Levels of Epinephrine and Norepinephrine in Blood and Tissues of Duck, Pigeon, Turkey, and Chicken (35034)

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The ratio of norepinephrine to epinephrine (N/E) in the blood and heart of most mammals and even some reptiles is greater than 1 (1), thus suggesting that the neurotransmitter is norepinephrine. The blood and heart of the chicken, like that of frogs, contain more E than N (2–4). However, this does not necessarily mean that E is the neurotransmitter in chickens, because the high concentration of E in chicken heart may be derived from the blood and that in turn from the adrenals or other organs. That the myocardium can extract catecholamines from the plasma is well known (5, 6).

Whether or not the chicken is representative of most avian species has not been determined and is an object of this study, where comparative levels of N and E in blood and tissues of ducks, pigeons, turkeys, and chickens are reported. Another objective is to determine the effects of anesthesia and excitement on plasma and tissue catecholamine levels.

Materials and Methods. Determinations of levels of N and E were made on the animals during the months of October, November, and December. The birds were removed from their pens with a minimum of excitement and taken to the laboratory where they were allowed to remain quietly in their crates for 30 min. Each bird was taken from its crate and blood was obtained by heart stab in the following amounts: for chickens, 10–15 ml each; turkeys, 30 ml; ducks, 10 ml; and for pigeons, 3 ml. All blood was taken within 30–40 sec and if difficulty was encountered in sampling, the sample was discarded. Since the total volume required for determination was approximately 30 ml, it was necessary to pool samples of several individuals, particularly for the pigeons.

The samples were centrifuged at 8–10°, and the resulting plasma (approximately 15 ml/sample) treated with perchloric acid reagent and centrifuged at 10°. The supernatant was kept in a freezer until analyzed by the aluminum oxide-trihydroxyindole method as modified by Anton and Sayre (1) and Lin and Sturkie (3). The developed fluorescence was measured in an Aminco-Bowman spectrophotofluorometer. Two sets of activation and fluorescence wavelengths (400 and 520, and 436 and 520 mμ) were employed for norepinephrine and epinephrine, respectively, both of which were determined from the same sample and calculated by the formula of Price and Price (7).

The analyses of catecholamines in the spleen and heart were conducted on a different group of birds not previously sampled for blood catecholamine determinations. All birds except those deliberately excited or sedated were caught, placed in a crate and allowed to remain quiet and undisturbed for 0.5 hr after which time each bird was removed and quickly decapitated. The heart and spleen were removed within 30 to 50 sec and instantly submerged in liquid nitrogen. The frozen tissues were then ground and stored in a freezer until analyzed by the same technique as for blood.

Results. Plasma levels of epinephrine (E) and norepinephrine are presented in Table I. Table I shows that the pigeon, chicken, and turkey have higher levels of E than N in the plasma with ratios of N/E of considerably less than 1, but the ratio is greater than 1 in the duck (2.60).

TABLE I. Norepinephrine and Epinephrine Levels (μg/liter) in Plasma of Pigeon, Duck, Turkey, and Chicken.

Species	Sex	Age (months)	No. of samples	No. of birds/ sample	Sample vol (ml)	Norepinephrine Mean	Norepinephrine SE	Epinephrine Mean	Epinephrine SE	Ratio N/E
Pigeon	Mix	12	8	9–14	27–42	0.225	0.158	5.761	1.414	0.39
Duck	Mix	2	10	3	30	0.828	0.071	0.319	0.114	2.60
Turkey	F	7–12	4	1	30	0.684	0.274	1.446	0.670	0.47
Chicken	M	17	10	3–4	30	0.843	0.211	8.68	0.983	0.097
	F	17–21	21	3–4	30	1.181	0.158	8.24	0.899	0.140

The data for tissue catecholamines are shown in Table II. The spleens of the duck, pigeon, and chicken contain higher levels of N than E and the actual values appear lower for the chicken, although more data are needed. The level of N is higher in the atria and ventricles than E for the duck and pigeon, but not the chicken where the level of E is higher.

Effects of Anesthesia and Excitement on Catecholamine Levels. The blood catecholamine levels reported for chickens (17–21 months of age) are lower than those reported by Lin and Sturkie (3), where handling and excitement of birds were probably at a maximum. The higher levels of N in the blood of females reported by Sturkie and Lin (2) are indicated herein, although the difference is not significant. A recent study from this laboratory on chickens of both sexes from 5 to 31 months reveals considerable differences in blood catecholamine levels, particularly N levels, but in all cases, the E levels were considerably higher than the N levels. Some of this variation may be related to sex and age differences, but much of it is probably caused by the conditions of handling, excitement, and blood sampling, factors which influence the release of adrenal and neuronal catecholamines. Accordingly, experiments

TABLE II. Norepinephrine (N) and Epinephrine (E) Levels in Tissues (μg/g) of Ducks, Pigeons (both sexes) and Chickens (males).

	Spleen		Right atrium		Left atrium		Right ventricle		Left ventricle	
	N	E	N	E	N	E	N	E	N	E
Duck										
No.	6	6	6	6	5	5	6	6	6	6
Mean	2.05	0.403	1.13	0.019	0.908	0.011	0.535	0.118	0.329	0.019
SE	0.224	0.339	0.15	0.019	0.454	0.011	0.141	0.077	0.056	0.007
Ratio N/E	5.08		59.4		82		4.5		17.3	
Pigeon										
No.	6	6	6	6	6	6	6	6		
Mean	2.605	0.027	1.536	—	1.297	—	2.082	0.218	1.347	—
SE	0.513	0.01	0.357	—	0.224	—	0.357	0.209	0.079	—
Ratio N/E	96		—		—		9.4		—	
Chicken (male)[a]										
No.	17	17	6	6	6	6	6			6
Mean	0.534	0.124[b]	0.959	1.485	0.791	1.211	0.302			0.469
SE	0.109	0.022	0.035	0.110	0.054	0.074	0.039			0.134
Ratio N/E	4.3		0.64		0.65			0.64		

[a] From data of Lin and Sturkie (1968).

[b] From Lin and Sturkie, unpublished data.

TABLE III. Norepinephrine (N) and Epinephrine (E) Levels of Plasma (μg/liter) and Atria (μg/g) of White Leghorn Males, Before and After Anesthesia.

	Anesthesia			
	Before		After	
	N	E	N	E
	Plasma			
No.	12	12	12	12
Mean	1.94	4.56	0.71	2.34
SE	0.17	0.83	0.15	.62
	Atria			
No.	4	4	4	4
Mean	0.32	0.33	0.46	0.05

were designed to test this supposition.

Blood samples were collected from trained males that had become adapted to their surroundings and handling. The birds were removed from their individual cages with a minimum of excitement and 15 to 30 ml of blood were taken by heart stab within 20 sec or less. A second group of trained males was anesthetized by injecting them intramuscularly (in their cages) with sodium pentobarbital (approx 90 mg/bird). As soon as the birds were under deep anesthesia (10–20 min), blood samples were collected by heart stab. Catecholamine levels were determined as previously described. The results are shown in Table III. It is apparent that the levels of N and E are considerably lower in anesthetized birds. The effects of excitement and lack of it (sedation) on tissue levels of N and E (atria) were also determined on a small number of birds. One group was caught and killed by cervical dislocation (maximum excitement), and another was sedated with pentobarbital sodium before the tissues were obtained and analyzed. The results are presented in Table III. The effects of anesthesia and lack of excitement decreased significantly the level of epinephrine in the heart, presumably by decreasing the amount released into the blood from other organs such as spleen and adrenals. The increase in atrial norepinephrine following anesthesia may mean that N is not released or metabolized as rapidly. Recent evidence by Quevedo and D'Iorio (8) indicates that sodium pentobarbital decreases the enzymatic breakdown of N by a partial inhibition of monoamine oxidase.

Discussion. The data reveal considerable variation in plasma and tissue levels of catecholamines and these levels mean little unless related to mechanisms of synthesis, release, and degradation, and factors influencing these.

The chicken, turkey, and pigeon have higher levels of epinephrine than norepinephrine in the blood and this probably reflects higher levels of epinephrine released by the adrenals of these species. Duck blood, however, like that of mammals, has a higher level of plasma N than E.

Considerable variation exists between the avian species with respect to levels of N and E in heart tissue, particularly atria. Ducks and pigeons have higher levels of N than E, and chickens less N. These differences are attributable in part to species differences in the ability to store and concentrate circulating E and also to factors influencing the release of E from other organs into the blood. This variation in release probably accounts for much of variation observed in blood and tissue levels of avian and mammalian species.

Preliminary studies from this laboratory reveal that electrical stimulation of the cardiac nerves of isolated chicken hearts causes the release of only norepinephrine and not epinephrine. This and other evidence suggests that neuronal N in the heart of chicken is bound and is the neurotransmitter and that E, derived mainly from circulating E, is only loosely bound, although this remains to be determined.

Summary. Epinephrine and norepinephrine were determined by spectrophotofluorometry on plasma of ducks, pigeons, turkeys, and chickens, and on certain tissues. The concentration of E was considerably higher than N in the plasma of all species except the duck where the N/E ratio was 2.60. Ducks and pigeons have higher levels of N than E in heart tissue (atria) and chickens have less N. Anesthesia or lack of excitement de-

creased significantly the plasma and atrial levels of N and E in chicken and the results suggest that the variation in release and synthesis of N and E from neuronal and adrenal sites probably accounts for much of the variation observed in blood and tissue levels of catecholamines.

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