

physiological refinement over their method in that air can be trapped in localized lung areas while adjacent lung tissues will remain relatively collapsed and resilient as is known to occur clinically. In addition, these animals remain in good health for long periods of time and are available for various types of experiments.

Summary. Partially obstructive endobronchial lesions have been produced in dogs by transbronchial injection of a plastic material. These lesions were composed primarily of large accumulations of vacuolated macro-

phages. The size of these masses was apparently dependent upon their age. The implication of this type of lesion in possible production of experimental emphysema has been discussed.

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Received June 24, 1960. P.S.E.B.M., 1960, v105.

Corticosterone Levels in Adrenal Effluent Blood of Some Gallinaceous Birds.* (26011)

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Collection of adrenal effluent blood from mammalian species by cannulation of the adrenal vein(1) is facilitated by the morphological relation of this vessel to the vena cava. Recently Phillips and Jones(2) made direct collections of adrenal venous blood from the capon by a procedure that involved obstruction of the blood flow through the vena cava and formation of a pocket within this vein into which flowed adrenal effluent blood. This method, while satisfactory for some types of experimentation, is laborious for routine collection, is unsuitable for repetitive sampling, and is acutely traumatic.

In domestic fowl the adrenal veins are located on the ventral surface of the glands; the left adrenal vein passes transversely from the adrenal to the vena cava whereas the right adrenal vein passes dorso-laterally to the caval vein(3). The nature of the venous drainage from the *left gland* permits collection of adrenal effluent blood from the bird. However, the intimate apposition of the ovary to the left adrenal gland restricts sampling from this vein to the *male* galliform. We

routinely make direct collections of adrenal venous blood by a simple, expedient manipulation that allows the animal to be used for study at a subsequent period.

Materials and methods. The male gallinaceous birds used in this study were 3 breeds of domestic fowl which were 10 to 20 weeks old and 1.5 to 2.5 kg in body weight; 5 to 12 month old ring-necked pheasants[†] about 1 kg in weight; and broad-breasted bronze turkeys[†] about 6 months old and approximately 12 to 14 kg in weight.

The birds were anesthetized by intravenous infusion of Nembutal. Peripheral blood was withdrawn either from the cephalic vein or from the heart. The body cavity was entered through incisions made in the skin and intercostal musculature between 6th and 7th ribs. A rib spreader was inserted and expanded to allow better access to the region of the adrenal gland. The presence of recrudescing gonads often necessitated retraction of the left testis for exposure of the adrenal vein. If sexually mature males are to be studied, it would be helpful unilaterally to castrate the animals.

* This study was financed in part by the Wisconsin Conservation Dept. with Federal Aid to Wildlife Restoration funds under Pittman-Robertson Project and by grant of Nat. Heart Inst.

[†] Made available through the courtesy of Wisconsin Conservation Dept. and Univ. of Wisconsin Dept. of Poultry Husbandry, respectively.

TABLE I. Corticosterone Levels of Peripheral and Adrenal Effluent Plasma of Gallinaceous Birds.

Animal	No.	$\mu\text{g}/100 \text{ ml plasma}$	
		Adr. effl.	Peripheral
Chickens			
Wisco White	8	$44.3 \pm 5.5^*$	$7.3 \pm .3^*$
New Hampshire	8	48.8 ± 3.7	12.3 ± 2.3
White Rock	8	37.0 ± 6.5	$9.5 \pm .2$
Pheasants	19	34.8 ± 4.7	$8.6 \pm .8$
" castrate	5	33.1 ± 7.2	$7.8 \pm .7$
Turkeys	6	42.4 ± 5.3	$7.8 \pm .4$

* Mean $\pm$ S.E.

Approximately 1.5 ml of blood were withdrawn in about 90 seconds from the left adrenal vein with a heparinized syringe fitted with a hypodermic needle the distal 5 mm of which had been bent to approximate a right angle. A 26 gauge needle was used for collecting adrenal effluent blood from the pheasant and domestic fowl, and a 22 gauge needle was used for the turkey. A pair of spectacle loupes is helpful for inserting the needle into the vein. Several samples can be obtained from the same bird if injury to the adrenal vein is kept at a minimum. Upon removal of the sample, the skin incision was closed with wound clips; the musculature between the costa was not sutured. The entire procedure can be completed in a matter of minutes.

In some instances blood was obtained from the turkey by cannulation. A 22 gauge hypodermic needle was separated from its shank, then inserted into one end of a piece of polyethylene tubing (0.034 inch I.D.). The cannula and bird were heparinized, and the needle was placed into the adrenal vein; blood was collected for 10 minute intervals.

The level of free corticosterone in aliquots of peripheral (1.0-2.0 ml) and adrenal effluent (0.2-0.4 ml) plasma was determined by the method of Silber *et al.* (4). Fluorescence was read in a Photovolt Corp. Fluorometer Model 54 and was quantitated relative to crystalline corticosterone. This method measures both corticosterone and hydrocortisone; however, we believe that the fluorescence attributable to the latter is negligible, since, as will be reported later, we were unable chromatographically to discern hydrocortisone in extracts of up to 50 ml of adrenal effluent plasma.

Results and discussion. Level of corticosterone in the sample of plasma is directly influenced by rate of removal of blood from the adrenal vein (Table II). Rapid withdrawal merely dilutes the level of corticosterone. However, if blood is withdrawn with a syringe at a rate of not more than 1.0 ml/min the amount of corticosterone present is not appreciably influenced by rate of flow of blood. Collection of blood with a syringe is more satisfactory for routine sampling than cannulation because of the ease by which blood can be obtained.

Plasma levels of corticosterone in the species of birds studied were surprisingly similar; values in adrenal effluent plasma were 4 to 6 times greater than peripheral levels (Table I). The amount of corticosterone in the peripheral plasma of the domestic fowl was similar to that reported by Urist and Deutsch (5). However, we found the level of corticosterone for adrenal blood of domestic fowl considerably less than the value Phillips and Jones (2) found in the capon. This difference may be due to variations in technics used for collecting blood and for determining amount of corticosterone. It is interesting to note that castration had no influence on adrenal activity in the pheasant (Table I).

Intravenous administration of 8 I.U. of ACTH (G. A. Breon & Co., El-Acorto Gel) to the pheasant increased the corticosterone in adrenal venous plasma 186% at 30 minutes and 253% at 60 minutes after injection (Table III). Wisco White cockerels showed in-

TABLE II. Effect of Rate of Blood Withdrawal on Plasma Concentration of Corticosterone in Turkeys.

Bird	Rate of blood removal (cc/min.)	Corticosterone ($\mu\text{g}/100 \text{ ml}$)
I	.8	31.0
"	6.0	4.5
II	1.1	36.0
"	2.7	7.7
III	.2	28.8*
IV	.2	29.9*
V	.2	31.1*
"	.3	43.2*
"	.4	16.4*
"	.4	28.0*

* Sample obtained by cannulation of adrenal vein.

TABLE III. Corticosterone Levels in Adrenal Effluent after Repeated Bleedings and ACTH.

Animal		μg corticosterone/100 ml plasma at			
		0 time	30 min.	60 min.	90 min.
Pheasants					
No treatment	(3)*	25.7 $\pm$ 5.5†	34.0 $\pm$ 12.3†	31.0 $\pm$ 3.7†	
8 IU ACTH IV	(4)	29.6 $\pm$ 5.4	84.6 $\pm$ 34.5	104.6	(2)
Chickens					
No treatment	(3)	27.9 $\pm$ 7.2	56.8 $\pm$ 15.3		26.3 $\pm$ 12.0
8 IU ACTH IV	(3)	30.7 $\pm$ 3.6	68.3 $\pm$ 14.5	143.7 $\pm$ 50.7	

* No. of animals.

† Mean $\pm$ S.D.

creases of 123% and 368% at these same times. Urist and Deutsch(5), using the method of Silber, report an 85% increase in corticosterone level of peripheral plasma of male fowl following administration of 400 I.U. of ACTH. Phillips and Jones(2) found a decrease in amount of corticosterone in adrenal effluent blood of the capon following ACTH treatment.

Summary. A method is described for obtaining samples of adrenal venous blood from the bird. This allows avian species to be used for routine *in vivo* studies designed to assess adrenal activity by direct measurement. Levels of corticosterone, expressed as $\mu\text{g} \%$, in adrenal effluent and peripheral plasma were 42.4 and 7.8 in turkeys and 34.8 and 8.6 in pheasants. In domestic fowl the values were 44.3 and 7.3 in Wisco Whites; 48.8 and 12.3

in New Hampshires, and 37.0 and 9.5 in White Rocks. ACTH (8 I.U. intravenously) increased the output of corticosterone from 29.6 to 104.6 $\mu\text{g} \%$ in pheasants and from 30.7 to 143.7 $\mu\text{g} \%$ in chickens 60 minutes after injection.

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Received July 8, 1960. P.S.E.B.M., 1960, v105.

Comparative Bilirubin Levels in Vitreous Body, Synovial Fluid, Cerebrospinal Fluid and Serum after Death. (26012)

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Further studies on postmortem chemistry (7-11) revealed in cases of jaundice presence of bilirubin not only in cerebrospinal fluid, where it was known to occur(1-6), but also in vitreous body and synovial fluid. This to our knowledge has not been reported before, and it seemed of interest, therefore, to obtain data on comparative concentrations in these fluids.

Materials and methods. Material from autopsies about 5½ hours after death on the average was based on a total of 78 male adult

bodies including 44 cases of jaundice and 34 non-jaundice control cases. Spinal fluid was obtained by cisternal puncture(7,9,10), vitreous body by puncture of the eye balls at the outer canthus with a 20 gauge needle and 10 cc syringe(11), and synovial fluid was aspirated from the knee joints with a 16 gauge needle and 10 cc syringe. Bilirubin determinations were performed by the method of Malloy and Evelyn(13) with the usual 1 minute modification for direct bilirubin(13). Floc- cules of mucoprotein forming in dilute syno-