TABLE I

COMPOSITION OF CHICK DIET[1]

Ingredient	Percent
Yellow corn	63.80
Alfalfa meal (17%)	2.50
Fish meal	2.50
Soybean oil meal (50%)	25.50
Ground limestone	0.60
Defluorinated rock phosphate	1.50
Salt	0.48
Manganese sulfate	0.02
Vitamin mix[2]	0.60
Coccidiostat premix[3]	2.50
Total	100.00

[1]Calculated analysis: crude protein, 21.54 percent, productive energy, 943 calories per pound, C/P ratio, 43.8; Methionine, 0.408 percent; Cystine, 0.313 percent; Calcium, 0.960 percent; Phosphorus, 0.692 percent; Manganese, 31.2 mg./lb.

[2]Calculated analysis: Vitamin A, 5349 I.U./lb.; Vitamin D, 340 I.C.U./lb.; Riboflavin, 3.01 mg./lb.; Niacin, 27.78 mg./lb.; Panthotenic acid, 6.67 mg./lb.; Choline, 718.0 mg./lb.

[3]2.5 pounds of premix supplies 0.013 lbs. of amprolium.

photoperiod. In experiments III and V a modification of this notation is used to characterize the superimposed photoperiods i.e., $\frac{\text{12L-12D, 60 watts}}{\text{24L-OD, } 7\frac{1}{2} \text{ watts}}$; under this system of notation the continuous light cycle (24L-OD) is placed as the denominator along with the intensity of the light source ($7\frac{1}{2}$ watts) while the diurnal light cycle is placed as the numerator along with the intensity of that light source (60 watts), to indicate it is the cycle that has been superimposed.

All data except that from Experiment IX were statistically analyzed and tested at the five percent level of probability. Significant differences between treatment means were separated by applying Duncan's Multiple Range Test.

Experiment I

This design was formulated to determine the effects of the selected physical factors of light on the development of ophthalmacrosis in the domestic fowl: hours of exposure to artificial light (0, 12 and 24 hours of light per 24 hours); various light intensities (0, 0.2, 1.6, 3.4, 3.6, 4.0, and 6.4 ft.-candles); and different wave lengths as produced by the use of blue, white and red light from 100 watt bulbs. Thirty birds were randomly assigned to each room and each treatment was duplicated. The birds were subjected to these treatments beginning at one day of age.

Five birds were sacrificed from each pen (ten birds per treatment) every two weeks for a period of ten weeks. Measurements were taken on body weights and eyeball weights at each of these five periods. An index of enlargement was calculated by expressing eye weight as percent of body weight. Beginning at four weeks of age the eyes of each bird were visually scored based on the degree of flattening of the cornea, reduction of the depth of the anterior chamber, and appearance of the fundus. Each bird was classified as having enlarged eyes or normal eyes while the score of the group was reported as the percentage of those birds with enlarged eyes. At four, six and eight weeks of age, measurements on testicular weight were taken while data on comb weights were reported at four and six weeks of age.

In the groups exposed to the three basic light treatments (0, 12, and 24 hours of light per 24 hours at an intensity of 4.0 ft.-candles) additional data were taken on the following: adrenal weights at eight weeks of age; pituitary and pineal weights at ten weeks of age.

The birds receiving 12 hours of light per day (a diurnal cycle) at an intensity of 4.0 ft.-candles were considered to be the controls.

Experiment II

This experiment was designed after it was suspected that the right eye of some birds might be larger than the left eye. It was hypothesized that since chicks are usually vaccinated in the right eye at this station that the vaccination process might have some effect on eye size.

Twelve-day-old male Leghorn chicks were started under each of the three basic lighting regimes (0, 12 and 24 hours of light per 24 hours at an intensity of 4.0 ft.-candles). Half of the chicks in each of these groups were vaccinated in the right eye and half were vaccinated in the left eye before housing. At eight weeks of age ten birds from each pen were sacrificed and weights of both the right and left eyes were taken. An analysis of variance was run to determine if any differences existed between right and left eye and between vaccinated and unvaccinated eyes.

In addition measurements on body weight, percentage of birds with crooked toes and with eye enlargements were recorded and an index calculated expressing the weight of the right eye as precent of body weight.

Experiment III

It was obvious from the first experiment that although exposure to continuous light would induce the development of ophthalmacrosis, exposure to continuous darkness resulted in an even greater enlargement. It was hypothesized from this information that exposure to light <u>per se</u> was not the cause and perhaps the development of the condition was tied to the lack of a diurnal light cycle.

Fifteen day-old male Leghorn chicks were started under each of three circadian light cycles. One group was exposed to a long photoperiod consisting of 22-1/2 hours of light per day. A second group was exposed to a short photoperiod consisting of 1-1/2 hours of light per day. The final group was exposed to a superimposed photoperiod which consisted of a continuous dim source of light (0.2 ft.-candle) which had previously been shown to induce ophthalmacrosis and a relatively bright source of light (4.0 ft.-candle) that was on for 12 of every 24 hours. This latter lighting program resulted in a continuous lighting regime that was broken into 12 hours of relatively bright light and 12 hours of relatively dim light every day, a modulating light cycle.

Ten birds from each treatment were sacrificed at eight weeks of age and data collected on body weight, eye weight, incidence of birds suffering from crooked toes and ophthalmacrosis and an index was calculated to express eye weight as percent of body weight.

Experiment IV

This experiment was designed to determine if the administration of a female hormone, castration, or pinealectomy might effect the development of ophthalmacrosis.

Pinealectomy was included for three primary reasons: first that Pincus (1956) had indicated that pinealectomized animals secrete nearly twice as much aldosterone as the controls and it was hypothesized that this latter hormone might be related to the development of ophthalmacrosis; secondly, that workers such as Danowski (1962) and Gittes and Chu (1956) had linked the pineal to the secretion of the sex hormones; finally, that the secretion of the pineal was influenced by the presence or absence of light.

The experimental groups and their treatments were as follows:

White Leghorn males were pinealectomized at one week of age and 14 were placed in each of six rooms and subsequently exposed to either continuous light, continuous dark or a diurnal light cycle with each light treatment being duplicated.

White Leghorn males were castrated at one week of age and 17 placed in each of six rooms and subsequently exposed to either continuous light, continuous dark or a diurnal light cycle with each light treatment being duplicated.

White Leghorn males were implanted with a 2 mg. estrogen pellet and 12 were placed in each of six rooms and subsequently exposed to either continuous light, continuous dark or a diurnal light cycle with each light treatment being duplicated.

Seventeen White Leghorn males were hypophysectomized at one week of age, however, all but a few died before the end of the experiment and no data was collected from these birds.

At eight weeks of age seven birds were sacrificed from each pen (14 per treatment) and measurements taken on body weight, eyeball weight, incidence of birds suffering from crooked toes and ophthalmacrosis. An index was calculated to express eye weight as percent of body weight.

Experiment V

This experiment was designed to determine the effects of the following treatments on the development of ophthalmacrosis: continuous darkness; continuous light; diurnal control (12 hours of light per 24 hours); a diurnal superimposed light cycle consisting of a continuous dim source of light (0.2 ft.-candles) and relatively bright source of light (4.0 ft.-candles) that was on for 12 of every 24 hours resulting in a continuous lighting regime that was broken into a modulating cycle of 12 hours of relatively bright light and 12 hours of relatively dim light every day; a diurnal superimposed light cycle consisting a continuous bright source of light (4.0 ft.-candles) and a relatively dim source of light (0.2 ft.-candles) that was on for 12 of every 24 hours resulting in a continuous lighting regime that was broken into a modulating cycle of 12 hours of relatively bright light and 12 hours of slightly brighter light; a long photoperiod (22-1/2 hours of light per day); a short photoperiod (1-1/2 hours of light per day); a dim diurnal light cycle (0.2 ft.-candles of light 12 hours per day).

Ten birds per treatment were sacrificed at four weeks of age and measurements taken on body weight, eyeball weight, incidence of birds suffering from ophthalmacrosis. An index was calculated to express eye weight as percent of body weight.

Experiment VI

An experiment was designed to determine the effects of the following light treatments and diets on the development of ophthalmacrosis: continuous dark; continuous light; diurnal light control; diurnal light, three percent added salt; diurnal light, riboflavin deficient; continuous light, plus protamone (iodinated casein) fed in the diet at the rate of 10 grams per 100 pounds of feed; continuous light, plus thiouracil fed at the level of 0.1 percent of the diet. The same basal diet was used for all treatment groups with modifications made where required.

Ten birds per treatment were sacrificed at four weeks of age and measurements taken on body weight, eyeball weight, incidence of birds suffering from ophthalmacrosis. An index was calculated to express eye weight as percent of body weight.

Experiment VII

An experiment was designed to determine the effects of light treatments on the development of ophthalmacrosis and on sodium levels of the blood plasma and the eyeball contents. The birds were exposed to continuous light, continuous dark or a diurnal light cycle from one day of age to the termination of the experiment. At four weeks of age 55 birds per light treatment were given radioactive sodium intraperitoneally. Five birds per treatment were examined and sacrificed at 30 minute intervals up to 5 hours after the injection while others were sacrificed at 24 hours after the injection. Data were taken on body weight, eyeball weight and incidence of ophthalmacrosis. An index expressing eye weight as percent of body weight was also calculated.

The birds sacrificed at one half hour and one hour after injection received the isotope 24 hours prior to that of the other groups and the sodium sample used was of higher specific activity than that prepared for the remaining groups. This accounts for the higher counts obtained for these two time intervals.

The means of body weight, eyeball weight and index were separated by Duncan's Multiple Range Test while those dealing with sodium uptake were compared after calculation of a standard error of the mean.

Experiment VIII

This experiment was designed to determine the effect of continuous light, continuous darkness and a diurnal light cycle with reserpine (1 ppm. of diet) and without reserpine. In addition, the effect of a long photoperiod (22-1/2 hours of light per 24 hours) and a short photoperiod (1-1/2 hours of light per 24 hours) without the influence of reserpine was investigated to supplement data previously gathered.

Ten birds per treatment were visually scored at eight weeks of age for incidence of ophthalmacrosis. Measurements were taken on body weight, eyeball weight and adrenal weight. In addition an index was calculated to express eye weight as percent of body weight.

Experiment IX

An experiment was designed to determine the effects of exposure of birds to a continuous lighting regime (continuous light or dark) or a diurnal light cycle. At eight weeks of age 12 of the birds from each treatment were equally redistributed among the three lighting regimes. The birds were then maintained under these new conditions for an additional 14 weeks, that is, until they were 22 weeks of age. The birds were visually examined for ophthalmacrosis at 8, 13, 18 and 22 weeks of age.

At 22 weeks of age the remaining birds were sacrificed and measurements taken on body weight, eyeball weight and adrenal weight. An index was calculated to express eye weight as percent of body weight. Birds were visually scored for the incidence of ophthalmacrosis and retinal tears.

No statistical study of data was made as excess mortality, particularly in the group previously exposed to continuous dark, resulted in extreme variation in numbers of birds remaining. Only two birds per treatment were sacrificed in the group previously exposed to continuous darkness and subsequently exposed to either continuous light or a diurnal light cycle. The group switched from continuous dark to continuous light yielded only three birds for sacrifice. The birds switched from continuous light or a diurnal light cycle to continuous darkness yielded only four birds per treatment for sacrifice while in the remaining groups seven birds were sacrificed per treatment.

Experiment X

It was noted during the routine of banding, dubbing and vaccinating chicks for numerous experiments that "normal" baby chicks as routinely hatched from incubators seemed to have little or no anterior chamber to the eye. The depth of the anterior chamber was one of the criteria by which birds were judged to be suffering from ophthalmacrosis.

It was hypothesized that chicks begin to suffer from ophthalmacrosis during development in incubators and that the manifestation of ophthalmacrosis in growing birds is merely a continuation of the development of this condition.

An experiment was designed to test this hypothesis in which one tray of eggs was set in each of three Jamesways incubators. The first and second incubators had a light suspended approximately 18 inches above the tray of eggs. In the first incubator the light operated continuously and the second incubator the light operated diurnally (12 hours of light per 24 hours). The third incubator had no light and served as the control.

Ten birds per treatment were sacrificed at one day of age and measurements were taken on body weight and eye weight and an index was calculated.

Experiment XI

This experiment was designed to determine if the pineal gland might be involved in the development of ophthalmacrosis in birds exposed to continuous light, continuous dark or a diurnal light cycle.

Day old chicks were divided into three groups. The first group was banded and set aside as unoperated controls. The second group was sham operated and banded and the third group was pinealectomized. The birds were then equally divided into six groups and subsequently placed in continuous light, continuous dark or a diurnal light cycle with each light treatment being duplicated. At eight weeks of age four birds from each treatment were sacrificed and measurements taken on body weight, eyeball weight, thyroid weight, adrenal weight, testis weight, bursa weight and visually scored for incidence of ophthalmacrosis and retinal tears. An index was calculated to express eye weight as percent of body weight.

Experiment XII

This experiment was designed to determine if the pineal gland is involved in the development of ophthalmacrosis in birds exposed to continuous light, continuous dark or a diurnal light cycle to an age approaching sexual maturity.

Day old chicks were divided into three groups. The first group was banded and set aside as unoperated controls. The second group was sham operated and banded while the third group was pinealectomized. The birds were then divided equally between three groups and subsequently placed in continuous light, continuous dark, or a diurnal light cycle. At 22 weeks of age four birds from each treatment were sacrificed and measurements taken on body weight, eyeball weight, thyroid weight, adrenal weight, testis weight, comb weight, and bursa weight. All birds were visually scored for incidence of ophthalmacrosis and retinal tears. An index was calculated to express eye weight as percent of body weight.

IV. RESULTS

Experiment I

The data on body weight are reported in Table II.

At two weeks of age the body weights of those birds exposed to continuous darkness were not significantly different from those of the controls which were the smallest in this test. By eight weeks of age these birds were the heaviest in the test and by ten weeks of age were significantly heavier than of the other birds with the exception of those birds receiving a very dim (7-1/2 watt bulbs - 0.2 fr.-candle) continuous source of light.

The mean weight of the control birds was consistently less than the weight of the other birds from the beginning of the measurements through eight weeks of age. At ten weeks of age the control birds were only about 80 grams heavier per chick than the smallest group in the test which were those exposed to continuous red light at a rather high intensity (100 watt red bulbs - 3.6 ft. candles).

Among the groups of birds exposed to continuous light differences in intensity and wave length of light resulted in no significant differences in body weight with the exceptions that the birds exposed to red light tended to be smaller while those exposed to the dimmest source of light (7-1/2 watt bulb - 0.2 ft.-candles) were consistently larger.

The data on absolute eye weight are reported in Table III.

TABLE II

THE EFFECT OF PHOTOPERIODICITY, LIGHT INTENSITY, AND COLOR ON BODY WEIGHTS OF CHICKS
EXPERIMENT I

Treatments			Weeks of age				
Watts	Ft.-candles	Color	2	4	6	8	10
			Average body weights (gms.)[1]				
0	0	--	119[ab]	307[bc]	579[a]	816[b]	1113[d]
7½	0.2	White	135[c]	330[c]	610[a]	815[b]	1088[d]
25	1.5	White	125[abc]	330[c]	586[a]	727[ab]	1037[c]
60C[2]	4.0	White	111[a]	261[a]	498[a]	705[a]	981[b]
60	4.0	White	132[bc]	315[c]	566[a]	794[ab]	1022[c]
100	6.4	White	127[bc]	312[c]	550[a]	739[ab]	1009[bc]
100	3.4	Blue	130[bc]	299[bc]	534[a]	814[b]	1040[c]
100	3.6	Red	121[abc]	275[ab]	514[a]	735[ab]	903[a]

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]The control group (60C) received alternating periods of 12 hours of light and 12 hours of darkness per 24 hour day. All other groups received continuous treatments.

TABLE III

THE EFFECT OF PHOTOPERIODICITY, LIGHT INTENSITY, AND COLOR ON EYE WEIGHTS OF CHICKS
EXPERIMENT I

Treatments			Weeks of age				
Watts	Ft.-candles	Color	2	4	6	8	10
			Average eye weights (mgs.)[1]				
0	0	--	846^{d}	1441^{d}	1965^{c}	2728^{c}	3212^{d}
7½	0.2	White	820^{cd}	1280^{c}	1763^{b}	2231^{b}	2779^{c}
25	1.5	White	788^{bc}	1696^{b}	1696^{b}	2232^{b}	2415^{b}
60C[2]	4.0	White	705^{a}	1022^{a}	1355^{a}	1617^{a}	1890^{a}
60	4.0	White	762^{b}	1168^{b}	1610^{b}	2173^{b}	2534^{b}
100	6.4	White	756^{b}	1168^{b}	1741^{b}	1998^{b}	2436^{b}
100	3.4	Blue	759^{b}	1256^{bc}	1691^{b}	2251^{b}	2554^{bc}
100	3.6	Red	792^{bc}	1218^{b}	1676^{b}	2232^{b}	2299^{b}

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]The control group (60C) received alternating periods of 12 hours of light and 12 hours of darkness per 24 hour day. All other groups received continuous treatments.

Regardless of the age of the birds, the eyeballs taken from the control birds (alternating periods of 12 hours of light and 12 hours of darkness) were significantly smaller than those removed from birds exposed to any of the other tratments. From four weeks of age through ten weeks of age the eyes removed from the birds reared in continuous darkness were significantly larger than those of birds exposed to the other light treatments and at two weeks of age matched only by the weight of the eyes removed from chicks exposed to the very dim continuous light source (7-1/2 watt bulbs, 0.2 ft.-candles). The only other detectable trend was that the eyes of birds subjected to 0.2 ft.-candles of light tended to be heavier than those exposed to greater intensities of continuous light.

The data on the calculated eye weight index (eye weight expressed as percent of body weight) are presented in Table IV.

It is obvious from these data that there is a steady decline in index value as the birds get older. This is an important consideration in that this reduction is primarily due to an increase in weight of the bird.

The index for those birds grown in continuous darkness was consistently greater than that of any of the other treatments regardless of age. From eight weeks of age on all of the treatments resulted in a significant increase in index value over that of the controls. Although the index value of the controls was significantly greater than several of the treatments at two weeks of age, by four weeks of age they were no different statistically from the smallest index figures, and by six weeks

TABLE IV

THE EFFECT OF PHOTOPERIODICITY, LIGHT INTENSITY, AND COLOR ON EYE WEIGHT INDEX VALUES OF CHICKS
EXPERIMENT I

Treatments			Weeks of age				
Watts	Ft.-candles	Color	2	4	6	8	10
			Average eye weight index values[1,3]				
0	0	--	.71[c]	.48[bc]	.35[b]	.33[d]	.29[c]
7½	0.2	White	.61[ab]	.39[a]	.29[ab]	.27[b]	.26[b]
25	1.5	White	.64[b]	.38[a]	.29[ab]	.33[d]	.24[b]
60C[2]	4.0	White	.65[b]	.40[ab]	.28[a]	.23[a]	.19[a]
60	4.0	White	.57[a]	.38[a]	.31[ab]	.27[b]	.24[b]
100	6.4	White	.60[ab]	.39[a]	.30[ab]	.31[c]	.25[b]
100	3.4	Blue	.59[ab]	.41[ab]	.33[b]	.28[b]	.25[b]
100	3.6	Red	.66[b]	.44[bc]	.33[b]	.31[c]	.28[c]

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]The control group (60C) received alternating periods of 12 hours of light and 12 hours of darkness per 24 hours day. All other groups received continuous treatments.

[3]Eye weight/body weight x 100.

of age had become the smallest.

The summary of visual score of eye enlargement and incidence of crooked toes is reported as percent of birds affected with the condition in Table V.

It is obvious from the visual scores that a significant amount of enlargement has taken place in all treatments except for the controls and by eight weeks of age it is a 100-0 relationship. The incidence of birds with crooked toes closely followed the visual score.

The data on adrenal, pituitary, and pineal weights are summarized in Table VI.

Although adrenal weights do not differ significantly in this experiment there was a tendency for those of birds exposed to a continuous unaltering lighting regime (continuous light or darkness) to be larger than the controls with those of birds exposed to continuous light being the largest.

There were significant differences between the controls and all treatments with regard to pituitary weights with these weights apparently being proportional to the amount of light received.

The pineal weights tended to be the greatest in the control birds followed by those birds exposed to continuous light; however, no statistical differences were established.

The data obtained on testis and comb weights are presented in Table VII.

According to the analysis no significant differences could

TABLE V

THE EFFECT OF PHOTOPERIODICITY, LIGHT INTENSITY, AND COLOR ON THE INCIDENCE OF OPHTHALMACROSIS AND CROOKED TOES EXPERIMENT I

Treatments			Weeks of age					
Watts	Ft.-candles	Color	4	6	8	8	10	10
			Percentage of birds affected[2,3]					
			Eyes	Eyes	Toes	Eyes	Toes	Eyes
0	0	--	100	100	100	100	90	100
7½	0.2	White	100	100	20	100	70	100
25	1.5	White	100	100	70	100	100	100
60C[1]	4.0	White	20	10	0	0	0	0
60	4.0	White	90	100	90	100	90	100
100	6.4	White	70	100	70	100	60	100
100	3.4	Blue	90	100	80	100	90	100
100	3.6	Red	100	90	70	100	60	100

[1]The control group (60C) received alternating periods of 12 hours of light and 12 hours of darkness per 24 hour day. All other groups received continuous treatments.

[2]Each bird was judged to have ophthalmacrosis if the depth of the anterior chamber of the eye was greatly reduced.

[3]Crooked toes as described in H. E. Biester, and L. H. Schwarte, Diseases of Poultry, Fifth edition, Ames: The Iowa State University Press, 1965.

TABLE VI

THE EFFECT OF PHOTOPERIOD ON ADRENAL WEIGHT, PITUITARY WEIGHT, AND PINEAL WEIGHT EXPERIMENT I

Hours of light/day	Average weight (mgs.)[1]		
	Adrenal[2]	Pituitary[3]	Pineal[3]
0	62.7[a]	60.3[a]	33.6[a]
24	65.9[a]	79.0[c]	36.6[a]
12	54.0[a]	68.9[b]	38.2[a]

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]Organ weights taken at eight weeks of age.

[3]Organ weights taken at ten weeks of age.

TABLE VII

THE EFFECT OF PHOTOPERIOD, LIGHT INTENSITY, AND COLOR ON TESTICULAR AND COMB WEIGHTS OF CHICKS EXPERIMENT I

Treatments			Average weight (mgs.)[1]				
Watts	Ft.-candles	Color	Testis[3]	Testis[4]	Testis[5]	Comb[3]	Comb[4]
0	0	--	74[a]	167[a]	277[a]	800[a]	3158[a]
7½	0.2	White	96[a]	206[b]	1073[a]	1122[a]	3823[a]
25	1.5	White	87[a]	458[c]	831[a]	799[a]	5646[a]
60C[2]	4.0	White	74[a]	174[a]	809[a]	931[a]	3391[a]
60	4.0	White	78[a]	312[d]	875[a]	796[a]	4420[a]
100	6.4	White	83[a]	241[c]	1223[a]	937[a]	3424[a]
100	3.4	Blue	98[a]	178[a]	900[a]	1316[a]	2581[a]
100	3.6	Red	94[a]	217[b]	767[a]	1144[a]	3422[a]

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]The control group (60C) received alternating periods of 12 hours of light and 12 hours of darkness per 24 hour day. All other groups received continuous treatments.

[3]Organ weights taken at four weeks of age.

[4]Organ weights taken at six weeks of age.

[5]Organ weights taken at eight weeks of age.

be established between the various treatments with regard to either testis or comb weights. The only exceptions were for testis weights at six weeks of age where weights seemed to increase according to the quantity and quality of light. The differences in comb weight at six weeks were approaching significance.

Experiment II

The comparisons between vaccinated and unvaccinated eyes and between right and left eyes are reported in Table VIII.

Neither one of the comparisons proved to be statistically significant. In comparing the overall means, the right eye tended to be slightly larger than the left, but in comparing individual birds, one-half of the individuals had right eyes that were larger, and one-half had left eyes that were larger, indicating no difference. Even in light of these statistics it was decided to always remove the right eye for weight measurements in subsequent experiments.

The data on body weight, right eye weight, index, incidence of crooked toes and enlarged eyes are reported in Table IX.

The individuals grown in continuous darkness were significantly heavier than those grown under continuous light or the diurnal light cycle. There was no statistical difference between the latter two groups in body weight.

The means of eye weights between all three groups were significantly different from each other, with the eyes of birds

TABLE VIII

COMPARISONS OF EYE SIZE BETWEEN INTRAOCULARLY VACCINATED AND UNVACCINATED CHICKS AND BETWEEN THE LEFT AND RIGHT EYEBALL OF INDIVIDUAL CHICKS
EXPERIMENT II

Treatment	Average eye weight (gms.)[1,2]
Vaccinated	2030
Unvaccinated	2027
Right eyeball	2041
Left eyeball	2016

[1]Differences were tested by analysis of variance and were found to be non-significant ($P \leq 0.05$).

[2]Eye weights taken at eight weeks of age.

TABLE IX

THE EFFECT OF PHOTOPERIOD ON EIGHT WEEK BODY WEIGHT, EYEBALL WEIGHT, INCIDENCE OF OPHTHALMACROSIS AND INCIDENCE OF CROOKED TOES OF CHICKS
EXPERIMENT II

Hours of light/day	Average weight (gms.)[1]			Percentage of birds affected	
	Body	Eyeball	Index[1,2]	Toes	Eyes
0	799[b]	2.409[c]	.30[b]	100	100
24	626[a]	1.880[b]	.31[b]	10	100
12	655[a]	1.634[a]	.25[a]	30	0

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

exposed to continuous darkness being largest followed by those of birds exposed to continuous light. When eye weight was expressed as percent of body weight (index) those birds exposed to a continuous lighting regime (continuous light or dark) had eyes that were significantly heavier than those of the control group.

It is obvious that the birds exposed to continuous darkness had a much higher incidence of crooked toes than the other two groups which were similar in this respect.

All of the birds exposed to a continuous lighting regime (continuous light or dark) suffered from ophthalmacrosis while none of the control birds exhibited this condition.

Experiment III

The data on body weight, eye weight, and the incidence of birds suffering from crooked toes and ophthalmacrosis are reported in Table X.

There were no significant differences in body weight, eye weight or eye weight index between any of the groups. The birds exposed to the long photoperiod (22-1/2L-1-1/2D) had the greatest body weight, eye weight, eye weight index and incidence of crooked toes; however, the incidence of ophthalmacrosis based on visual scores was very low. The birds exposed to the superimposed $\left(\frac{\text{12L-12D} \quad \text{60 watts}}{\text{24L-0D} \quad 7\frac{1}{2}\text{ watts}}\right)$ light regime had the lowest eye weight, eye weight index, and the lowest incidence of crooked toes and none of these birds showed any evidence of ophthalmacrosis based

TABLE X

EFFECT OF LONG, SHORT, AND SUPERIMPOSED PHOTOPERIOD ON EIGHT WEEK BODY AND EYEBALL WEIGHT, INCIDENCE OF OPHTHALMACROSIS AND CROOKED TOES OF CHICKS--EXPERIMENT III

Light-dark cycle	Average weight (gms.)[1] Body	Eye	Index[1,2]	Percentage of Birds affected Toes	Eyes
$22\frac{1}{2}$L-$1\frac{1}{2}$D	687[a]	1.774[a]	.26[a]	60	90
$1\frac{1}{2}$L-$22\frac{1}{2}$D	703[a]	1.025[a]	.28[a]	80	20
12L-12D, 60 watts / 24L-0D, $7\frac{1}{2}$ watts	695[a]	1.727[a]	.25[a]	5	0

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

on visual scores. The analysis revealed that differences in eye weight were approaching significance.

Experiment IV

The data on body weight, eye weight, incidence of crooked toes and ophthalmacrosis as well as an eye weight index in which eye weight is expressed as percent of body weight are presented in Table XI.

Although none of the differences in body weight are significantly different there was a tendency for the birds treated surgically to be smaller than the estrogen implanted birds.

Regardless of whether the birds received estrogen, were castrated or were pinealectomized all birds grown under the diurnal light cycle had the smallest eyes. These differences were significant except in the case of those birds receiving estrogen and grown in continuous light. All of the birds grown in continuous darkness regardless of secondary treatment had eyes that were significantly heavier than those of birds subjected to either continuous light or a diurnal light cycle. The eyes of those birds exposed to continuous light were intermediate in weight between those exposed to either continuous darkness and the diurnal light cycle regardless of the secondary treatment.

In all cases, those birds exposed to a diurnal light cycle had a significantly smaller eye weight index. The birds exposed to continuous darkness had a consistently greater eye

TABLE XI

THE EFFECT OF CASTRATION, ESTROGEN IMPLANTATION AND PINEALECTOMY IN CONJUNCTION WITH VARIOUS PHOTOPERIODS ON BODY AND EYEBALL WEIGHT, INCIDENCE OF OPHTHALMACROSIS AND CROOKED TOES OF EIGHT WEEK OLD CHICKS--EXPERIMENT IV

Treatment		Average weight (gms.)[1]			Percentage of birds affected	
	Hours of light/day	Body	Eyeball	Index[1,2]	Toes	Eyes
Estrogen[3]	12	741[a]	1.644[a]	.23[a]	29	0
	24	681[a]	1.826[ab]	.27[b]	57	100
	0	720[a]	2.371[c]	.33[c]	43	100
Castration[4]	12	684[a]	1.734[ab]	.26[a]	29	0
	24	671[a]	1.912[b]	.29[b]	71	100
	0	646[a]	2.450[c]	.38[d]	57	100
Pinealectomy[4]	12	744[a]	1.678[a]	.22[a]	14	0
	24	651[a]	1.917[b]	.30[bc]	14	100
	0	626[a]	2.561[c]	.41[d]	100	100

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

[3]Birds implanted with a 2 mg. estrogen pellet at one week of age.

[4]Birds were operated on at one week of age.

weight index than those exposed to the other light treatments regardless of the secondary treatment. These differences were significant in all cases except of those birds given estrogen implants.

The incidence of crooked toes was greated in all groups exposed to either continuous light or dark regardless of secondary treatment except in the pinealectomized birds where those receiving continuous light had no greater incidence.

There was no ophthalmacrosis in the groups receiving a diurnal light cycle while all of the birds exposed to continuous light or dark developed ophthalmacrosis regardless of secondary treatment.

Experiment V

The data on body weight, eye weight, eye weight index and the incidence of birds with ophthalmacrosis are reported in Table XII.

There were no significant differences in body weight, regardless of treatment.

The eye weights of those birds exposed to the 12L-12D (control), the 12L-12D of bright light superimposed on 24L-OD of dim light, the 22-1/2L--1-1/2D dim light treatments were significantly smaller than those from any of the other treatments. The eye weights of those birds exposed to 24L-OD, the 12L-12D of dim light superimposed on 24L-OD of bright light, 1-1/2--22-1/2D, were significantly larger than the treatments mentioned

TABLE XII

THE EFFECT OF VARIOUS PHOTOPERIODS AND LIGHT INTENSITIES ON FOUR WEEK BODY AND EYE WEIGHT AND THE INCIDENCE OF OPHTHALMACROSIS--EXPERIMENT V

Treatment		Average weight (gms.)[1]			
Light-dark cycle	Watts	Body	Eyeball	Index[1,2]	Percentage of birds with ophthalmacrosis
0L-24D	60	332[a]	1.469[c]	.44[b]	100
24L-0D	60	308[a]	1.289[b]	.42[b]	100
12L-12D	60	308[a]	1.080[a]	.36[a]	0
12L-12D / 24L-0D	60 / 7½	326[a]	1.124[a]	.35[a]	10
12L-12D / 24L-0D	7½ / 60	305[a]	1.292[b]	.44[b]	80
22½L1½D	60	298[a]	1.096[a]	.37[a]	70
1½L-22½D	60	323[a]	1.365[b]	.43[b]	30
12L-12D	7½	319[a]	1.165[a]	.37[a]	0

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

above, but were significantly smaller than those of birds exposed to OL-24D.

The eye weight index values of those birds exposed to the 12L-12D (control), the 12L-12D of bright light superimposed on 24L-OD of dim light, the 22-1/2--1-1/2D, and the 12L-12D dim light treatments were significantly smaller than those from any other treatment.

The incidence of ophthalmacrosis was 100 percent in those birds exposed to either 24L-OD or OL-24D while the 12L-12D (control) and 12L-12D of dim light birds showed no visual evidence of ophthalmacrosis. The only other two groups showing a significant incidence of the condition were the birds exposed to the 12L-12D of dim light superimposed on 24L-OD of bright light, 1-1/2--22-1/2D, and 22-1/2L--1-1/2D, with the 1-1/2L--22-1/2D and the 12L-12D of bright light superimposed on 24L-OD of dim light being comparable with the controls.

Experiment VI

The data on body weight, eye weight, eye weight index and the incidence of birds with ophthalmacrosis are reported in Table XIII.

At four weeks the birds receiving thiouracil were smaller than any of the birds from the remaining treatments. These differences were significant except in the case of those grown on a riboflavin deficient diet and exposed to continuous light.

The eyes of birds exposed to continuous dark were significantly heavier than those from birds exposed to any of

TABLE XIII

THE EFFECT OF DIET AND PHOTOPERIOD ON FOUR WEEK BODY AND EYEBALL WEIGHT AND THE INCIDENCE OF OPHTHALMACROSIS--EXPERIMENT VI

Treatment		Average weight (gms.)[1]			
Hours of light/day	Diet	Body	Eyeball	Index[1,2]	Percentage of birds with ophthalmacrosis
0	Basal	332[b]	1.469[c]	.44[b]	100
24	Basal	308[b]	1.289[b]	.42[ab]	100
12	Basal	308[b]	1.080[a]	.36[a]	0
12	Basal + 3% Salt	269[ab]	1.049[a]	.40[ab]	0
12	Riboflavin deficient[3]	317[b]	1.150[ab]	.37[a]	0
24	Riboflavin deficient[3]	266[ab]	1.196[b]	.48[b]	80
24	Basal + Protomone[4]	315[b]	1.232[b]	.40[ab]	80
24	Basal + Thiouracil[5]	240[a]	1.105[ab]	.46[b]	80

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

[3]Essentially the basal diet with corn substituted for alfalfa and soybean oil meal substituted for fish meal and no added riboflavin in the diet.

[4]Added to basal diet at the rate of 10 gms./100 lbs. of feed.

[5]Added to basal diet at the rate of 0.1 percent.

the other treatments. All of the birds exposed to continuous light were significantly heavier than those exposed to a diurnal light cycle, regardless of the nature of the diet. The only exception to this was those birds exposed to continuous light and receiving thiouracil.

The eye weight index values of the different treatment groups did not differ greatly and this was undoubtedly due, in part, to variation in body weight. Those birds receiving continuous light or dark had significantly higher values than those receiving a diurnal light cycle, regardless of the nature of the diet with the exception of those birds receiving added salt in the diet. Apparently the salt added to the diet resulted in an increase in the eye weight index value.

The visual score of the incidence of ophthalmacrosis was zero for all of the birds receiving a diurnal light cycle and 80-100 percent in the case of those exposed to continuous light or dark regardless of dietary treatment.

Experiment VII

The data on body weight, eye weight, eye weight index and incidence of ophthalmacrosis are presented in Table XIV.

Of the three groups, the birds kept in continuous darkness had the heaviest body weights at four weeks of age, followed by those exposed to continuous light; however, these differences were non-significant.

The birds kept in continuous darkness had significantly

TABLE XIV

EFFECT OF PHOTOPERIOD ON FOUR WEEK BODY AND EYE WEIGHT, INDEX AND INCIDENCE OF OPHTHALMACROSIS--EXPERIMENT VII

Hours of light/day	Average weight (gms.)[1]		Index[1,2]	Percentage of birds with ophthalmacrosis
	Body	Eyeball		
0	273[a]	1.239[b]	0.46[b]	100
24	264[a]	1.037[a]	0.41[ab]	100
12	257[a]	0.957[a]	0.38[a]	0

[1]Averages with different superscripts differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

larger eyes than those of birds exposed to either continuous light or a diurnal light cycle. The eyes of birds exposed to continuous light were heavier than those of birds exposed to the diurnal light cycle; however, this difference was non-significant.

Index values of eye weights followed a similar pattern with the largest value belonging to the birds exposed to continuous darkness followed by those birds exposed to continuous light. The index value of those birds exposed to continuous darkness was significantly greater than that of birds exposed to a diurnal light cycle with the index value of those birds exposed to continuous light not being significantly different from either of the other two treatments.

All of the birds exposed to continuous light or continuous dark had ophthalmacrosis while none of those birds exposed to a diurnal light cycle showed evidence of the condition.

The data on sodium uptake by the blood and by the eyeball are presented in Figures 1, 2, and 3.

The birds exposed to the diurnal light cycle or continuous light seem to absorb the sodium into the bloodstream and subsequently eliminate it at a faster rate than those birds exposed to continuous darkness as can be seen in Figure 1.

Sodium appeared to be transferred into the contents of the eyeball at a faster rate in birds exposed to the diurnal light cycle than in the other two groups up to four hours after administration of the isotope; however, from four hours on these

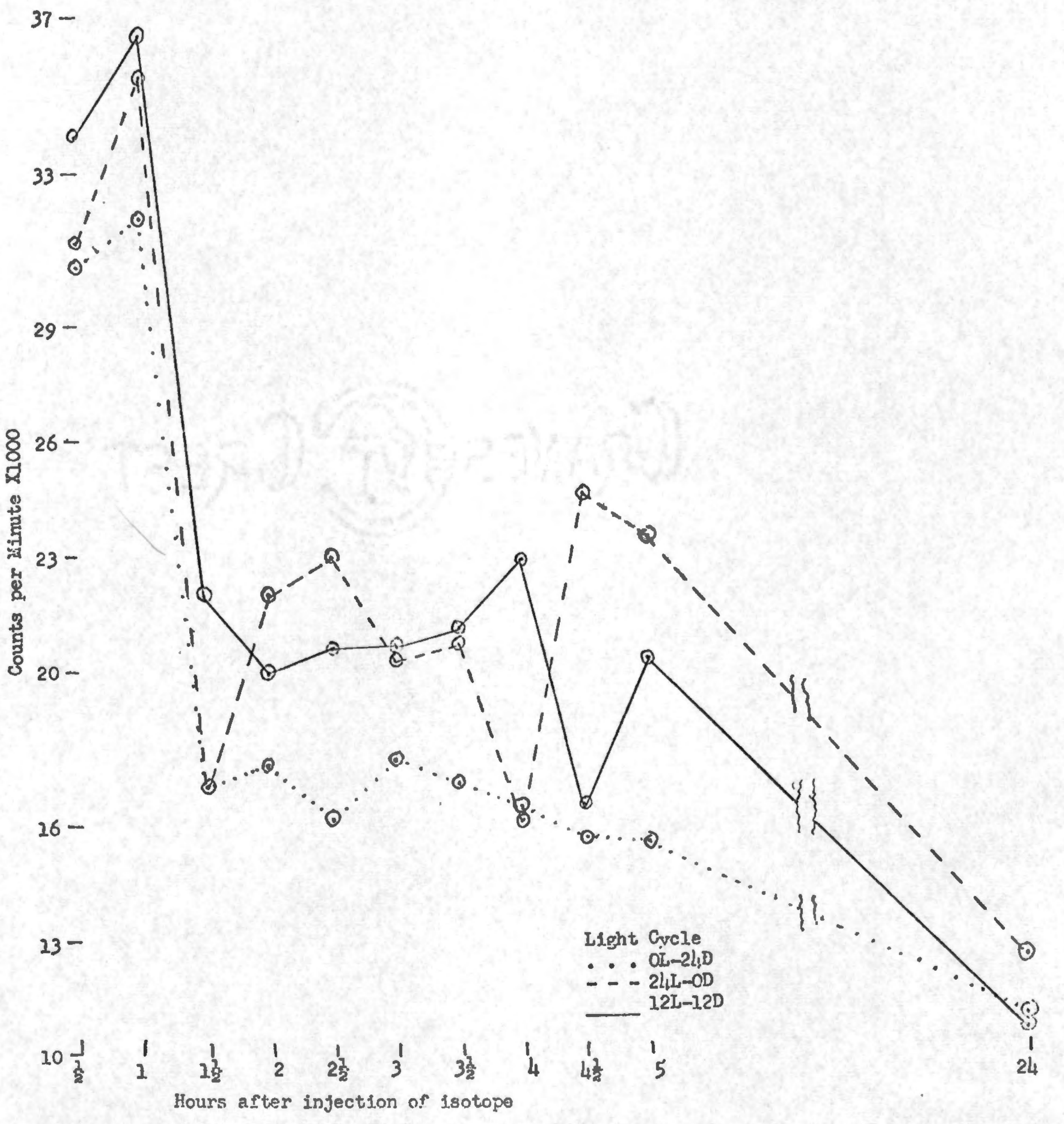

Figure 1

Blood Uptake of Sodium as Affected by Photoperiod
Experiment VII

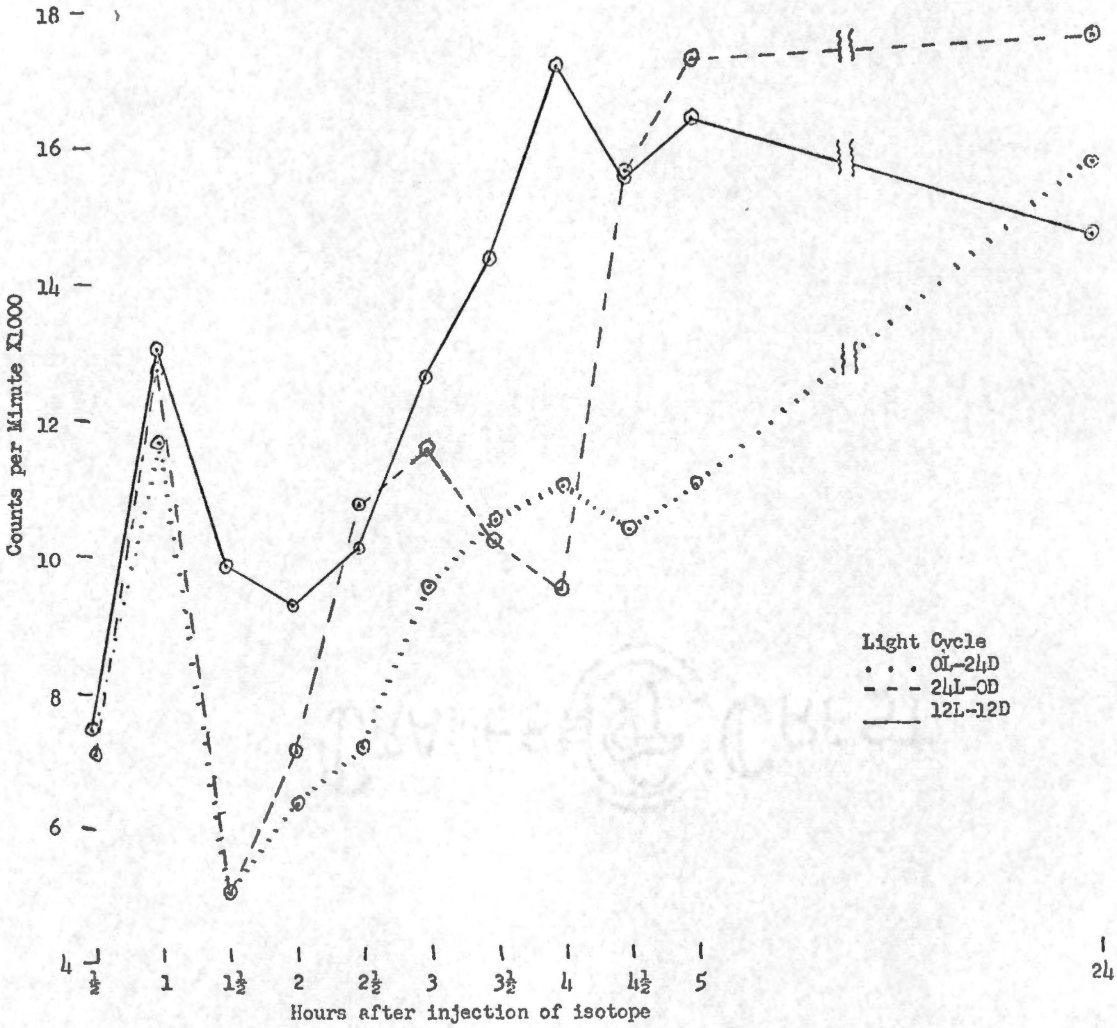

Figure 2

Eye Uptake of Sodium as Affected by Photoperiod
Experiment VII

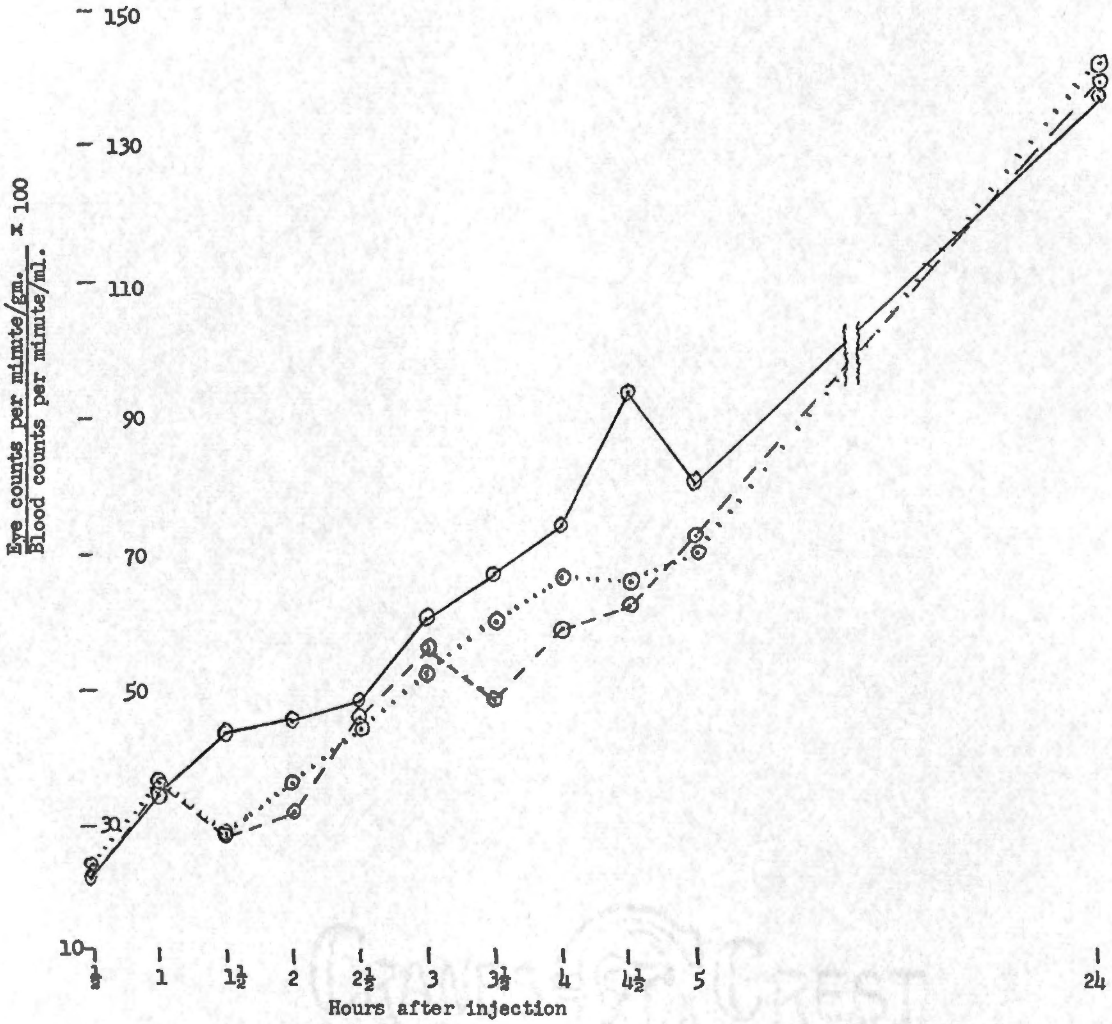

Figure 3

Sodium Uptake by the Eye From the Blood as Affected by Photoperiod
Experiment VII

birds began to lose sodium from the eye. The sodium uptake of the eyeball of birds exposed to either continuous light or continuous darkness seemed to parallel each other up to four hours after administration of the isotope at which time the uptake of sodium by the group exposed to continuous light greatly exceeded that of the birds from the other two light treatments up to five hours after administration and climbed gradually from five to twenty-four hours after administration. From four to twenty-four hours after administration of the isotope the birds exposed to continuous darkness showed an almost linear uptake of sodium. These comparisons are easily seen in Figure 2 (page 47).

When the uptake of sodium in the eyeball is compared to the uptake of sodium by the blood the increase is almost linear from the beginning of the experiment to its conclusion. These comparisons are presented in Figure 3 (page 48).

When sodium uptake of the eyeball was expressed as percent of the standard it was directly related to the degree of ophthalmacrosis, while sodium's appearance in the blood did not appear to be related to the degree of ophthalmacrosis.

None of the comparisons in Table XIV (page 44) or in Figures 1, 2, or 3 (pages 46, 47, and 48) are significantly different from each other, due to the tremendous variation within each treatment group.

Experiment VIII

The data on body weight, eyeball weight, adrenal weight, eye weight index and incidence of ophthalmacrosis are reported in Table XV.

The body weights of those birds exposed to continuous darkness were significantly heavier than those of all of the other treatments except those exposed to the short (1-1/2L--22-1/2D) photoperiod. The addition of 1 ppm reserpine to the diet did not significantly effect body weight except for the significant reduction in body weight for those birds fed reserpine and exposed to continuous light.

The eye weights of those birds exposed to continuous darkness were significantly heavier than the eyes of birds exposed to any of the other treatments. In those birds exposed to either continuous darkness or a diurnal light cycle the addition of reserpine to the diet tended to increase eye weight while in those birds exposed continuous light added reserpine resulted in a reduced eye weight. This latter observation is clouded by the variation in body weight.

The eye weight index values were significantly smaller for those birds exposed to a diurnal light cycle except when compared to those birds exposed either the long (22-1/2L--1-1/2D) or short (1-1/2--22-1/2D) photoperiod. The addition of reserpine to the diet did not significantly effect index values.

The adrenal weights of those birds exposed to continuous light were significantly reduced by the addition of reserpine to the diet while the addition of reserpine to the diets of those birds exposed to either continuous dark or a diurnal light

TABLE XV

THE EFFECT OF PHOTOPERIOD AND RESERPINE ON EIGHT WEEK BODY, EYE, AND ADRENAL WEIGHTS AND THE INCIDENCE OF OPHTHALMACROSIS--EXPERIMENT VIII

Light-dark cycle	Average weight (gms.)[1]				Percentage of birds with ophthalmacrosis
	Body	Eye	Adrenal	Index[1,2]	
12L-12D	564[ab]	1.66[a]	37.5[ab]	.30[a]	0
12L-12D (R)[3]	602[ab]	1.72[ab]	39.8[b]	.29[a]	0
24L-0D	620[b]	2.11[c]	40.2[b]	.34[b]	100
24L-0D (R)[3]	541	1.85[b]	32.6[a]	.34[b]	100
0L-24D	705[c]	2.55[d]	35.8[ab]	.36[b]	100
0L-24D (R)[3]	754	2.67[d]	39.2[b]	.35[b]	100
22½L-1½D	539[a]	1.68[a]	32.6[a]	.32[ab]	40
1½L-22½D	650[bc]	1.98[bc]	36.2[ab]	.31[ab]	0

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

[3]Reserpine was present in the diet at the level of 1 ppm.

cycle increased adrenal weights, however, these latter differences were not significant. No clear cut pattern could be established from this data.

All of the birds exposed to either continuous light or dark developed ophthalmacrosis regardless of the presence or absence of reserpine in the diet. The birds exposed to the long and short photoperiod were nearly normal when visually scored for ophthalmacrosis with 40 percent and 0 percent of the birds showing enlargement respectively.

Experiment IX

The data on the incidence of ophthalmacrosis and retinal tears are presented in Table XVI.

At eight weeks of age when the switch to the new lighting regimes took place, all of the birds previously exposed to either continuous light or dark had ophthalmacrosis while none of those birds exposed to a diurnal light cycle exhibited the condition. Regardless of the initial treatment from 0-8 weeks of age all birds subjected to a diurnal light cycle from eight weeks had taken on a normal appearance to the eyes by the thirteenth week of age and their eyes remained normal in appearance throughout the remainder of the experiment. The visual scores of those birds placed on continuous light at eight weeks of age did not change during the remainder of the experiment, i.e., those that were enlarged remained enlarged and those that were normal remained normal. The eyes of birds

TABLE XVI

THE EFFECT OF SWITCHING PHOTOPERIODS AT EIGHT WEEKS OF AGE ON THE INCIDENCE OF OPHTHALMACROSIS AND RETINAL TEARS--EXPERIMENT IX

Hours light/day		Ophthalmacrosis Age in weeks				Retinal Tears
0-8 weeks	8-22 weeks	8	13	18	22	22
				percent		
12	0	5	50	50	100	75
12	24	0	0	0	0	0
12	12	0	0	0	0	0
24	0	100	100	100	100	100
24	24	100	100	100	100	71.4
24	12	100	0	0	0	42.9
0	0	100	100	100	100	100
0	24	100	100	100	100	100
0	12	100	0	0	0	100

that were switched from continuous light to continuous dark or those that remained on continuous darkness throughout the experiment were enlarged upon the first and all subsequent examinations while 50 percent of those that were switched from a diurnal light cycle to continuous darkness exhibited typical signs of ophthalmacrosis after five weeks on this latter regime. Upon the examination at 18 weeks of age 50 percent of the birds had eyes that were still enlarged and in the remainder the fundus had become so obvious as to be observable without the aid of instruments while at 22 weeks of age all birds exhibited typical signs of ophthalmacrosis. All of the birds exposed to continuous light or dark for the entire 22 weeks of the experiment had substantial retinal tears while 75 percent of those birds switched from a diurnal light cycle to continuous darkness also had retinal tears. None of the birds that remained on the diurnal light cycle for the entire 22 weeks showed any evidence of retinal tears. All of the birds switched from continuous darkness to a diurnal light cycle had retinal tears while over 40 percent of those switched from continuous light to a diurnal light cycle had retinal tears. All of the birds switched from continuous darkness to continuous light had retinal tears while over 70 percent of those remaining on continuous light for the entire 22 weeks of the experiment had retinal tears. None of the birds that were switched from a diurnal light cycle to continuous light showed any evidence of retinal tears.

The data on body weight, eye weight, eye weight index and adrenal weight are reported in Table XVII.

At 22 weeks of age those birds that had been switched from continuous darkness to either continuous light or a diurnal light cycle were lighter in weight than those exposed to either continuous light or a diurnal light cycle from hatching to eight weeks of age. The heaviest birds in the test were those switched from continuous light to a diurnal light cycle. Those birds exposed to continuous darkness the last 14 weeks of the experiment had approximately the same body weight regardless of the former light treatment.

Eye weight and incidence of ophthalmacrosis were greatest for those birds exposed to continuous darkness followed by those exposed to continuous light, regardless of whether the birds received these treatments from 0-8 weeks of age, 8-22 weeks of age or 0-22 weeks of age indicating a definite interaction between the former and subsequent photoperiod on the development of ophthalmacrosis.

The adrenal weights of those birds exposed to the same photoperiod for the entire 22 weeks of the experiment were greatest for those birds exposed to continuous darkness followed by those of birds exposed to continuous light, i.e., directly related to the degree of ophthalmacrosis. The exact opposite was true of those birds exposed to a diurnal light cycle the first eight weeks of the experiment while the adrenal weights of the birds exposed to either continuous light or dark for the first eight weeks of the experiment appeared to have no relation to

TABLE XVII

THE EFFECT OF SWITCHING PHOTOPERIODS AT EIGHT WEEKS OF AGE ON BODY, EYE AND ADRENAL WEIGHTS OF CHICKENS AT TWENTY-TWO WEEKS OF AGE EXPERIMENT IX[1]

Hours of light/day		Average weights (gms.)			
0-8 weeks	8-22 weeks	Body	Eye	Adrenal	Index[2]
12	0	1642	3.58	.1052	.222
12	24	1861	2.80	.0760	.150
12	12	1785	2.65	.0870	.148
24	0	1573	3.63	.0838	.237
24	24	1617	3.54	.0961	.219
24	12	1781	2.72	.0851	.153
0	0	1695	4.60	.0983	.271
0	24	1680	3.94	.0948	.235
0	12	1618	3.65	.0737	.226

[1]No statistical analysis was run on these data as excess mortality resulted in extreme variation in the number of birds remaining (see page 17).

[2]Eye weight/body weight x 100.

the development of ophthalmacrosis.

Experiment X

The data on body weight, eye weight and eye weight index are presented in Table XVIII.

The chicks hatched without light in the incubator were significantly heavier than those incubated in continuous light. Although the chicks receiving a diurnal light cycle during incubation were intermediate in weight they were not significantly different from either of the other two treatments.

While there were no significant differences between treatments with regard to eye weight, those chicks incubated in either continuous light or dark had heavier eye weights than those incubated in a diurnal light cycle.

The eye weight index values of those chicks exposed to continuous light were significantly greater than those of the other two treatments; however, differences in body weight have affected these results.

Experiment XI

The data on body weight, eye weight, eye weight index, thyroid weight, adrenal weight, testis weight, bursa weight and incidence of ophthalmacrosis and retinal damage are presented in Table XIX.

In the birds exposed to continuous darkness or a diurnal light cycle the unoperated controls were the heaviest, followed

TABLE XVIII

THE EFFECT OF PHOTOPERIOD DURING INCUBATION ON BODY AND EYE WEIGHT OF CHICKS EXPERIMENT X

Hours of light/day	Average weight (gms.)[1]		
	Body	Eye	Index[2]
12	39.5[ab]	45.7[a]	1.16[a]
24	38.1[a]	47.3[a]	1.25[b]
0	41.2[b]	46.2[a]	1.12[a]

[1]Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

TABLE XIX

THE EFFECT OF PINEALECTOMY IN CONJUNCTION WITH PHOTOPERIOD ON EIGHT WEEK BODY, EYE, THYROID, ADRENAL, TESTIS, AND BURSA WEIGHTS, INCIDENCE OF OPHTHALMACROSIS AND RETINAL TEARS--EXPERIMENT XI

Hours of light per day	Surgical Treatment	Average weight (gms.)[1]						Index[1,2]	Percentage of birds affected	
		Body	Eye	Thyroid	Adrenal	Testis	Bursa		Eye	Retina
0	Pinealectomy	748^{b}	2.58^{c}	$.101^{a}$	$.088^{a}$	2.11^{a}	3.24^{a}	$.346^{b}$	100	25
0	Sham	789^{bc}	2.78^{c}	$.091^{a}$	$.088^{a}$	1.94^{a}	4.69^{a}	$.354^{b}$	100	50
0	Control	813^{c}	2.66^{c}	$.109^{a}$	$.086^{a}$	2.23^{a}	3.58^{a}	$.328^{b}$	100	37
24	Pinealectomy	679^{a}	1.93^{b}	$.116^{a}$	$.081^{a}$	3.40^{ab}	3.50^{a}	$.285^{ab}$	100	0
24	Sham	744^{b}	1.96^{b}	$.157^{a}$	$.082^{a}$	4.16^{b}	4.19^{a}	$.265^{ab}$	100	0
24	Control	678^{a}	1.83^{b}	$.140^{a}$	$.080^{a}$	4.92^{b}	3.80^{a}	$.272^{ab}$	100	12.5
12	Pinealectomy	696^{ab}	1.74^{ab}	$.104^{a}$	$.082^{a}$	2.42^{a}	3.08^{a}	$.252^{ab}$	0	0
12	Sham	702^{ab}	1.76^{ab}	$.121^{a}$	$.098^{a}$	2.23^{a}	3.22^{a}	$.257^{ab}$	0	0
12	Control	717^{ab}	1.68^{a}	$.117^{a}$	$.091^{a}$	2.60^{a}	3.98^{a}	$.236^{a}$	0	0

[1]Averages with different superscripts differ significantly ($P \leq 0.05$).

[2]Eye weight/body weight x 100.

by the sham operated birds with the pinealectomized birds being the lightest. These differences were non-significant except in the comparison between the controls and the pinealectomized birds grown in total darkness. The birds exposed to continuous light and sham operated were significantly heavier than the pinealectomized or control group exposed to continuous light. The birds grown in continuous darkness were consistently heavier than those grown under either of the other two light treatments, regardless of surgical treatment.

The sham operation or pinealectomy increased eye weights although these differences were not statistically significant within a single light treatment. The eyes of birds grown in continuous darkness were significantly heavier than those from the birds of the other two light treatments, regardless of surgical treatment. The eye weights of those birds exposed to continuous light were heavier than those of birds exposed to a diurnal light cycle; however, these differences were only significant in comparing those grown in continuous light with the control birds grown under a diurnal light. The sham operated birds had consistently heavier eyes than the pinealectomized birds within a light treatment; however, these differences were non-significant.

Eye weight index values were the greatest for those birds grown in continuous darkness followed by those birds grown in continuous light, with the smallest values being from those birds exposed to a diurnal light cycle, regardless of surgical treatment. These differences were only significant between the control birds exposed to a diurnal light cycle and all of the birds exposed to continuous darkness, regardless of surgical treatment. Pinealectomy or sham operation did increase index values when compared to the unoperated control within a light treatment; however, these differences were non-significant.

Thyroid weights were not significantly different from each other, regardless of light or surgical treatment; however, these differences were approaching significance. The birds exposed to continuous dark seemed to have the lightest thyroids, with those birds exposed to continuous light being the heaviest.

Adrenal weights were not significantly different from each other and no trend could be detected.

The testis weights of those birds exposed to continuous dark or a diurnal light cycle were not significantly different from each other; however, the former group was consistently lighter than the latter when compared within a surgical treatment. Within these two groups the sham operated birds had the lightest tests with the controls having the heaviest. The birds exposed to continuous light had consistently heavier testes than those of birds exposed to the other two light treatments; these differences were significant except in the case of the pinealectomized birds.

Bursa weights were not significantly different from each other regardless of light or surgical treatment; however, these values were approaching significance. The pinealectomized birds had the lightest bursa weights within a light treatment; however, differences in bursa weights seemed to be related to differences in body weight rather than to treatment *per se*.

All of the birds exposed to either continuous light or continuous darkness exhibited ophthalmacrosis upon visual examination while none of those birds exposed to a diurnal light cycle exhibited any signs of the condition, regardless of surgical treatment. There was no evidence of retinal damage as scored by the unaided eye in any of the birds exposed to a diurnal light cycle and none in those birds exposed to continuous light and subsequently sham operated or pinealectomized while 12.5 percent of the unoperated controls exposed to continuous light showed evidence of retinal damage. The birds exposed to continuous darkness had 25, 50 and 27 percent incidence of retinal damage in the pinealectomized sham operated and control groups respectively.

Experiment XII

The data on body weight, eye weight, eye weight index, thyroid weight, adrenal weight, comb weight, testis weight, bursa weight, incidence of ophthalmacrosis and retinal damage are presented in Table XX.

When compared within a surgical treatment the birds exposed to a diurnal light cycle had the heaviest body weights. When compared within a light treatment, the controls were the heaviest followed by the pinealectomized birds with the lightest

TABLE XX

THE EFFECT OF PINEALECTOMY IN CONJUNCTION WITH PHOTOPERIOD ON NINETEENTH WEEK BODY, EYE, THYROID, ADRENAL, COMB, TESTIS AND BURSA WEIGHTS, INCIDENCE OF OPHTHALMACROSIS AND RETINAL TEARS
EXPERIMENT XII

Hours of light per day	Treatment	Average weight (gms.)[1]							Index[1,2]	Percentage of birds affected	
		Body	Eye	Thyroid	Adrenal	Comb	Testis	Bursa		Eye	Retina
0	Pinealectomy	1665^{a}	4.61^{d}	$.129^{a}$	$.082^{a}$	$.054^{a}$	8.5^{ab}	$.464^{a}$	$.279^{c}$	100	50
0	Sham	1523^{a}	4.08^{c}	$.102^{a}$	$.085^{a}$	$.061^{a}$	12.1^{b}	$.511^{a}$	$.269^{c}$	100	75
0	Control	1693^{a}	4.27^{c}	$.100^{a}$	$.089^{a}$	$.063^{a}$	9.9^{b}	$.639^{a}$	$.252^{c}$	100	100
24	Pinealectomy	1534^{a}	3.12^{b}	$.211^{a}$	$.092^{a}$	$.057^{a}$	5.7^{a}	$.429^{a}$	$.205^{b}$	100	0
24	Sham	1478^{a}	2.99^{ab}	$.156^{a}$	$.091^{a}$	$.066^{a}$	7.0^{ab}	$.513^{a}$	$.204^{b}$	100	0
24	Control	1665^{a}	2.92^{ab}	$.102^{a}$	$.105^{a}$	$.078^{a}$	7.4^{ab}	$.555^{a}$	$.175^{ab}$	100	50
12	Pinealectomy	1771^{a}	2.59^{a}	$.116^{a}$	$.105^{a}$	$.066^{a}$	10.3^{b}	$.369^{a}$	$.147^{a}$	0	0
12	Sham	1683^{a}	2.52^{a}	$.182^{a}$	$.101^{a}$	$.056^{a}$	8.1^{ab}	$.396^{a}$	$.150^{a}$	0	0
12	Control	1819^{a}	2.56^{a}	$.208^{a}$	$.116^{a}$	$.066^{a}$	8.9^{ab}	$.508^{a}$	$.142^{a}$	0	0

[1] Averages with different superscripts within a column differ significantly ($P \leq 0.05$).

[2] Eye weight/body weight x 100.

birds being those that were sham operated. None of these differences were significant; however, an analysis revealed that they were approaching significance.

The eye weights of the birds exposed to continuous darkness were significantly heavier than those from any of the other treatments. The birds exposed to continuous light were the next heaviest; however, only those birds of this group that were pinealectomized had eye weights that were significantly heavier than those of the birds exposed to a diurnal light cycle. When compared within a light treatment the pinealectomized birds had the heaviest eyes followed by the unoperated controls except in the case of the birds exposed to continuous light where the sham operated birds had heavier eyes than the controls. None of these differences were significant.

The eye weight index values when compared within a surgical treatment were significantly greater for the birds exposed to continuous darkness followed by those birds exposed to continuous light. In this latter group the differences were significant except in the case of those controls.

The thyroid weight seemed to be related to the amount of light received per day and were lightest for those birds exposed to continuous darkness; however, these differences only approached significance. In the birds exposed to either continuous light or continuous darkness, the heaviest thyroid weights were found in those birds that had been pinealectomized followed by the sham operated group with the thyroids from the controls being

being the lightest; however, in the group exposed to a diurnal light cycle this order was reversed.

Adrenal weights were greatest in the birds exposed to a diurnal light cycle, followed by those birds exposed to continuous light, with the group exposed to continuous darkness having the lightest adrenals regardless of surgical treatment. When compared within a light treatment the controls had consistently heavier adrenals. None of these differences approached significance.

The comb weights were not significantly different from one another; however, in the birds exposed to continuous light or continuous darkness the comb weights were the heaviest for the unoperated controls followed by the sham operated birds. In the group exposed to a diurnal light cycle the sham operated birds had the smallest combs with the size of the combs of the other groups being equal.

The testis weights of the birds exposed to continuous light were lighter than those exposed to either continuous darkness or a diurnal light cycle. The only significant difference was between the pinealectomized birds exposed to continuous light and a diurnal light cycle with the latter having the heaviest testes.

Comparing birds within a light treatment, those that were pinealectomized had the lightest bursas with the controls having the heaviest bursas and the sham operated birds having bursas of intermediate weight. In general the bursas of those birds exposed to continuous darkness were the heaviest followed by those exposed to continuous light with the group exposed to a diurnal light cycle having the lightest bursas. None of these differences were significant.

All of the birds exposed to either continuous light or continuous darkness exhibited ophthalmacrosis upon visual examination while none of those birds exposed to a diurnal light cycle exhibited any signs of the condition, regardless of surgical treatment. There was no evidence of retinal damage as scored by the naked eye in any of the birds exposed to a diurnal light cycle and none in those birds exposed to continuous light and subsequently sham operated or pinealectomized while 50 percent of the unoperated controls in an unoperated group exposed to continuous light showed evidence of retinal damage. The birds exposed to continuous darkness had 50, 75 and 100 percent incidence of retinal damage in the pinealectomized, sham operated and control groups respectively.

V. DISCUSSION

The data indicate a definite relationship between photoperiod and body weight. At one day of age the advantage seemed to lie with the birds exposed to continuous darkness; however, these differences were non-significant. From two through six weeks of age regardless of the color or intensity of the light, the birds exposed to continuous light were consistently heavier than those exposed to either continuous dark or a diurnal light cycle, and in most cases these birds were significantly heavier than the controls that had been exposed to a diurnal light cycle. At eight and ten weeks of age the birds grown in continuous darkness were consistently heavier and in the majority of cases they were significantly heavier than the controls that had been exposed to a diurnal light cycle. The one major exception to this was in the birds that had either been castrated or implanted with estrogen in which cases the control birds were the heaviest; however, these differences were non-significant. By 19 weeks of age the birds receiving a diurnal light cycle had become consistently heavier even when surgical treatment, i.e., pinealectomy, or sham operation, was superimposed over light treatment and this trend continued through 22 weeks of age which is the greatest age to which any of the birds were carried. A possible explanation for differences in body weight up through ten weeks of age is that one significant part of the syndrome developing upon exposure to either continuous light or continuous darkness is the extreme calmness and docile condition of these birds. This would allow these birds to be more efficient converters of food since they do not waste energy flying or running about. In addition since lighting conditions do not signal a period of sleep as in the diurnal light cycle, the birds may tend to eat at all times throughout the day rather than eating in a restricted period such as is the case for birds exposed to a diurnal light cycle, and therefore, consume more food.

The fact that the birds exposed to continuous light are heaviest to six weeks of age could possibly be due to the inability of chicks grown in continuous darkness to find sufficient food early in the growing period until they fix the location of the feeders and waterers. This hypothesis is not sufficient to explain the fact that the birds exposed to a diurnal light cycle become the heaviest as they approach sexual maturity. One possible clue here is the fact that the birds exposed to a diurnal light cycle and either castrated or implanted with estrogen were also the heaviest at eight weeks of age, indicating a possible interaction between the release of significant amounts of sex hormones and a diurnal light cycle.

The data on the development of ophthalmacrosis definitely demonstrate that continuous light does cause the eyeballs to enlarge when compared to the controls, regardless of the color or intensity of the light, but more strikingly that continuous darkness causes the enlargement to an even greater extent.

This latter fact is not reported in the literature. On the basis of these findings the hypothesis was developed that ophthalmacrosis is not due merely to the presence or absence of light but is due to a lack of a diurnal light cycle by which the chicken could set its biological clock(s).

Partial proof of this hypothesis was found in the results from Experiment III, which demonstrated that breaking the day into a short or long photoperiod or exposing the birds to a superimposed photoperiod results in less apparent eye enlargement. The superimposed lighting regime as discussed earlier is really a continuous lighting regime consisting of a modulating diurnal light cycle of 12 hours of bright light and 12 hours of dim light per day. This is definite proof that the pre-existing literature is incorrect in simply stating that ophthalmacrosis is due to exposure to continuous light as no indication of ophthalmacrosis was found in the birds exposed to the continuous light of the superimposed lighting regime. The results from Experiment V strongly re-enforce these data and provide additional evidence in support of the hypothesis. It was clearly indicated by these data that the greater the difference in intensity between the two phases of the light cycle the more nearly normal were the birds, but that even a rather subtle difference in the two phases would lessen the degree of ophthalmacrosis. All three experiments designed specifically to test this hypothesis indicated that even short breaks of 1-1/2 hours per 24 in a continuous lighting regime (continuous light or dark) would reduce the severity of the condition.

It appears that at hatching chicks incubated in the normal manner are already suffering from ophthalmacrosis and that subsequently placing the birds under a diurnal light cycle causes the eyes to return to normal before any damage is done; however, if they are placed in either continuous light or continuous darkness at hatching the condition continues to worsen, vision is impaired and severe unreparable retinal damage, i.e., retinal tears, may occur if the birds are not soon placed under a diurnal light cycle.

Regardless of whether the birds are placed under a continuous lighting regime early (0-8 weeks of age) or later in life (8-22 weeks of age) they develop the typical symptoms of ophthalmacrosis; however, the appearance of the symptoms require much longer exposure later in life than they do earlier in life. This may be because the longer the birds are exposed to a diurnal light cycle the more firmly entrenched in their nervous system is this diurnal rhythm and the longer is required for this rhythm to fade. The birds that are exposed to a continuous lighting regime from one day of age for eight weeks (continuous light or dark) all develop typical signs of ophthalmacrosis; however, if switched to a diurnal light cycle at eight weeks of age all return to normal appearance. This indicates that by re-establishing the external diurnal rhythm the internal diurnal rhythm is also restored and the condition begins to correct itself. When the birds exposed to the diurnal light cycle from 8 weeks of age were examined at 22 weeks of age the degree of retinal damage was directly related to the degree of ophthalmacrosis; however, the birds were examined only macrosopically for evidence of retinal tears. When the birds were switched from a diurnal light cycle to either continuous light or continuous dark at eight weeks of age all were normal with respect to ophthalmacrosis, and as far as outward appearance is concerned none of the birds developed the typical symptoms of ophthalmacrosis even after being exposed to continuous light for 14 weeks; however, examination of the enucleated eye showed that ophthalmacrosis had begun to develop in these birds. This is in contrast to those birds switched from a diurnal light cycle at 8 weeks of age to continuous dark for the remaining 14 weeks of the experiment. After only 5 weeks on this latter regime 50 percent of the birds developed typical signs of ophthalmacrosis and after ten weeks all had some outward evidence of the development of ophthalmacrosis while at 22 weeks of age 75 percent of the birds had suffered retinal damage. One possible explanation for this phenomenon and the fact that birds subjected to continuous dark are more severely affected than those exposed to continuous light, is that when the birds exposed to continuous light close their eyes to sleep they change the intensity of the light incident on the retina and therefore the type of signal relayed to the brain; however, this is not true of the birds subjected to continuous dark. It may be that variation in intensity of light incident on the retina signals the biological clock(s) to reset and are therefore temporarily synchronized although not as effectively as in the case of birds subjected to a diurnal cycle of light; these birds are therefore intermediate with respect to the development of ophthalmacrosis between the controls that receive an unvarying diurnal signal and the continuous dark group that receives no variation in intensity. It may be argued that other external stimuli such as the activity of the caretaker, noise, changes in humidity, temperature, to name a few might allow the bird to "key" on these rather than light, and it is logical to assume that these influences tend to cloud the results; however, light seems to be such an important "time cue" for chickens that the effects of these secondary signals are for the most part overcome.

It was obvious from all of the experiments that the degree of retinal damage was directly related to the degree of ophthalmacrosis. The retinal tears seemed to be due to over-stretching of this delicate tissue from the accumulation of excess fluid in the posterior chamber of the eye. It appears that up to the age of twenty-two weeks, the outer coats of the eye are elastic enough to stretch with the increased amount of fluid while the retina, being more fragile, merely ruptures.

The data on adrenal weights were extremely variable and no significant differences were found between groups of birds regardless of the light treatment they had been placed on or the surgical treatment they were subjected to. The differences in adrenal weight could not be tied to the degree of eye enlargement as the results varied from experiment to experiment; however, there was a slight indication in the data from the birds sacrificed in Experiments IX, XI, and XII, that adrenal size may be reduced in birds suffering from ophthalmacrosis. This agrees with the findings of Hogg (1967). It must be remembered that endocrine organ output cannot be gauged from the fact that the organ in question is larger or smaller than the controls. Hogg (1967) demonstrated that the corticosterone output of the adrenals was positively related to the degree of ophthalmacrosis while the adrenal size was negatively related. The fact that adrenal size was affected by the light treatment and not that it was consistently larger or smaller for a specific treatment is the important point, since size and secretion rate are not specifically related.

In the case of the pituitary weights from birds exposed to continuous light, continuous dark or a diurnal light cycle, all were significantly different from each other with the size of the pituitary being related to the amount of light received per day and not to the degree of ophthalmacrosis. This is to be expected as light is especially important in regulating pituitary secretion. This says nothing about the type of secretions or the receptivity of their target organs.

The thyroid is one of the target organs of the pituitary and although pituitary and thyroid weights are not available from the same experiments it should be noted that thyroid weights, although not significantly different, seemed to be positively related to the amount of light received per day as was the case with the pituitaries removed in an earlier experiment. These were the same trends pointed out by Hogg (1967). Although differences in body weight cloud the issue, the data from Experiment VI indicate that addition of thyroid hormone-like material in the form of protamone might reduce the severity of the condition while added thiouracil may increase the severity of the condition. It must be pointed out that these differences are non-significant and would not be considered at all if these data did not correspond to the thyroid weights of Experiment XI and XII.

Up to eight weeks of age testis size seems to be directly related to the quality and quantity of light received; however, as the birds approach maturity, testis size seems to be inversely related to the quantity of light to which the birds are exposed. Up to eight weeks of age, testis size and comb size have a fairly direct relationship; however, as the birds approach maturity the relation between testis size and comb size is not good. This is another example of the size of an endocrine organ not indicating the level of secretion of the organ as comb size is highly positively correlated with hormone secretion. In the birds receiving a continuous lighting regime (continuous light or dark to 19 weeks of age) the pinealectomized birds had smaller testes and smaller combs, indicating a possible relationship. This trend was becoming evident in those birds exposed to continuous light to 8 weeks of age, but not in those exposed to continuous darkness. None of these differences were significant.

The data from Experiment IV indicate that the presence of female hormone in the circulatory system may reduce the severity of ophthalmacrosis though it had no effect on the incidence of the abnormality as determined by visual score, while pinealectomy seemed to increase the severity of the condition. This latter statement is supported by the data from Experiment I which indicate that pineal weight in the intact bird is inversely related to the severity of ophthalmacrosis and the data from Experiments XI and XII that indicate also that pinealectomy increases the severity of ophthalmacrosis. According to Gaston and Menaker (1968), the pineal is associated with internal diurnal rhythm and affected by external diurnal light rhythms and may well be influential in the secretion of the major hormones of the body.

Sturkie (1965) reported that bursa weight is inversely related to adrenal cortex secretion and might serve as an indirect measure of adrenal activity. The bursa weights of Experiment XI and XII, were inversely related to the degree of ophthalmacrosis and serve as further evidence that excess adrenal cortical secretion may be involved in the development of this abnormality.

The natural assumption is that if adrenal cortical secretions are increased, then certain minerals, especially sodium, might appear in excess amounts in the eye. The birds exposed to either a diurnal light cycle or continuous light took sodium into the blood stream and subsequently gave it up faster than the birds exposed to continuous darkness and up to five hours after administration of sodium the uptake of sodium into the eye was directly related to its appearance in the blood; however, from 5 to 24 hours the birds suffering from ophthalmacrosis showed a continued increase in amounts of sodium taken into the eye while the controls showed a decline. This latter data lends strong support to the involvement of sodium in the development of this abnormality. The addition of sodium to the diet in the form of sodium chloride did not seem to affect the development of ophthalmacrosis to any significant degree, however.

In addition to eye involvement, certain strains of birds showed marked increases in the incidence of crooked toes that corresponded to the degree of ophthalmacrosis. This type of crooked toe is not related to the curled toe paralysis as exhibited by riboflavin deficient birds. Riboflavin deficient birds do not show any greater incidence of eye involvement than birds fed a normal diet and apparently this nutrient is not related to the development of ophthalmacrosis.

Another significant part of the syndrome developing in birds suffering from ophthalmacrosis is their extreme calmness and docile appearance when compared with the normally "nervous" controls. The feeding of reserpine, which is a tranquilizer that has a calming effect on the birds, to those birds exposed to continuous darkness or a darkness or a diurnal light cycle tended to result in larger body and eye weights while in those exposed to continuous light the opposite was true. From these data it appears that ophthalmacrosis is not contributed to by the calmness of the birds, but that this is a secondary condition that develops at the same time; however, these data are important in that they lend support to the hypothesis that the calmness and docile condition contribute to increased body weights by allowing the birds to be more efficient converters of food since they do not waste energy flying or running.

Based on these data, a hypothesis is presented that ophthalmacrosis develops as a result of the lack of a diurnal light cycle by which the chicken can set its biological clock(s). Apparently this lack of a diurnal light cycle sets up a stress condition due to the lack of appropriate signals reaching the hypothalamic region of the brain. This possibly results in the release of adrenocorticotrophic hormone and perhaps other as yet unidentified releasing factors that act on the adrenal cortex. This stimulates the release of the mineralo-corticoids of the adrenal cortex that act directly on sodium, potassium and water metabolism. This results in a general systemic upset due to improper mineral balance, improper acid and base balance and the adrenal hormones action on other endocrine organs, specifically the thyroids. Increased amounts of sodium enter the eye and water follows causing a physical stretching of the eye similar to that caused by blowing up a balloon. The more delicate retina may tear after a significant amount of enlargement has occurred; however, even if retinal tears do not develop vision is undoubtedly impaired severly by the alteration of eye shape as well as the change in the make up of the occular fluids.

VI. SUMMARY

A series of twelve experiments was conducted to study factors in the development of ophthalmacrosis (eye enlargement) in chickens. Treatments included various photoperiods, colors and intensities of light, extirpation of endocrine organs, application of hormones and certain feed additives.

It was found that exposure of chicks to continuous light, regardless of the color or intensity, caused the eyeballs to enlarge, but more strikingly that continuous darkness caused the enlargement to an even greater extent. Breaking the day into long or short photoperiods or exposing the chicks to a superimposed photoperiod (continuous light broken into twelve hours of bright and twelve hours of dimmer light per day) resulted in less apparent eye enlargement than was found in the birds exposed to a continuous lighting regime (continuous light or dark). When birds were exposed to the diurnal light cycle the eyes appeared to be unaffected.

Chicks hatched from eggs incubated in continuous darkness appeared to have ophthalmacrosis at hatching time; however, when the chicks were subsequently placed under a diurnal light cycle they returned to normal. When these chicks were placed under a continuous lighting regime upon hatching the condition continued to worsen and permanently impaired vision resulted if the birds were not soon placed under a diurnal light cycle.

Regardless of whether the birds were placed under a continuous lighting regime from 0-8 weeks or from 8-22 weeks of age, they developed ophthalmacrosis; however, the symptoms appeared more slowly in birds during the 8-22 week period. When birds with ophthalmacrosis were switched to a diurnal light cycle from 8-22 weeks all returned to normal but with varying degrees of retinal damage. Switching birds with normal eyes to a continuous lighting regime from 8-22 weeks resulted in varying degrees of ophthalmacrosis and retinal damage. In all cases the degree of retinal damage was positively related to the degree of ophthalmacrosis. It was concluded that the retinal damage was due to over-stretching of the delicate tissue resulting from the accumulation of excess fluid in the posterior chamber of the eye. Up to at least 22 weeks of age the outer two coats of the eye are elastic enough to give with the increased amount of fluid, while the retina being more fragile, merely ruptures.

There was a slight indication that adrenal size is reduced in birds with ophthalmacrosis. This agrees with the findings of Hogg (1967) who also pointed out that adrenal size was inversely related to adrenal output in birds with ophthalmacrosis.

Pituitary and thyroid weights were positively related to the amount of light received per day and not to the degree of ophthalmacrosis.

It was found that the addition of protamone to the diet, implantation with estrogen or castration reduced the severity of ophthalmacrosis while the addition of thiouracil to the diet of pinealectomy increased the severity of ophthalmacrosis;

however, these differences were statistically non-significant.

That the adrenal cortex is involved in the development of ophthalmacrosis is indicated by the findings that bursa weights were inversely related to the degree of ophthalmacrosis and also by the fact that sodium uptake by the ocular fluids was positively related to the degree of ophthalmacrosis.

There was a defininte relationship between photoperiod and body weight. Up to the time of sexual maturity the birds exposed to a continuous lighting regime were heavier than those exposed to a diurnal light cycle; however, as the birds approached sexual maturity the reverse was true.

Based on these findings, the following hypothesis is presented: ophthalmacrosis develops as a result of the lack of a diurnal light cycle by which the chicken can set its biological clock(s). This lack of a diurnal light cycle sets up a stress condition due to the absence of appropriate signals reaching the hypothalamic region of the brain. This results in the release of adrenocorticotrophic hormone and perhaps other as yet unidentified releasing factors that act on the adrenal cortex. This stimulated the release of the mineralo-corticoids of the adrenal cortex that act directly on sodium, potassium and water metabolism resulting in a general systemic upset due to improper mineral balance, improper acid and base balance and the adrenal hormones action on other endocrine organs, specifically the thyroids. Increased amounts of sodium and water enter the eye causing a physical stressing of the eye. The more delicate retina may tear after a significant amount of enlargement has occurred; however, even if retinal tears do not develop vision is undoubtedly impaired severely by the alteration of eye shape as well as the change in the make up of the ocular fluids.

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VITA

Robert Allen Voitle was born in Parkersburg, West Virginia, on May 12, 1938. He was educated in the elementary schools of that city and was graduated from Parkersburg High School in June of 1956. The following September, he entered West Virginia University, receiving a Bachelor of Science in Agriculture in January, 1962. He subsequently entered the Graduate School of West Virginia University, holding a graduate assistantship. In January 1964, he received his Master of Science degree. He entered The University of Tennessee Graduate School, February 1964, as an "Assistant in Poultry." In June 1969, he was granted a Doctor of Philosophy in Animal Science. He is a member of both Sigma Xi and Gamma Sigma Delta.

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