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THE ADRENAL GLAND
OF THE HOUSE SPARROW (PASSER DOMESTICUS)
AND ITS SEASONAL CHANGES

- by -

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ABSTRACT

The present paper is the study of the adrenal glands of the house sparrow (Passer domesticus) with special emphasis on seasonal changes. Prior to this paper hardly any investigations have been made to determine the relationship between the avian adrenals and the breeding cycle. A number of birds were trapped and subjected to artificial illumination, in addition to which birds were shot just outside of Edmonton. All sections were fixed in a bichromate solution and stained with Delafield's haematoxylin and eosin. In addition, a representative number of adrenals were fixed in osmic acid to determine the fat concentration and thus the physiological activity of the glands.

A general description is given of the adrenal gland of Passer domesticus, the first account (to my knowledge) of an adrenal gland of the house sparrow. A complete histological survey is also given of a series of experimental and wild birds, including the osmicated glands. In addition, a histological description of the testes of the experimental birds is given to correlate the investigation done on adrenal glands in relation to the breeding cycle.

By a "paper-weight" method the volume of

medullary and cortical tissues of a series of experimental and wild birds was calculated, tabulated (Table III and IV) and graphically presented (Graph I)

Conclusions are drawn from the histological, physiological and volumetric data obtained, and a summary given at the end of this thesis.

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INTRODUCTION

In recent years the extreme importance of the adrenal gland in the general metabolic and physiological processes of all higher animals is becoming more and more apparent. Especially in the last few years when the full significance of the adreno-cortical hormone, cortisone, and its stimulating hormone secreted by the anterior pituitary gland, ACTH, is being uncovered, investigators are becoming more aware of the incompleteness of their knowledge concerning the endocrine processes and their general relationship with the body processes. At the present time the whole of medical science is being revolutionized by the findings being disclosed about the connection between disease in general, both psychosomatic and organic, and the endocrine system.

The adrenals of all animals consist of two distinct tissues, the cortical and the medullary or chromaffin tissue. Each has its own pre-embryonic and post-embryonic history, histological structure and physiological functions. Just why these two entirely different types of tissue should be found together is quite obscure. Indeed, in certain fishes (Elasmobranchs) the analogues of these two tissues are separate.

The presence of the adrenalin - containing chromaffin cells has been demonstrated in invertebrates (most Annelids, Cephalopods, Cyclostomata and Molluscs) where they are in most cases situated in close connection with large vessels and nerve tracts. Cortical cells do not appear in these lower forms of life, being found only in animals higher than invertebrates. In Salaceans, unlike most other animals, the adreno-cortex (or "Inter-renal" body) is anatomically distinct from the chromaffin accumulation. In amphibia there is no distinct demarcation between cortical and medullary cells, the two being intimately mixed. In Reptilia and Aves the cortical and chromaffin cells are also irregularly mingled, there being no obviously distinct layers of cortex or medulla as in higher vertebrates. In Mammalia and other higher vertebrates the adrenal is arranged with the chromaffin tissue in the centre of the gland (hence the term "Medulla") and the cortex and outer enveloping rim of tissue.

Embryologically, the medullary cells and the sympathetic ganglion cells have a common origin in all animals.

The following is quoted from "Physiological Basis of Medical Practice " (2) by Best and Taylor:

"They develop from primitive cell masses which have separated from the neural crest. Migrating from their sites of origin these masses of ectoderm cells undergo differentiation along two paths, some into sympathetic ganglion cells, others into chromaffin tissue. In the abdomen on either side of the midline a relatively large mass of chromaffin tissue becomes enveloped by cortical cells to constitute the adrenal medulla. Other smaller masses persist as accessory chromaffin tissue in association with the ganglia and plexuses of the sympathetic. Such chromaffin collections are called paraganglia. On the other hand, sympathetic ganglion cells may be found scattered among the cells of the adult adrenal medulla. The cortex is developed from the mesoderm. It arises as a bud from the coelomic epithelium covering the inner side as the fore part of the mesonephros. The coelomic epithelium immediately behind this area gives rise to the germinal epithelium from which in turn the sex glands develop."

Adrenal Cortex

The cortex in a mammal is differentiated into three distinct zones. From the outside of the gland inward they are:

1. The zona glomerulosa - these cells are

arranged in a circular or oval pattern, i.e., cells are arranged in blocks; the cells are not frothy, therefore believed to be young cells.

2. The zona fasciculata - here the cells are arranged in long columns usually two cells thick. The columns are separated one from the other by blood sinuses which empty into a large central sinus in the centre of the medulla. These cells are frothy indicating an extremely active physiological function.
3. The zona reticularis - is composed of a network of cell cords. These cells are larger and stain more heavily than the cells in the zona fasciculata.

In castrated males and often in normal female mammals there appears between the zona reticularis and the medulla another zone referred to as Zone X. This zone is quite different from the cortical tissue or the chromaffin tissue and is believed to be due to a compensatory action of the adrenal gland to overcome a deficiency of androgen secretion.

The physiological activity of the cortical cells is indicated by lipid granules which are clearly brought out by specific fixing and staining methods involving

fresh-frozen sectioning and fat stains, or by fixing in an osmium tetroxide solution. In a paper written by A. B. Kar (22), the author quotes Flexner and Grollman (9) as suggesting that the lipoid content is low when the functional activity of the cortex is low and is increased when the secretory activity is supposed to be increased. Kar, in a later paper (23), concurs with the view that the increase in the lipoid material of the cortex is suggestive of a greater secretory activity on the part of the cortex. Although the proof of this statement is lacking, it is suggestive, but until further experimentation is conducted, it will remain unsubstantiated. Selye believes that "the lipoid granules in the cortical cells are not the hormones of the cortex, but consist mainly of cholesterol and neutral fats. Since, however, the cortical hormones are highly fat soluble (much more soluble in fats than in plasma) it is possible that the visible lipoid granules act as solvents in which the cortical hormones are stored."

The cortex of the adrenal, unlike the medulla, is essential to life. Removal of more than about five-sixths of this part of the adrenal causes death within a few days. Some twenty crystalline steroid compounds have been isolated from the adrenal cortex which exhibit in greater or less degree the physiological properties of crude cortical extracts. Corticosterone is related to

glyconeogenesis and muscle efficiency. Desoxycortico-sterone (which has lost the oxygen atom on C₁₁ found in corticosterone) is involved in the control of salt and water metabolism, that is, the conservation of water and electrolytes in the body. Besides these compounds, with actions specific for the adrenal cortex, steroids with actions of estrone, progesterone, and the male hormone such as testosterone respectively, have been isolated from the adrenal cortex. The last mentioned principle has been named adrenosterone.

Adrenal Medulla

The medulla is composed of closely packed groups of polyhedral cells with extremely ragged cytoplasm containing chromaffin granules which are looked upon as the mother substance of the medullary secretion. The cell groups are separated by blood sinuses which empty into a central vein.

It remains unexplained why, in all vertebrates except the elasmobranchs, the primordial cells of the medulla travel a long distance in order to unite with those of the cortex. All the blood reaching the medulla has to travel through the cortical sinusoids first; this suggests that products of the cortical cells may have to go through further transformation in the medulla, but such a possibility

is entirely conjectural. Since both cortical and medullary hormones play an important role in the general-adaption-syndrome (see Footnote), the demand for both types is essentially great during exposure to non-specific damage.

Footnote - The definition of the general-adaption-syndrome as given by Hans Selye in his text "Endocrinology" is as follows:

"Experimental and clinical observations have shown that in addition to the specific adaptive reactions (e.g. serological reactions to specific antigens, muscular hypertrophy subsequent to physical work, proliferation of the epidermis where the skin is exposed to pressure or friction) there are certain physiological mechanisms which help to raise resistance to damage as such. In these latter reactions the endocrine system plays a prominent part and they occur irrespective of the specific nature of the evocative damaging agent.

The sum of all these non-specific system's reactions of the body which ensue under long continued exposure to stress has been termed the 'general-adaptive-syndrome'. It is characterized by a number of morphological and functional changes. Among the most prominent of these are: enlargement of the adrenal

Perhaps the anatomic union of these tissues is advantageous because, during emergencies, relaxation of the sphinctors in the adrenal veins suddenly supplies the general circulation with large amounts of both cortical and medullary hormones stored in the gland's spacious sinusoids. Stimulation of the adrenal nerves elicits an immediate release of chromaffin granules into the bloodstream, without causing any detectable secretion of cortical lipoid granules. Conversely, denervation of the adrenals prevents the secretion of adrenalin, without interfering with cortical function, therefore secretions of cortical hormones are not significantly influenced by specific motor nerves, while that of adrenalin is almost entirely dependent upon nervous stimuli.

The medulla, as far as is known, secretes only adrenalin. In times of stress, for example, fright, rage, pain, asphyxia, exposure to cold etc., the medullary secretion, it is believed, reinforces the sympathetic nervous system. Through this hormonal-nervous co-operation of the several bodily reactions associated with such states of emergency are raised to maximum efficiency. This involves

"cortex with increased corticoid-hormone secretion, involution of the thymus and of other lymphatic organs, and certain metabolic changes and variations in the resistance of the organism."

a rise in general blood pressure accompanied by a dilation of the vessels of the contracting skeletal muscles, hyperglycemia which indicates demobilization of the carbohydrates stored in the liver, discharge of more erythrocytes from the spleen and thus a greater oxygen capacity, and other general conditions suitable for an emergency reaction.

REVIEW OF LITERATURE

It seems necessary, at this point, to review all investigation done on avian adrenals, which is very slight. Indeed, that is one of the chief reasons for undertaking this investigation. No work has been done on wild birds, as all previous investigations were confined to pigeons and domestic fowl. As very little has been done on a possible adrenal cycle (to my knowledge), this outline will be very general and will include investigations carried out on adrenals from the embryological, histological, physiological and other aspects.

E. O. Hohn (18), while investigating the seasonal changes in the gonads and adrenals of the mallard, noted that the adrenal glands of both sexes show no significant seasonal variations as regards weight. Histologically the females show a relative increase of cortical tissue at the height of the breeding season, while in males no significant season variations are observed.

V. J. Hays (15) investigated the origin and development of the adrenal gland in birds. His work showed that the origin of bird adrenals is similar to that of a mammalian adrenal, i.e. that the anlagen which gives rise to the cortical substance of the bird adrenal appear as groups of cells which migrate dorsally from the peritoneal

epithelium. The chromaffin substance is derived from in-different cells which wander in from the anlagen of the prevertebral sympathetic plexus. He found that the vessels of the arterial system are found almost entirely in the cortical substance, and that the entire venous system is derived from the subcardinal veins, the vessels inside the gland itself being sinusoid in character.

An investigation has been made by Miller and Riddle (29) into the cytology of the pigeon adrenal cortex. They state that the tendency for the cortical strands is to be composed of a double row of cells with their long axis in the transverse plane of the strand. One end of each columnar-like cell faces on the blood channels.. In the midst of the large masses the cells are usually longer in one axis than the other, but the orientation is not as clear. In this same paper they also try to show that since the cortical tissue of the pigeon adrenal is not a single, homogeneous structure, it is not surprising that it does not show the distinct zonation commonly found in the mammalian adrenal. However, the usual histological fixatives show certain gross differences between areas of the cortex. At the periphery the cells are large, contain considerable cytoplasm and full, round or slightly oval nuclei; the chromatin network is thin and diffuse, and there are usually two nucleoli, one taking the acid stain,

the other basic. Towards the middle of the gland the cells elongate and eventually become smaller. With the decrease in cell volume the nucleus decreases somewhat and the chromatin is more compact. Shrunken dying cells with pycnotic nuclei and cytoplasm can be seen in the centre of the gland. Because cell proliferation occurs at the periphery of the glands and cell death occurs at the centre, and because of further cytological evidence given in this paper, the authors believe that the current view of the life history of mammalian cortical cells as propounded by Salmon and Swemer (41), Walton and Zwemer (49) and others is valid also for the adrenals of pigeons. Following hypophysectomy they found that there was a loss of weight in the bird adrenals chiefly due to the reduced volume of cortical cells, and a smaller total number, diminished by death and a lower rate of replacement.

Sauer and Latimer (42) investigated sex differences in the proportion of the cortex and the medulla in the chick adrenal. They indicated that the female chickens have approximately thirty percent more adrenal cortex, in proportion to its body weight, than the male, although the gross weight of the glands is about the same in the two sexes. The amount of adrenal cortex is much more variable in the female than in the male, suggesting a physiological variation.

James C. Perry (36) doing research on the English sparrow, showed that birds brought to breeding conditions by artificial illumination, and normally breeding birds, if given adrenalin injections in one-half minim daily doses, would result in a regression to the resting juvenile condition of the gonads, accessory and secondary sex characteristics. The evident antagonism is interpreted as being directed against the secretion, release or activity of the gonadotrophic hormones of the anterior pituitary. The mode of action of the adrenalin antagonism, whether directly or indirectly through the nervous system or indirectly through changes in the metabolic rate has not been determined. As a possible explanation, Perry states that increased daily exposure to the spring daylight in the English sparrow brings about increased physiological activity. This increased physiological activity diminishes the available adrenalin in the blood, and as it falls below its gonadal antagonistic value, the anterior pituitary-gonad mechanism is suppressed. This explanation is quite unsound from a physiological viewpoint because only in a state of emergency are significant amounts of adrenalin secreted by the chromaffin tissue, and its physiological effects last only a short duration because of the rapid oxidation of this hormone in the tissue. This evidence of adrenalin gonadal antagonism is further substantiated by Perry (37) and by Wheeler, Searcy and Andrews

(51). Perry showed that subcutaneous daily injections of adrenalin given to female rats caused prolonged and repeated diestrus, follicular atrophy, reduction in absolute and relative ovarian weight, reduction in weight and size of the uterus and vagina and a decrease in the pituitary weight. Wheeler et al investigated the effect of adrenalin upon semen production in domestic fowl. Intramuscular injections of adrenalin was followed by an almost immediate decrease in semen volume with a forty-eight percent reduction at the end of a thirty day injection period. Volume rose to pre-injection levels following the cessation of treatment. The total number of sperm per ejaculation decreased fifty to seventy percent by adrenalin injection. Marked disintegration of the cells of the seminiferous tubules was present seven weeks after cessation of injections.

The relation between size of testes and requirement of cortical extract in adrenalectomized drakes was investigated by Bulbring (4). In adrenalectomized drakes, it was found that the sensitiveness of the bird to cortical deficiency changes. In spring much more extract is needed to keep operated birds alive than at other times of the year. The size of the testis at that time is also much greater. Average weight of the testis was determined for the different months and it was found that the weight of both testes in the winter is about three g. In February the weight increases, is considerably more in April and

reaches a maximum of 78 g in May, after which there is a rapid decrease to the lowest figure in July. When for each month the dose was calculated which kept adrenalectomized drakes alive for sixteen hours, it was found that the dose was low for winter (0.07 cc's per hour), while in April as much as 0.3 cc's per hour was needed, in May and June less and in July 0.05 cc's was usually sufficient. If these amounts of extract are plotted together with the weights of testes for different months, two parallel curves result. The weight curve of the testes reaches its peak in May, whereas the dose curve reaches its maximum one month earlier.

Parkes and Selye (35) showed in sub-total adrenalectomized birds, the feathers which grew back on the spot the operation was made were abnormal, much like the capon type of feather. This temporary change of the feather to capon type after sub-total adrenalectomy is evidence, in their view, of testes disturbance and may be taken to indicate that the adrenals, possibly by gonadotrophic hormones, play some part in maintaining testis activity.

Hartman and Knouff (13) have examined chromaffin patterns in bird adrenals. They found that in young birds, the chromaffin tissue cells formed only a few cell groups which were connected by means of almost imperceptible bridges.

With increasing age the chromaffin tissue increased and showed a strand-like arrangement. In old birds the chromaffin cells might form a more or less distinctly connected peripheral border due to their increased number. The pattern of the chromaffin tissue depends somewhat on the relative amounts of this tissue in the gland. When small it is arranged in distinct islets scattered through the interrenal tissue. As it increases in amount, the islets become larger and often elongated, tending to unite. If a relatively large proportion of chromaffin tissue is present, it may form a meshwork throughout and also cover appreciable amounts of the surface of the gland. Characteristic patterns may be found in certain species. However, there may be considerable variation in the amount of chromaffin material among species of the same genus.

Kar (22) investigated the action of male and female sex hormones on the adrenals of fowl. Injections of diethylstilbestrol into adult female fowl failed to produce any effect on the adrenal weight. This treatment caused an increase in size of the vacuoles of the cortex. Testosterone propionate treatment was without any effect on the adrenal weight of adult female fowl. A decrease in the size of the vacuoles of the cortex ensued as a result of this treatment.

Herrick and Torstveit (16) did research on the effects of adrenalectomy in fowl. The glands were destroyed

by cauterizing. Operated birds were maintained for eighty days by giving injections of cortical hormone and continued treatment with sodium chloride. Birds receiving no treatment died in six to fifteen hours after complete adrenalectomy. The combs of adrenalectomized chickens became greatly reduced in size. Changes appear as early as the second day after complete adrenalectomy. The testis of operated birds also became smaller. The histological structure of the testis was altered, indicating marked degeneration, and the walls of the tubules were so broken down that the lamina were obscure. The authors believe that the adrenal glands have an important influence in maintaining the testes of chickens. Experiments have not as yet indicated the nature of this influence, or whether it reaches the testis directly or indirectly. Likewise, it has not been determined whether the influence on the comb is due to lack of adrenal secretion directly, to reduced testicular activity, or to some other indirect effects of adrenalectomy. The changes in the comb begin so soon after adrenalectomy, at which time the testis is still large, that it is questionable whether reduced testicular activity is the entire answer.

Hewitt (17) found that bilateral adrenalectomy in the adult chicken produces atrophy and degeneration of the testis and the external appearance of the bird tends

towards that of a capon. In the hen of the same species and variety the presence of the adrenal cortex seems necessary for the conversion of the rudimentary right gonad to a testis-like organ following ovariectomy. In bilaterally adrenalectomized birds this conversion did not take place.

Uotila (48) noted that two cytomorphic states --the fuchsinophile and pale cells respectively can be regularly and clearly differentiated in the adrenocortical tissue of fowl when stained with Broster and Vine's ponceau fuchsin or Masson trichrome stain after Orth's fixation. Cytological criteria suggest that the pale cells are precursors of active fuchsinophile cells which in turn become converted into degenerating fuchsinophile cells. The number of fuchsinophile cells and the strength of the fuchsinophile reaction is greater in males than in females, and greater in immaturity than in adult animals. Androsterone (and to a lesser extent estrin) suppress this reaction in males but not in females. Several anterior pituitary extracts failed to produce any definite changes in the fuchsinophile reaction. These observations afford no definite proof that the fuchsino-ophile reaction is specifically correlated with the alleged masculinizing function of the adrenal cortex.

A. G. Kar, in another paper (24) again connected

with the adrenal cortex-testicular relations in the fowl indicated that the adrenal cortex is greatly hypertrophied after castration. This hypertrophy can be prevented by administration of testosterone propionate or diethylstilbestrol. The sex difference in proportion to the adrenal cortex is neutralized by castration. Further, castration increases the lipid content back to normal again. The cytological evidence shows that the so called fuchsino-philia are nothing but mitochondria. The mitochondria do not take any part in the formation of the cortical lipid. The result of histo-chemical tests indicate an increase in the cortical hormone level in hypertrophied cortex, and it is suggested that the increased amount of mitochondria in the castrated cortex plays some unknown role in transforming the cortical sterols into androgens. Kar finally advances a hypothesis according to which one of the functions of the hypertrophied cortex in the castrated fowl is to provide a greater amount of androgen in order to take over the extra-sexual functions of the gonad.

Miller and Riddle (28) investigated the cellular response to insulin in adrenals of pigeons. In tests made on forty-one normal white Carneau pigeons, daily injections of one to sixty units of insulin during ten hours to ten days, enlarged the adrenal, produced mitosis and cellular activity. These cytological effects were more pronounced

in cortical than in medullary cells. Wholly comparable effects were produced in the adrenals of sixteen other birds, previously deprived of their anterior pituitary glands. The cytological changes in the cortex following insulin are identical with those produced by adrenotrophic pituitary extracts.

The relationship of these results to the "alarm reaction" is discussed by the authors. They pointed out that the work of others has shown that the mammalian adrenal reacts to toxic substances and various operative procedures by enlarging, one part of a syndrome which Selye calls the "alarm reaction". On Selye's view, (45) such adrenal enlargement is a secondary result of the action of the alarming stimulus upon the pituitary. He obtained no adrenal enlargement in hypophysectomized animals subjected to "alarming" stimuli. In the studies of Miller and Riddle cortical enlargement and loss of chromaffin granules in the medulla, duplicate changes associated with the "alarm syndrome". It is entirely certain, however, that following insulin these particular changes occur in pigeons deprived of their anterior pituitary with the same facility as in the intact animals. Thyroxins and estrogens are also effective in stimulating adrenals in both normal and hypophysectomized pigeons--but insulin has marked ability to enlarge the adrenal, notably in hypophysectomized birds.

Since the adrenal is involved in carbohydrate metabolism, it seems more than coincidental that this gland, particularly cortex, responds especially well to insulin, which suggests that both cortical and medullary parts of the adrenal actively oppose the ability of insulin to decrease the blood sugar, and that this occurs in hypophysectomized pigeons.

MATERIALS AND METHOD

This investigation was prompted by research done by W. Rowan (40) and others, on the sexual cycle of juncos and Starlings and the induction of a complete sex cycle prematurely by the use of artificial illumination. Little work has been done on bird adrenals in general, and nothing at all on the problem of a possible cycle in the adrenals of wild birds analogous to the sexual cycle. Nothing has been done to try to integrate the activity of the bird adrenal with the functions and activities of the other endocrine glands. Therefore, the problem arose--is there a cycle in the adrenal of the resident wild bird and if so what relationship has it to the breeding cycle and to the endocrine system of the bird as a whole? Because of the necessary delimitation involved in any investigation only the cyclic aspect of this problem will be emphasized.

The bird chosen for this investigation was the house sparrow (Passer domesticus). The reasons for this selection were twofold. Firstly, a wild bird was desired in contrast to domesticated or semi-domesticated birds such as chickens and pigeons used in most experiments cited. Secondly, the house sparrow, being a permanent resident of this area, is thus much more easily accessible to capture or shooting. These birds are quite well suited for this type of experiment because of their size, because

of their hardiness and their feeding habits, although they have one serious drawback which was clearly manifested, and that is their extreme nervousness and irritability. This objection was outweighed by their availability.

A very primitive arrangement was used to capture these birds. The trap was a simple wooden frame covered with wire which was propped up by a stick to which was attached a long piece of string. In this way, the trap was dropped on sparrows feeding underneath. About twenty-five sparrows were captured in this manner and placed in four cages, approximately six birds to each cage.

Illumination experiment

The procedure used in this experiment is very similar to that used by W. Rowan (40) in his migration and breeding experiments. The purpose in using this procedure was to create the complete breeding cycle prematurely by increasing each day the amount of light to which the birds were exposed.

The illumination experiment was commenced in January, 1950. The cages in which the birds were kept were covered in front and on top by wire, the sides and back being of wood. The illumination was produced by 100 watt fluorescent bulbs which were about two feet above the cages.

The temperature in the room remained constant at 24 degrees Centigrade and the birds were kept well fed on millet, oats and rape.

Lighting was commenced on January 9. For the first several days the birds were exposed to ten hours of light, after which the illumination time was increased by ten minutes per day. Under normal conditions the day lengths at this time of the year are increasing at the rate of about three minutes per day, so that these birds were getting an abnormal increase in illumination of approximately seven minutes per day. This was kept up until the fifteen hour limit was reached. At this time the birds were kept at fifteen hours for about two weeks, following which the lighting was cut down by fifteen minutes each day. A bird was sacrificed twice a week or after an increase in illumination of approximately one-half hour. Due to the fact that it was very difficult to trap sparrows because of their nervous nature, not enough birds were trapped to be able to keep a large enough batch of both experimental birds and controls. Therefore, all the birds were kept as experimentals. As a substitute for this lack of a control batch, birds were shot in the open at fairly regular intervals. Wild birds can be considered to be legitimate controls, except that they were not confined to cages.

Method

The adrenals from both experimental individuals and the wild birds were removed immediately after killing so that degeneration of these glands after death was kept to a minimum. Also removed with the adrenals were the gonads, thyroid and pituitary glands. After removal the adrenals were fixed in Regaud's fluid. This fixative consists of four parts of three percent potassium bichromate with one part of forty percent formalin (formaldehyde). After twenty-four hours in this solution, the glands were put in a three percent potassium bichromate solution for seventy-two hours. This was followed by washing in running water for twenty-four hours. Regaud recommended four days fixation followed with eight days further chromation in three percent potassium bichromate for mitochondria, but this is excessive because it interferes with nuclear staining. Dehydration was next carried out by moving the material from fifty percent to one-hundred percent alcohol and then into chloroform, and finally into wax. It was soon found that the chloroform hardened the tissue too much, particularly the testes, so that chloroform was replaced by xylol which was found much more satisfactory.

The wax blocks were then cut on the microtome into sections ten μ thick and mounted on slides. Serial sections were made in all cases. All sections were stained

with Delafield's haematoxylin and eosin to produce a uniform set of slides, therefore any differences cannot be attributed to a varied method of staining.

Some of the glands were fixed for fat staining. The method chosen in this case was used by Flexner and Grollman (9). The fixing solution consisted of one part twenty-five percent neutral formalin, nine parts Zenkers without acetic acid and one and one-half parts of two percent osmic acid. The tissue was kept in this fixative for forty-eight hours (renewing it after twenty-four hours), rinsed in six changes of distilled water, put in the two percent osmic acid for twenty-four hours, and then washed in water for one-half hour with numerous changes. Dehydration was then effected by the usual method, although the tissue was kept in each change of alcohol for a very short time so that the fat should not be washed out. After the alcohols, the tissue was put through a solution of one half absolute alcohol and one half xylol (for fifteen minutes) then xylol (for fifteen minutes) and finally embedded in paraffin. Serial sections were cut at six mu and mounted on slides. Several slides were stained with Vine's Ponceau stain as a trial (Archives of Pathology 1939;27:1030). This method proved to be unsatisfactory because when the sections were washed with xylol and the cover slips fixed over them, the fat was completely dissolved out of the sections. As a result, the following method

was used: the wax was dissolved out by placing the slide in xylol for thirty seconds and following this, the sections were run through the alcohols very quickly and into water, the cover slips being mounted on the slides with glycerine jelly. In this way the fat concentration was well preserved due to the absence of fat solvents in the process. The effects of xylol were very slight as the sections were kept in this solution for a very short period of time, and the weak effect it did have was cancelled out because all slides were similarly treated.

HISTOLOGICAL STUDY OF THE SPARROW ADRENAL

General Description of the Adrenal of the House Sparrow

As has been stated before, the adrenal gland of all higher vertebrates is composed of two distinct types of tissue, the cortical and medullary (or chromaffin) tissue. In birds, these terms do not refer to the position of these tissues in the gland. In the sparrow, the cortical tissue is found throughout the gland, with the groups of medullary tissue interspersed everywhere (See Plates I to V). Generally speaking, the medullary tissue lies more to the centre of the gland with the cortical masses usually larger at the periphery. In some cases the medullary tissue lies in thin strands immediately adjacent to the capsule except at scattered places where a strand of cortex touches the capsule. The capsule itself is a very loose and poorly constituted sheath of connective tissue around the gland. In many cases it is poorly defined and is not clearly distinguishable from the peripheral cortical cells. It is generally accepted that there is a differentiation of capsular cells into cortical cells, but special techniques would be required to demonstrate this in the present case, e.g., by trypan blue injections (41) and not enough material was available to investigate this line of study.

The blood vessels in general are distributed along the periphery of the gland--in the capsule or just at the edge of the cortical tissue. From these, capillaries are given off which form an intricate network throughout the whole gland, between the strands of cortical and medullary cells. These capillaries finally radiate in the direction of a central sinus situated approximately in the centre of the gland, which in turn forms the efferent vein.

The cortical cells are found in several configurations, but there is a tendency for the cortical strands towards the centre of the gland to be composed of a double row of cells with their long axis in the transverse plane of the strand. One end of each columnar-like cell faces on the blood channels. In many cases the cell walls are very difficult to distinguish and the cells are usually longer in one axis than the other in these large masses. Around the periphery the cortical cells have a more glomerular configuration, that is they form a circular group of cells like an acenous without the inside lumen. These cells do not have such a long axis as those cells found in the long strands. Some cortical cells are not found in any set configuration but are an amorphous group of cells.

In many of the glands a large number of mitotic figures were seen at the periphery just underneath the

capsule, while few mitotic figures were observed internally. It is now generally accepted that mammalian cortical cells arise in the peripheral portion of the gland, migrate inward towards the medulla, become senescent and disappear in the inner zone of the zona reticularis. New cortical cells are formed by mitosis and by the differentiation of certain cells in the capsule. Thus the observation of many mitotic figures at the periphery tends to confirm this hypothesis.

Since the adrenal cortical tissue of a bird is not a homogeneous area as in mammals, it is not surprising that it does not show the distinct zonations found in the mammalian adrenal. However, in the sparrow the usual histological fixatives show certain gross differences between areas of the cortex. At the periphery the cells are large, contain considerable cytoplasm and full, round or slightly oval nuclei with usually two nucleoli. The chromatin network in the nuclei is thin and diffuse. This area I refer to as Zone Y. In this area there is usually little medullary tissue present and when found is only a thin strand one to two cells deep just inside the capsule. The Zone Y does not usually run all the way round the gland. Quite often it is crescent shaped or may be found only in one corner of the gland. Towards the middle of the gland the cells elongate and eventually become smaller. With

the decrease in cell volume the nucleus decreases somewhat and the chromatin is more compact. Shrunken, dying cells with pycnotic nuclei and thin diffuse cytoplasm can be seen in the centre of the gland.

The medullary cells are quite irregular in shape arranged in rounded groups or short cords in an anastomosing network beside which is usually found a capillary. The cells are polygonal and are smaller than those of the cortex. When the tissue is fixed in a fluid containing potassium bichromate, very fine brown granules may be seen distributed throughout the cell bodies. This constitutes the "chromaffin" reaction characteristic of chromaffin tissue, which elaborates adrenalin. In the mammal these cells are closely associated with myelinated nerve fibres, but in the sparrow very few nerve cells were observed, although a large nerve tract adjoining the adrenal was seen in practically all cases.

As to the average volume of the medullary and the cortical tissues, an attempt will be made later in this paper to show that these vary with the seasons. Miller and Riddle (29) took the average of a number of measurements in the pigeon adrenal which indicated that sixty-five percent of the gland is cortex, thirty-two percent medulla and three percent blood space, lymphoid tissue etc. In that paper they did not even consider seasonal changes,

and I will try to show that this ratio of volumes changes with the seasons. They also did not find any difference between males and females such as Sauer and Latimer (42) reported for adult fowl.

HISTOLOGICAL OBSERVATIONS OF A SERIES OF ADRENALS

The glands from illuminated birds will be described separately from those of the birds shot outside.

Illuminated Birds

Bird #11 - Eleven Hours - Female

No marked Zone Y seen. The cortical cells are quite small and closely packed together. The cytoplasm is not frothy and there is no regular arrangement of the cells, i.e. they are not arranged in columns. The nuclei are quite pale and open. The medullary cells are irregularly shaped, with nuclei quite obscure. The blood sinuses are not noticeable except for a few small ones at the periphery just under the connective tissue capsule.

#114 - Eleven Hours and Forty Minutes - Male

No marked Zone Y is clearly discernable. The cortical cells are arranged in columns while those cells near the periphery are more in glomerular bundles. The blood sinuses at the side of each bundle are quite noticeable--they lead into the large central sinus found typically in most of the glands examined. The cortical cells are small and closely packed, but the cytoplasm is more frothy than the eleven hour gland. The medullary cells are polygonal with light nuclei.

#115 - Twelve Hours - Male

No Zone Y seen. The peripheral cortical cells are not arranged in columns as are the more central cells. Also the latter cells are larger and frothier than the peripheral cortical cells. There is no change in the medullary cells.

#119 - Twelve Hours and Forty Minutes - Male

Zone Y is clearly distinguishable. The cortical cells at the periphery are arranged in an indifferent manner. They are larger than the internal cells, stain deeper and the cytoplasm is much less frothy than those cells found closer to the centre of the gland. The nuclei in general seem to be deeper staining than those in the previous specimens. The nuclei of the Y Zone cells are more pale, less granular than the nuclei of the more active cells.

#122 - Thirteen Hours and Thirty Minutes - Female

Similar to #119.

#123 - Fourteen Hours - Male

Zone Y is clear and distinct. The cells in this area are larger, deeper staining (cytoplasm) and not frothy. Nuclei are also not granular. This raises the possibility that these cells are young, which when younger cells are formed peripherally to them are pushed in towards the centre, and as they mature

they become physiologically active. Another possibility is that they have a different physiological function from the other cortical cells. There are numerous blood sinuses present in this gland, peripherally and internally.

#124 - Fifteen Hours - Female

The cortical cells in the Y Zone are thoroughly permeated with large vacuoles. The cells in the centre are frothy but not vacuolated. The blood sinuses are very prominent with the capillaries observable all through the gland, and the drainage very marked by radiating towards the central sinus. This gland has thus every sign of being physiologically active.

#133 - Fifteen Hours - Male

Zone Y is very clear. There are no large vacuoles anywhere, the central cells of the cortex are very frothy and the numerous capillaries radiate towards the central sinus. The medulla is in smaller patches than before.

#134 - Fifteen Hours - Female

Zone Y distinctive. Vacuolization is quite widespread throughout most of the cortex except in most of Zone Y. The nuclei of the vacuolated cells are smaller and quite granular in contrast to the larger

and paler nuclei of the Zone Y cells. The medullary patches in these sections (not quite as positive in the other fifteen hour glands) are largely pushed in towards the centre of the gland. Thus it seems to take on more the configuration of a mammalian adrenal gland, although the central area in these sections are still permeated with cortical tissue. Very little medulla is found in the Zone Y, which in this case extends all the way around the gland. As in #129, the blood capillaries are very conspicuous.

#140 - Fifteen Hours - Male

This bird died during the night and the adrenal glands were removed in the morning, approximately eight hours after death. Large vacuoles are seen throughout the whole of the outer portion of the cortex. The Y Zone is not clearly demarcated from the rest of the cortex, probably because of vacuolization. The inner cortical cells are not very frothy.

#141 - Fourteen Hours and Forty-five Minutes - Male

In this specimen the Y Zone is not very prominent. The cytoplasm of the cells is very frothy and capillaries are very prominent all through the gland. The medulla seems to be broken up into smaller patches than in previous specimens. The capillaries here are most prominent close to medullary patches and seem to

radiate out from these patches, through the strands of cortical cells, to the central sinus. This is probably because the active cortical cells crowd out the capillaries which look like slits between the columns of cells.

#155 - Twelve Hours - Male

There is slight vacuolization, or frothiness, in the area just inside, and partially including Zone Y, which is clearly differentiated. There are little or no medullary cells found in this area. Frothiness in the inside cells not as noticeable as in the fifteen hour glands.

#157 - Eleven Hours and Thirty Minutes - Male

Zone Y is very evident and heavily vacuolated, while the central cells are frothy. The cells in the Y Zone are more regularly arranged (i.e. in columns) than in the other previous specimens. The nuclei are pushed up against the back walls of the cells. In some areas the Y Zone cells are perfectly normal. In these latter cells the nuclei are normal, i.e. quite pale and open, while in the vacuolated (active) cells the nuclei have become granular (Pycnotic) in many cases, much the same as the cells in the central area. This may be an indication that the Y Zone is an area of largely immature cells which become more mature as they move towards the centre of the gland.

#158 - Ten Hours and Thirty Minutes - Male

A layer of medullary cells one to three cells thick runs as a sort of capsule around the outside of the gland just inside the connective tissue capsule. A great number of arterioles are also seen just outside the capsule, some long and narrow, others relatively small. Internal cortical cells are quite frothy in one area, but general frothiness not as prevalent as in previous glands. The Y Zone is distinct. Except for the thin layer of cells on the outside, the medullary tissue is in patches radiating towards the centre of the gland.

#159 - Ten Hours and Fifteen Minutes - Female

Vacuolization is seen in the Y Zone. Internally the cells are frothy and the capillaries are very prominent.

#160 - Ten Hours - Male

Cells in all areas extremely frothy and even vacuolated in spots, except in one large area of the Y Zone. Froth is even all through the gland except in the Y Zone.

#161 - Ten Hours - Female

Y Zone quite wide and cells arranged strikingly in columns. Central area slightly frothy. Medulla radiates towards centre of gland. Extremely vascular, but not so in Y Zone which is found only on one side of the gland.

#162 - Ten Hours - Female

Frothiness quite marked and vacuolization prevalent in peripheral regions other than Y Zone, which is not prominent. Capillaries very evident.

The above histological description of experimental birds indicates a general trend, in the adrenal glands of both the male and female, towards physiological activity paralleling the breeding cycle.

Wild Birds

The majority of these birds were shot in the vicinity of Edmonton, while some were caught in a mouse trap. The date indicates the day these birds were killed and the number of hours indicates the day length of those specific dates. These day lengths were obtained from the Edmonton Meteorological Station and were calculated from the daily sunrise and sunset. In most cases the adrenal glands were removed within an hour after the bird was shot. All the glands were treated in the manner described previously.

Histological Observations

#107 - December 14 - Seven Hours and Thirty Minutes - Female

The cortical cells are not frothy. No Zone Y is

evident and the cell arrangement is indifferent except in one corner where the cells are arranged in columns. The nuclei are large and quite pale, no sharp granulation being present. The capillary system is completely lacking.

#108 - January 8 - Seven Hours and Forty Minutes - Female
Cortical cells are the same as in #107. The medulla is in large masses throughout the gland. These masses are in long outstretched shapes.

#112 - January 21 - Eight Hours and Twenty Minutes - Male
Similar to the two specimens above.

#116 - January 21 - Eight Hours and Twenty Minutes - Male
No Y Zone visible. The cortical cells are the same as above with slight frothiness in the central regions. The medulla is in large masses especially along the edge of the gland. The capillary system is becoming more prominent than previously.

#117 - January 29 - Eight Hours and Forty Minutes - Female
Zone Y has appeared in small areas around the periphery. The central cortical cells are much more frothy than in #116, and the nuclei are pycnotic. Most of the medulla is at or near the edge of the gland, and very little of it is in the central area.

#125 - February 15 - Nine Hours and Forty-five Minutes - Female
Cortical cells frothy in central area and no Y Zone
is apparent.

#127 - February 15 - Nine Hours and Forty-five Minutes - Male
The medulla is extensive and lengthy, large patches
appear prominently. The Y Zone is absent. Generally
the cortical cells are in a glomerular configuration,
although some are in the columnar arrangement.

#131 - February 22 - Ten Hours and Fifteen Minutes - Male
The Y Zone is definite in this gland. Central cells
are not in columns but more glomerular in arrangement.
Frothiness not too apparent, with the capillaries
mostly at the periphery. The medulla is in small
patches scattered throughout the whole of the gland
except in the Y Zone.

#137 - March 1 - Ten Hours and Forty-five Minutes - Male
Zone Y present along one side. Fuzziness not extensive
in inside cortical cells - similar to #131.

#138 - March 1 - Ten Hours and Forty-five Minutes - Female
Cortical cells not frothy but gland very vascular.
The medulla is in small patches.

#144 - March 8 - Eleven Hours and Fifteen Minutes - Male
Y Zone clearly distinguishable from the rest of the
cortical cells. The central cells are quite frothy

and the nuclei are becoming pycnotic. These cells are arranged in long columns or in a type of syncytium, i.e. nuclei with no cell walls visible. The medulla is in very large masses, the cells being more granular than usual possibly indicating physiological activity.

#149 - March 15 - Eleven Hours and Forty-five Minutes - Female
Y Zone quite clearly distinguishable. Medulla is fairly extensive. The gland in general is very vascular. The central cells are conspicuous in long two-celled columns and appear frothy.

#150 - March 15 - Eleven Hours and Forty-five Minutes - Male
Very similar in nature to #149. The medullary groups of cells are smaller in the centre of the gland than further towards the periphery, probably because the central cells are active physiologically.

#156 - March 22 - Twelve Hours and Forty-five Minutes - Male
Central cells are very frothy with some slight vacuolization in the Y Zone which is not too clearly distinguishable from the other cortical cells. The medulla is in small patches hardly discernable at the extreme periphery. The capillaries are quite prominent.

#165 - June 1 - Sixteen Hours and Thirty Minutes - Female
Vacuolization quite marked. What appears to be the Y Zone runs across one end of the gland, i.e. does not run along the edge of the gland. The medulla is

generally in long strands a few cells deep. The gland is very highly vascular. Vacuolization is most marked around the edge of the gland.

#167 - June 9 - Sixteen Hours and Fifty Minutes - Male

The Y Zone is not clear and the gland in general does not appear to be physiologically active. The central cortical cells are very slightly frothy, if at all. No capillaries are visible at all.

#168 - June 20 - Seventeen Hours - Male

Zone Y distinct in several areas of periphery. This specimen is quite vascular although the cells do not indicate a great deal of activity. Only in one area are the cortical cells comparatively frothy.

#170 - June 28 - Seventeen Hours - Female

This gland is very vascular. The cortical cells are similar throughout the whole gland, i.e. there is no Y Zone as distinguished from the inner cells. These cells do not appear to be active at all. The medulla is in large patches.

#171 - July 6 - Sixteen Hours and Fifty Minutes - Juvenile

There is an unusual amount of medulla in this gland. One small area of the cortex, triangular in shape, is the Y Zone. In general this specimen is very vascular and the cortical cells are quite frothy.

The cells in these frothy areas are similar to those in the adult, i.e. the cell walls are not visible, the cytoplasm is stringy and the nuclei, pushed up against the back of the cell, is quite pycnotic. The cells in the Y Zone resemble those in the adult.

#172 - July 6 - Sixteen Hours and Fifty Minutes - Male

Very similar to the juvenile described above. It is very vascular and the medulla is quite extensive, but the cortical cells appear very inactive.

#173 - July 19 - Sixteen Hours and Twenty-five Minutes - Female

Y Zone is at one end of the gland and the cortical cells in the centre are in long columns and are quite widely separated one from the other. This gland appears to be quite active although very few capillaries are visible.

#174 - July 19 - Juvenile

The Y Zone along one edge of the gland clearly shows the differentiation between these cells and those closer to the centre. There is quite a large amount of medulla, but not as much as that found in #172, and the capillaries are not very obvious.

#176 - July 22 - Sixteen Hours and Fifteen Minutes - Male

The medulla is very extensive and found in large groups. This gland is quite vascular and the central

cortical cells, arranged in long narrow columns in some places, appear fairly active. These central cells are not as spread out as those in the female of July 19. A Y Zone is clear although these cells are smaller than those in other glands.

#180 - August 29 - Fourteen Hours - Female

This gland is fairly vascular but inner cortical cells do not appear active. The medulla is in large, stringy groups, with rows four to five cells thick running along the periphery of the gland. The Y Zone is present.

#182 - October 4 - Eleven Hours and Thirty Minutes - Female

Medulla is in large masses. The peripheral cortical cells are in columns and in acenous arrangements while the central cells, which are not active, are in an indifferent arrangement. Most of the cells in this gland resemble the Y Zone cells in previous specimens.

These observations are similar to those made on the experimental birds, i.e. that a general physiological cycle is present in wild birds, with the cycle reaching its peak approximately at the same time that the apex of the breeding cycle is reached.

Testis Development In Exberimental Birds

The close relationship between the breeding cycle in the sparrow and the variation in day-length, naturally or artificially produced, has been adequately proven by W. Rowan. In this illumination experiment the procedure used, closely follows that used by the above investigator in his papers (40). As this description of the seasonal development of the gonads is placed in this thesis to show the parallel between the breeding cycle and adrenal changes, a complete account will only be given of the testis development in the experimental birds. The testes were fixed in a formol saline, cut in sections ten mu thick and stained with Delafield's haematoxylin and eosin.

Observations of Testis

Eleven Hours and Forty Minutes

The testis was about 0.9 mms in diameter, (measured along the long axis). This specimen is essentially in the winter stage. The tubules are small and spermatogonia are seen along the basement membrane. The lumina are full of debris. No interstitial cells are observed and the interstices are entirely filled with thick connective tissue fibres arranged in layers around the tubules.

Twelve Hours

The testis has started the process of recrudescence,

having a diameter of just over 1 mm. The tubules are still small and lumina largely with debris. The spermatogonia are more numerous and a few have moved towards the centre of the tubule. These spermatogonia are larger than those against the basement membrane. A few mitotic divisions were observed. The connective tissue in the intertubular spaces is thinner than that in the Eleven hours and forty minute testis, but interstitial cells were not observed.

Twelve Hours and Forty Minutes

The testis is much the same stage as the twelve hour testis. The interstices are more open but it is hard to distinguish interstitial cells in these sections, although a few were observed. There are more spermatogonia near the centre of the tubules and they are conspicuously larger than those adjacent to the tubule wall. Debris in the lumina is still prominent.

Fourteen Hours

The testis is considerably larger than the twelve hour and forty minute specimen, having a diameter of about 2.5 mms. The tubules are much enlarged and the interstitial tissue is greatly developed. Many degenerating spermatogonia are observed in the centre of the tubules. Spermatocytes can be observed farther in from the basal layer of spermatogonia. Mitotic figures are seen in this area.

Fourteen Hours and Twenty Minutes

The diameter of the testis is 3 mms. It is similar to the fourteen hour testis, but spermatocytes are more plentiful and a few spermatids are observed. Tubules are much enlarged and the interstitial tissue seems to be pushed more towards the triangles between the tubules.

Fifteen Hours

The testis diameter is 5.5 mms. The tubules are so large that most of the interstitial tissue is almost completely crowded out. All the stages of the germ cells are observed except that there are few mature sperm. The metamorphosing spermatids can be clearly seen. The cells of Sertoli are very vague.

Later Fifteen Hour Testis

The diameter is about 6.8 mms. Tubules are very large and no interstitial tissue is seen. Mature sperm are very conspicuously arranged in groups and the spermatids are also numerous. The cells of Sertoli are still not very distinct. The lumina of the tubules are now quite large with no debris present. Before these birds were killed a smear was taken of the extruded cloaca and no sperm were manifested. When these same birds were killed minutes later and smears made, the sperm was voluminous. Perhaps by killing them a certain nervous mechanism was set off which enabled the sperm to be ejected.

Illumination Decreasing

Fourteen Hours and Forty-five Minutes

The testis measured 4 mms. Signs of degeneration are quite clear. Mature spermatozoa in a degenerating condition can be seen in the lumina together with metamorphosing spermatids.

Thirteen Hours and Fifteen Minutes

The diameter is 3.5 mms. Spermatocytes are observed in active degeneration and the lumina are being clogged with debris.

Twelve Hours

The diameter is about 2.5 mms. Disintegration is general throughout the whole tubule. The lumina of the tubules are choked with waste material, degenerate cells etc. These degenerate cells seem to agglutinate.

Eleven Hours and Thirty Minutes

Much the same as the twelve hour testis. Tubules are smaller and becoming like a winter testis.

Ten Hours and Thirty Minutes

About 1.8 mms in diameter. Very much like winter testis. Interstitial tissue becoming more prominent. Tubules are much smaller than before.

Ten Hours

About 1.5 mms in size. Similar to ten hour and thirty minute testis.

Testes of Wild Birds

These testes underwent essentially the same cycle as that described in the experimental birds, so that it will not be repeated here. But the development of the testis of the wild birds during the later winter and early spring seems to be a step ahead of the experimental birds. For example, the size of the fourteen hour and twenty-five minute testis of the wild bird is about 3.5mms. Only after the experimental birds were kept at fifteen hours for about two weeks did the testis reach its maximum size. This indicates that there was a lag in development of the experimental testis due to the abnormal increase in illumination, i.e. the testes did not develop fast enough to keep up to the increase in illumination per day.

OBSERVATIONS ON OSMICATED ADRENALS

The process used in treating these adrenals has been described previously (page 28). These adrenals were cut at 6 μ . The medulla remained colorless while the cortical tissues stained densely.

Adrenals of Experimental Birds

#115 - Twelve Hours

Droplets heaviest at the centre of the gland and quite light around the periphery. The droplets in central cells are mostly discrete and in general fat concentration is not very heavy (see Plate VI).

#122 - Thirteen Hours and Thirty Minutes

The osmicated fat concentration is very thin at the centre and heavier at the periphery. In general this gland is quite lightly impregnated. The fat granules are mostly discrete but around the edges they tend to run together into larger masses of fat (see Plate VI).

#128 - Fifteen Hours

This gland is very heavily impregnated with fat. In the central cells the fat globules have amalgamated to form large masses. Going towards the periphery the particles become more distinct and discrete, nevertheless large masses are also observed near the periphery. The concentration of osmic-reactive steroids is heaviest in the central area of the gland (see Plate VI).

#133 - Fifteen Hours

The concentration of fat in this specimen is also very heavy. At the extreme periphery fat globules are thin and discrete. This area is no doubt the Y Zone.

Illumination Decreasing

#152 - Thirteen Hours and Fifteen Minutes

Heavy concentration in the centre of the gland. As in all other sections examined, at the periphery the fat droplets thin out considerably to the point where it is found in very small specks at the extreme periphery (see Plate VI).

#155 - Twelve Hours

The concentration of fat in central cortical cells is medium and falls off considerably towards the periphery where the fat droplets are mere specks. In the central area of the gland some of the fat droplets have merged, but most of them are discrete (see Plate VII).

#160 - Ten Hours

The concentration of fat droplets in this gland is quite evenly distributed over most of the gland. In general the fat concentration is quite thin throughout. In one area of the periphery the droplets are very few and fine. This area is most likely the Y Zone. The rest of the periphery is perhaps more heavily invested with steroid than are the internal cortical cells (see Plate VII).

Adrenals of Wild Birds

In the case of the wild birds, unfortunately some of the fixing did not take properly, due probably to the fact that the osmic acid became ineffective. Consequently only the initial part of this stage of the work was satisfactory.

#127 - Nine Hours and Forty-five Minutes

The central area has a medium concentration of fat droplets. Again towards the periphery the concentration drops (see Plate VII).

#131 - Ten Hours and Fifteen Minutes

The concentration is much the same as in #127, but more cells seem to be impregnated with droplets and the concentration extends farther to the periphery. Only at the extreme periphery do the fat droplets fade in number.

#136 - Ten Hours and Forty-five Minutes

Much the same as #131.

#147 - Eleven Hours and Forty-five Minutes

Concentration is thinner in the centre, and fades out along one side of the gland which probably is the Y Zone (See Plate VII).

#156 - Twelve Hours and Forty-five Minutes

Much the same as in #131.

The osmicated glands thus give further evidence in support of the belief that there is a hypertrophy of the adrenal cortex of the house sparrow at the time that the gonads have become active.

EXAMINATION OF ADRENAL GLANDS FOR CHANGE
IN PERCENTAGE VOLUME OF CORTEX AND MEDULLA

Because of the extreme intermingling of the medullary cells in the cortical tissue, the ordinary methods of determining percent volumes were considered to be extremely inadequate if not technically impossible to carry out. The method adopted in this thesis was introduced by Hammar who used it extensively in the study of thymus, and dealt with it in detail in a special paper (described by Scammon and Scott (43)). He sectioned the thymus serially, projected the sections on heavy, well-made paper, traced them, cut out their various areas and weighed them. He then computed the area from the weight of the paper by determining a weight-area ratio by weighing a sample of the paper of known area.

There are three chief sources of error in the area-by-weight method when paper is employed:

1. Those involved in cutting out the elaborate outlines of various structures.
2. The variability in moisture content and therefore in the weight-area ratio of the paper.
3. Variation in the thickness of the individual sheets of the paper stock used in the study.

The first error is small in the hands of a careful operator and can hardly be avoided by any method. The error of moisture content is a very real one and should be taken into consideration whenever this method is employed. It has been shown that there is a rapid, though not constant absorption of water by both ledger and bond papers with increasing atmospheric humidity. There is also a change in the area of the paper due to its expansion with the increased moisture in the air.

Part of the difficulty of moisture absorption can be avoided by using Atwell and Woodworth's technique of storing the paper cuttings in a desiccator for a week and weighing upon removal. Weighing should be carried out immediately after removal of the paper from the desiccator because it absorbs a large percentage of its final moisture content within twenty minutes after exposure to the air.

The variability in the thickness in various parts of sheets of commercial bond paper is also hardly negligible. To test this factor, Scammon and Scott (43) used large sheets of paper employed in their laboratory for the area-weight method. These sheets were placed on a film surface, and fifty discs having a diameter of approximately 1.8 cms were punched from them at random with a hollow steel cylinder whose lower margin was ground to a knife edge. A similar set of discs, fifty in number, approximately 4 mms in

diameter, were cut in the same manner. These discs were then weighed on a delicate analytical balance. To eliminate, as far as possible the psychological element in this experiment, two observers were employed in making the weighings, one adjusting the analytical balance used, while the other, a totally disinterested party, recorded each weight. A coefficient of variability was found to be quite considerable.

Since absolute values are not necessary in this investigation, these errors do not take on the same importance as indicated in the above papers in which the determinations required quantitative results as an essential factor. In the case of this study, relative values are the most important thing, and not absolute values.

The use of heavy paper was at first thought desirable for this work with adrenals. Because of the delicate configuration of the medullary groups of tissue, however, it was deemed inadvisable from a mechanical standpoint to use this heavy paper, i.e. the cutting out of the pieces would be inaccurate. Therefore, two types of paper were considered for this purpose, ordinary bond paper and foolscap. In order to determine which one to use, a number of sheets of uniform style of both types were weighed piece by piece and the average taken of both groups separately. The deviation of each sheet from the average was

obtained and the mean deviation calculated. The percentage error was determined on the basis of the mean deviation (see Tables I and II). The foolscap showed a much smaller percentage error, and for this reason was chosen as the best paper of the two to be used.

The sections of each gland were projected through a microprojector and reflected by a mirror on to an adjustable stand. As each gland was serially sectioned, to get a representative number of sections, every tenth section was used.

TABLE I
WEIGHTS OF BOND PAPER

<u>Weight of Paper (gms)</u>	<u>Average Weight</u>	<u>Deviation</u>
3.5819	3.5199	+0.0620
3.5173		-0.0026
3.5412		+0.0213
3.5212		+0.0013
3.4916		-0.0213
3.4910		-0.0219
3.5162		-0.0037
3.4852		-0.0347
3.4855		-0.0344
3.4952		-0.0247
3.5486		+0.0287
<u>3.5634</u>		<u>+0.0435</u>
42.2383		0.3001

Mean Deviation = 0.0250

Percent of Greatest Deviation = 1.73%

Percent of Mean Deviation = 0.71%

(100) 100%

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	87	88	89	90	91	92	93	94	95	96	97	98	99	100
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	68	69	70	71	72	73	74	75	76	77	78	79	80	81	82	83	84	85	86	87	88	89	90	91	92	93	94	95	96	97	98	99	100

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TABLE II
WEIGHTS OF FOOLSCAP

<u>Weight of Paper (gms)</u>	<u>Average Weight</u>	<u>Deviation</u>
3.4985	3.5197	-0.0212
3.5218		+0.0021
3.5174		-0.0023
3.5173		-0.0024
3.5634		+0.0437
3.5486		+0.0289
3.4952		-0.0245
3.4855		-0.0342
3.4852		-0.0345
3.5162		-0.0035
3.4910		-0.0287
3.4916		-0.0281
3.5221		+0.0024
3.5412		+0.0215
3.5570		+0.0373
3.5380		+0.0183
<u>3.5442</u>		<u>+0.0245</u>
59.8342		0.3581

Mean Deviation = 0.0211

Percent of Greatest Deviation = 1.24%

Percent of Mean Deviation = 0.60%

TABLE III

PERCENT VOLUME OF MEDULLA AND CORTEX
IN EXPERIMENTAL BIRDS

<u>Sex & Number</u>	<u>Duration of Illumination</u>	<u>Weight of Paper</u>		<u>% of Medulla</u>	<u>% of Cortex</u>
		<u>Medulla</u>	<u>Cortex</u>		
111-F	11 hours	2.0806	8.5231	19.62	80.38
115-M	12	2.2591	9.5379	19.15	80.85
119-M	12:40	2.3540	7.5505	23.77	76.23
122-F	13:30	2.0628	7.8532	20.80	79.20
129-F	15	1.5333	10.1793	13.09	86.91
133-M	15	1.5056	11.0419	11.98	88.02
134-F	15	1.3375	11.1990	10.67	89.33
152-M	13:15	2.5111	11.5492	17.86	82.14
155-M	12	1.9845	10.3250	16.12	83.88
157-M	11:30	1.7605	8.0787	17.89	82.11
158-M	10:30	2.4030	9.5160	20.16	79.84
162-F	10	2.4372	10.9680	18.18	81.82

TABLE IV
PERCENT VOLUME OF MEDULLA AND CORTEX

Sex & Number	Duration of Illumination	IN WILD BIRDS		% of Medulla	% of Cortex
		Weight of Paper Medulla	Cortex		
116-M	8:45	2.2316	7.3150	23.37	76.63
132-F	10:15	2.2043	8.3340	20.91	79.09
137-M	10:45	2.7477	11.9543	18.69	81.31
138-M	10:45	2.2351	10.3311	17.78	82.22
142-M	11:15	2.3616	6.9222	25.44	74.56
149-F	11:45	3.7551	9.5862	28.15	71.85
150-M	11:45	2.9550	7.2540	28.94	71.06
154-M	12:15	2.6910	8.4035	24.25	75.75
156-M	12:45	1.5445	9.3843	14.13	85.87
165-F	16:40	1.5337	10.4818	12.76	87.24
167-M	16:55	1.8120	9.9114	15.46	84.54
168-M	17	2.1198	10.2412	17.15	82.85
[171	Juvenile	3.6115	8.8734	28.93	71.07]
173-F	16:25	2.1771	11.9862	15.37	84.65
[174	Juvenile	3.4826	11.0012	24.04	75.96]
176-M	16:15	1.9823	9.8475	16.75	83.24
180-M	14	2.5349	10.0076	20.21	79.79
182-F	11:30	2.2245	8.0471	21.59	78.41
185-M	9:35	2.8475	11.0121	20.55	79.45

Juveniles Not Included in Graphs.

TABLE

OF THE AREA OF THE SURFACE OF THE EARTH

TABLE I

NO.	AREA	PERCENT	C. OF THE AREA	C. OF THE AREA	PERCENT
1.	100.00	100.00	100.00	100.00	100.00
2.	99.99	99.99	99.99	99.99	99.99
3.	99.98	99.98	99.98	99.98	99.98
4.	99.97	99.97	99.97	99.97	99.97
5.	99.96	99.96	99.96	99.96	99.96
6.	99.95	99.95	99.95	99.95	99.95
7.	99.94	99.94	99.94	99.94	99.94
8.	99.93	99.93	99.93	99.93	99.93
9.	99.92	99.92	99.92	99.92	99.92
10.	99.91	99.91	99.91	99.91	99.91
11.	99.90	99.90	99.90	99.90	99.90
12.	99.89	99.89	99.89	99.89	99.89
13.	99.88	99.88	99.88	99.88	99.88
14.	99.87	99.87	99.87	99.87	99.87
15.	99.86	99.86	99.86	99.86	99.86
16.	99.85	99.85	99.85	99.85	99.85
17.	99.84	99.84	99.84	99.84	99.84
18.	99.83	99.83	99.83	99.83	99.83
19.	99.82	99.82	99.82	99.82	99.82
20.	99.81	99.81	99.81	99.81	99.81
21.	99.80	99.80	99.80	99.80	99.80
22.	99.79	99.79	99.79	99.79	99.79
23.	99.78	99.78	99.78	99.78	99.78
24.	99.77	99.77	99.77	99.77	99.77
25.	99.76	99.76	99.76	99.76	99.76
26.	99.75	99.75	99.75	99.75	99.75
27.	99.74	99.74	99.74	99.74	99.74
28.	99.73	99.73	99.73	99.73	99.73
29.	99.72	99.72	99.72	99.72	99.72
30.	99.71	99.71	99.71	99.71	99.71
31.	99.70	99.70	99.70	99.70	99.70
32.	99.69	99.69	99.69	99.69	99.69
33.	99.68	99.68	99.68	99.68	99.68
34.	99.67	99.67	99.67	99.67	99.67
35.	99.66	99.66	99.66	99.66	99.66
36.	99.65	99.65	99.65	99.65	99.65
37.	99.64	99.64	99.64	99.64	99.64
38.	99.63	99.63	99.63	99.63	99.63
39.	99.62	99.62	99.62	99.62	99.62
40.	99.61	99.61	99.61	99.61	99.61
41.	99.60	99.60	99.60	99.60	99.60
42.	99.59	99.59	99.59	99.59	99.59
43.	99.58	99.58	99.58	99.58	99.58
44.	99.57	99.57	99.57	99.57	99.57
45.	99.56	99.56	99.56	99.56	99.56
46.	99.55	99.55	99.55	99.55	99.55
47.	99.54	99.54	99.54	99.54	99.54
48.	99.53	99.53	99.53	99.53	99.53
49.	99.52	99.52	99.52	99.52	99.52
50.	99.51	99.51	99.51	99.51	99.51
51.	99.50	99.50	99.50	99.50	99.50
52.	99.49	99.49	99.49	99.49	99.49
53.	99.48	99.48	99.48	99.48	99.48
54.	99.47	99.47	99.47	99.47	99.47
55.	99.46	99.46	99.46	99.46	99.46
56.	99.45	99.45	99.45	99.45	99.45
57.	99.44	99.44	99.44	99.44	99.44
58.	99.43	99.43	99.43	99.43	99.43
59.	99.42	99.42	99.42	99.42	99.42
60.	99.41	99.41	99.41	99.41	99.41
61.	99.40	99.40	99.40	99.40	99.40
62.	99.39	99.39	99.39	99.39	99.39
63.	99.38	99.38	99.38	99.38	99.38
64.	99.37	99.37	99.37	99.37	99.37
65.	99.36	99.36	99.36	99.36	99.36
66.	99.35	99.35	99.35	99.35	99.35
67.	99.34	99.34	99.34	99.34	99.34
68.	99.33	99.33	99.33	99.33	99.33
69.	99.32	99.32	99.32	99.32	99.32
70.	99.31	99.31	99.31	99.31	99.31
71.	99.30	99.30	99.30	99.30	99.30
72.	99.29	99.29	99.29	99.29	99.29
73.	99.28	99.28	99.28	99.28	99.28
74.	99.27	99.27	99.27	99.27	99.27
75.	99.26	99.26	99.26	99.26	99.26
76.	99.25	99.25	99.25	99.25	99.25
77.	99.24	99.24	99.24	99.24	99.24
78.	99.23	99.23	99.23	99.23	99.23
79.	99.22	99.22	99.22	99.22	99.22
80.	99.21	99.21	99.21	99.21	99.21
81.	99.20	99.20	99.20	99.20	99.20
82.	99.19	99.19	99.19	99.19	99.19
83.	99.18	99.18	99.18	99.18	99.18
84.	99.17	99.17	99.17	99.17	99.17
85.	99.16	99.16	99.16	99.16	99.16
86.	99.15	99.15	99.15	99.15	99.15
87.	99.14	99.14	99.14	99.14	99.14
88.	99.13	99.13	99.13	99.13	99.13
89.	99.12	99.12	99.12	99.12	99.12
90.	99.11	99.11	99.11	99.11	99.11
91.	99.10	99.10	99.10	99.10	99.10
92.	99.09	99.09	99.09	99.09	99.09
93.	99.08	99.08	99.08	99.08	99.08
94.	99.07	99.07	99.07	99.07	99.07
95.	99.06	99.06	99.06	99.06	99.06
96.	99.05	99.05	99.05	99.05	99.05
97.	99.04	99.04	99.04	99.04	99.04
98.	99.03	99.03	99.03	99.03	99.03
99.	99.02	99.02	99.02	99.02	99.02
100.	99.01	99.01	99.01	99.01	99.01

THE AREA OF THE SURFACE OF THE EARTH

PERCENT VOLUME of CORTICAL TISSUE in EXPERIMENTAL BIRDS

PERCENT VOLUME of CORTICAL TISSUE in WILD BIRDS

— Development of Testis in
Experimental Birds

— Percent Volume of Cortical
Tissue in Experimental Birds

CONCLUSIONS

An examination of the medulla-cortical configuration of the series of both experimental and wild birds shows a great deal of variation. In some cases the medullary tissue is found in large masses in the central area of the gland, while some have medullary tissue in long strands. Others again have the medullary cells in small discrete masses. Some have thin strands of these cells running along the edge of the gland and generally the medulla is spread throughout the whole gland, while in others the peripheral regions are largely devoid of chromaffin cells. Plates I to V give a representative cross-section of adrenal glands.

Regardless of this, a general pattern can be discerned through the whole series, in both experimental and wild birds. In the winter condition the medullary tissue is in large masses which is also found at the periphery. As spring conditions are approached, the medullary bodies seem to be broken up into smaller groups and are generally pushed away from the periphery towards the centre. This is partially due to the emergence of Zone Y at the time of the breeding season as a prominent area at the periphery of the gland. These cells are large and compact, thus pushing the medullary cells towards the central area of the gland. Also, the central cells become activated and in

some cases push the chromaffin tissue into long, string-like groups which radiate towards the central sinus. Also, the gland becomes more vascular, a natural result of, or prerequisite to, an increased physiological activity of the cortical cells. After the breeding season the gland tends to return to its winter stage, but this process is very gradual. There are of course many variations and intermediate configurations, but this general over-all pattern is definite. F. A. Hartman, R. A. Knouff & Co. (13) have attempted to show that every Avian species has a characteristic chromaffin pattern. I believe that they have completely overlooked the possibility of seasonal changes and that there is no characteristic chromaffin pattern in individual species, but that there is a general tendency for the chromaffin pattern to change with the seasons. The general chromaffin pattern is also dependent largely on the amount of medullary tissue present in the gland; as the amount of medullary tissue varies, the pattern will vary. Plates I to V will indicate the great variability of the adrenal gland in one species of bird. Plate III shows that even in a number of representative sections taken from one adrenal a certain amount of variation in configuration is shown.

From the study of the percentages of medullary and cortical cells in both series, there is definitely a clear indication of a cycle. These results have been tabu-

lated in tables III and IV from which graphs have been drawn. From the graphs it will be seen that there is a cycle in both the experimental and the wild birds which closely parallel each other and which approximately follows the breeding cycle.

The increase in the volume of the cortical tissue may not be an actual increase in the number of cortical cells, but they may have merely increased in size, crowding out the medullary tissue and cutting it up into smaller bundles. It may be a combination of the two, but the histological examination of the series seems to corroborate the latter contention as being the more plausible explanation.

An examination of the graph shows that the experimental birds seem to lag behind the wild birds in the cycle. For example, the peak of the graph is reached in the experimental birds at the illumination of fifteen hours, while in the wild birds the peak of the cycle is reached at approximately fourteen hours. I think the probable explanation is that the experimental birds, due to the abnormally rapid increase in illumination, are several steps behind in their development than the wild birds in their natural environment.

The dip in both graphs may be connected with the development of the gonads. Further study along this line would no doubt shed more light on this problem.

Several investigators have noted a difference in the amount of medullary tissue found in both sexes. In this study no difference has been noted in the sexes which could not be attributed to individual variability. The juveniles show a greater volume of medullary tissue than do the adults shot at the same period.

The histological and physiological examination of both series supports the belief that a cycle is present. An examination of the cortical cells of a winter adrenal gland show that they are comparatively inactive. As the spring breeding conditions are approached the cells appear to become more active and the peak is reached during the breeding season. This is quite natural because of the sex hormones known to be elaborated by the adrenal cortical cells. After the short breeding season the cortical cells in the central area of the gland, which during the breeding season are the most active, become degenerate and die, to be replaced by younger and less active cells being pushed in from the periphery by proliferation. Thus after the period of recrudescence of the gonads the cortical cells of the adrenals generally take on an inactive appearance. This process is much more gradual than the building up of activity.

This study indicates that the life cycle of the cortical cells generally accepted is substantially true. It is apparent throughout the whole series of glands by

the appearance of the cortical cells that the young cells are at the periphery and that the older degenerating cells are in the central area. To determine whether new cortical cells are elaborated by the capsule would require special techniques and is beyond the scope of this thesis.

The osmicated adrenal glands show a definite activity cycle. In general the central cortical cells show a greater concentration of lipoid than the peripheral cortical cells. It is believed by Flexner and Grollman, Kar and others, that the cells which show little or no stainable lipoid are the youngest type and that in the course of their migration inward the droplets increase both in number and in size. As the breeding season is approached the concentration of osmicated fats increased greatly in some of the peripheral areas. With the passing of the breeding season there is a gradual recession in the amount of lipoid present. It is generally believed that when the lipoid content is low the functional activity of the cortex is low and is increased when the secretory activity is supposed to be increased. This study tends to substantiate this contention.

Thus from these various studies there is suggestive evidence that a cycle correlated to the breeding cycle is present in the adrenal gland of the house sparrow. In the light of this positive evidence it seems that there should

be a revaluation of many investigations which have already been carried out on the Avian adrenal gland. More investigations along other lines are required to establish more exact relationship between the pituitary gland, the adrenal cycle and the gonadal cycle in the house sparrow.

SUMMARY

In the present survey twenty-four experimental and about sixty wild bird adrenal glands of the house sparrow (Passer domesticus) were examined microscopically.

1. A general description of the adrenal gland of the house sparrow was made.
2. No typical medullary tissue configuration was seen, but a general pattern of change was observed. This change was due to the amount of medullary tissue found in the gland, the emergence of the Y Zone and the physiological activation of the central cortical cells with the approach of the breeding season.
3. Histological examination of both series showed a general activity cycle of the cortical cells. This was substantiated by the investigation of adrenal glands fixed in an osmic acid solution to bring out the steroid content of the glands.
4. An area-by-weight method was used to determine changes in volume of medulla and cortex and it was shown that during the breeding season the volume of medullary tissue decreases considerably.

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PLATE I

CONFIGURATION OF ADRENAL GLANDS
OF EXPERIMENTAL BIRDS

Figure I	Bird #111	11 hours	Female
Figure II	#114	11 hours 40 mins.	Male
Figure III	#115	12 hours	Male
Figure IV	#119	12 hours 40 mins.	Male
Figure V	#123	14 hours	Male
Figure VI	#124	15 hours	Female

Explanation:

Medullary groups of cells.

Blood sinusoids.

Cortical Tissue.

Y Zone.

I

II

III

IV

V

VI

6. Cl 100 1

1. $\frac{1}{2} \times \frac{1}{2} = \frac{1}{4}$

 $\frac{1}{2} \text{O}_2 + \frac{1}{2} \text{C}_2\text{H}_2 \rightarrow \text{CO} + \text{H}_2$

W. L. F. Co.

PLATE II
CONFIGURATION OF ADRENAL GLANDS
OF EXPERIMENTAL BIRDS

Figure I	Bird #134	15 hours	Female
Figure II	#152	12 hours 40 mins	Male
Figure III	#155	12 hours	Male
Figure IV	#157	11 hours 30 mins	Male
Figure V	#158	10 hours 30 mins	Male
Figure VI	#160	10 hours	Male

Explanation:

Medullary groups of cells.

Blood sinusoids.

Cortical Tissue.

I

II

III

IV

V

VI

PLATE III
CONFIGURATION OF ADRENAL GLANDS
OF EXPERIMENTAL BIRDS

Figures I to VI Bird #133 fifteen hours Male

Explanation:

- Medullary groups of cells
- Blood sinusoids
- Cortical tissue
- Y Zone

I

II

III

IV

V

VI

The following is a list of the
 names of the persons who have
 been appointed to the various
 positions in the office of the
 Secretary of the State.

John A. Smith	John A. Smith	John A. Smith
John A. Smith	John A. Smith	John A. Smith
John A. Smith	John A. Smith	John A. Smith
John A. Smith	John A. Smith	John A. Smith
John A. Smith	John A. Smith	John A. Smith
John A. Smith	John A. Smith	John A. Smith
John A. Smith	John A. Smith	John A. Smith

: c. 1870

Office of the Secretary of the State

Colonial Office

Office of the Secretary of the State

PLATE IV
CONFIGURATION OF ADRENAL GLANDS
OF WILD BIRDS

Figure I	Bird #108	7 hours 40 mins.	Female
Figure II	#116	8 hours 20 mins.	Male
Figure III	#131	10 hours 15 mins.	Male
Figure IV	#138	10 hours 45 mins.	Female
Figure V	#144	11 hours 15 mins.	Male
Figure VI	#150	11 hours 45 mins.	Male

Explanation:

Medullary groups of cells

Blood sinusoids

Cortical tissue

I

II

III

IV

V

VI

PLATE V
CONFIGURATION OF ADRENAL GLANDS
OF WILD BIRDS

Figure I	Bird # 167	16 hours 50 mins.	Male
Figure II	# 173	16 hours 25 mins.	Female
Figure III	# 175	Juvenile	
Figure IV	# 176	16 hours 15 mins.	Male

Explanation:

Medullary groups of cells

Blood sinusoids

Cortical tissue

I

II

III

IV

TABLE

of the

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PLATE VI
PHOTOMICROGRAPHS OF OSMICATED
ADRENAL GLANDS

Experimental Birds

Figure I	Bird #115	12 hours	Male
Figure II	#122	13 hours 30 mins.	Female
Figure III	#128	15 hours	Female
Figure IV	#152	12 hours 40 mins.	Male

I

II

III

IV

PLATE VII
PHOTOMICROGRAPHS OF OSMICATED
ADRENAL GLANDS

Experimental Birds

Figure I	Bird #155	12 hours	Male
Figure II	#160	10 hours	Male

Wild Birds

Figure III	#127	9 hours 45 mins.	Male
Figure IV	#156	12 hours 45 mins.	Male

I

II

III

IV

B29761