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**A QUANTITATIVE COMPARISON OF THE SYMPATHOMIMETIC
AMINE CONTENT OF THE LEFT AND RIGHT
ADRENAL GLANDS OF THE CAT**

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Much work has been undertaken concerning the effects of various chemical and physical stimuli on the amine content of one adrenal gland of the animal, the other being used as a control. Some of the early workers (Elliott, 1912; Stewart & Rogoff, 1916) calculated the adrenaline content of the adrenal per gland. More recent workers (Bülbring & Burn, 1949; Crawford & Outschoorn, 1951) assume that the adrenaline and noradrenaline content of the two glands is the same per gram of tissue and thus they express their results in $\mu\text{g}/100\text{ mg}$ of gland. Still other workers (Satow, 1938; Hökfelt & McLean, 1950) have expressed their results in three ways: in $\mu\text{g}/\text{mg}$ of gland, in $\mu\text{g}/\text{gland}$ and in $\mu\text{g}/\text{kg}$ of body weight. Butterworth & Mann (1956), when comparing the amine content of the two adrenals of the cat, subsequent to depletion and resynthesis of the amines in one of them, found it necessary to ascertain whether the adrenaline and noradrenaline contents of the two glands were the same per gland or per unit weight of gland. Thus the main object of this paper is to determine the relationship between the amine contents of the two adrenals of each cat. Certain other statistically derived data concerning normal cat adrenals are included.

METHODS

Sources. The adrenal glands were obtained from cats which were employed for various types of experimental work. The glands that were used for determining the normal adrenaline and noradrenaline content were obtained by careful removal under chloralose anaesthesia before any other experiment was performed. Those that were used for comparing the amine content of the left and right glands of each animal were obtained subsequent to various experimental conditions. The adrenals of ovariectomized, pregnant and lactating cats are excluded from the results of

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Table 1, since it is known that such conditions affect the weight of the glands. They are included in Table 2 since, even under such conditions, the two glands should bear the same relationship to each other as in the non-pregnant animal.

Preparation of the extracts of the adrenal glands. Immediately the gland was removed it was freed of connective tissue, weighed on a microtorsion balance and a 50 mg/ml. extract prepared by grinding in 0.1 N-HCl with 300 mg of acid-washed sand per 100 mg of gland. The extract was centrifuged at 5000 r.p.m. for 2 min and the supernatant fluid withdrawn. After checking that the extract was definitely acid, it was stored in an airtight container at just above 0° C.

Assay methods. The adrenaline and noradrenaline content of the extracts was determined by differential biological assays, using the blood pressure of the hexamethonium-treated cat and the acutely denervated nictitating membrane of the cat as separate test preparations. The amines were calculated as the laevo isomers of the base, by the formula originated by Bülbring (1949). In addition, a qualitative paper chromatographic analysis similar to that of Shepherd & West (1951) was made, using *n*-butanol-acetic acid-water as the solvent and potassium iodate as a spray to locate the catechol amines.

RESULTS

No significant difference ($P > 0.05$) was found between (1) the weight of adrenal tissue in the male, castrated male and female cats, either when expressed as mg or as mg/kg of body weight (Table 1); (2) the weights of the left and of the right adrenal glands in either the male or the female cats (Table 2); (3) the sexes with respect to the total amine content when expressed as per 100 mg or as per gland (Table 3), and (4) the sexes with respect to the percentage of noradrenaline in the glands (Table 3).

As can be seen from Table 5, 28 out of 36 pairs of glands show a closer relationship when the results are expressed as per gland than as per 100 mg of gland, i.e. $\frac{\text{total amine in left}}{\text{total amine in right}}$ per gland is nearer to 1 than $\frac{\text{total amine in left}}{\text{total amine in right}}$ per 100 mg of gland. There was no difference between the sexes in this respect. Four pairs of glands show an identical total amine content. Of these, two pairs were of identical weight and two differed by only 1 mg. Four pairs show a closer relationship per 100 mg than per gland. These pairs differed in weight by 3, 5, 16 and 38 mg (1.1, 2.6, 11.4 and 18.1% respectively); 3 and 5 mg represent only insignificant volumes of extract. Thus only two out of 32 pairs of glands fail to conform with the statement that the total amine content of the two glands of any cat is the same, irrespective of the difference in weight of the two glands. Statistical analysis of these results (Table 4) gives a variance ratio (F) of 2.25, which is greater than the value of 1.89 given in the tables at $P = 0.05$. Since the variance for the results expressed as per gland is the smaller figure, the better method of expressing the amine content is per gland and not as per unit weight of gland. There was no significant difference ($P > 0.05$) between the percentages of noradrenaline in the left and right glands.

By means of the chromatographic analysis the presence of adrenaline and noradrenaline in the glands was confirmed. No other catechol amines were detected.

TABLE 1. Weight of cat adrenal tissue

	mg			mg/kg		
	Castrated males	Males	Females	Castrated males	Males	Females
Mean	431	401	419	111.4	131.1	148.1
No. of observations	21	41	48	21	41	48
s.d.	± 121.4	± 160.3	± 141.4	± 35.2	± 49.9	± 39.9
s.e.	± 26.5	± 25.0	± 20.4	± 7.67	± 7.79	± 5.76
<i>t</i>	0.837		0.557	1.802	1.752	
<i>P</i>	> 0.05		> 0.05	> 0.05	> 0.05	

TABLE 2. Difference in weight (mg) of the two adrenal glands of each cat

	Males		Females	
	Difference in weight of right and left glands (R - L)	Percentage difference $\left(\frac{R-L}{R} \times 100\right)$	Difference in weight of right and left glands (R - L)	Percentage difference $\left(\frac{R-L}{R} \times 100\right)$
Maximum	+ 100	+ 24.76	+ 70	+ 21.43
Minimum	- 58	- 26.44	- 40	- 36.36
Mean	—	+ 1.34	—	+ 1.28
No. of observations	—	62	—	60
s.d.	—	± 11.96	—	± 10.64
s.e.	—	± 1.52	—	± 1.37
<i>t</i>	—	0.112	—	0.121
<i>P</i>	—	> 0.05	—	> 0.05

TABLE 3. Amine content (μ g) of cat adrenal glands

	Total amine/100 mg		Total amine/gland		Noradrenaline (%)	
	Males	Females	Males	Females	Males	Females
Maximum	385.8	404.7	627.4	991.5	89.9	90.7
Minimum	91.1	50.3	177.9	100.6	18.7	13.3
Mean	184.1	146.9	330.6	311.4	58.1	53.4
No. of observations	22	16	22	16	22	16
s.d.	± 72.7	± 83.3	± 116.1	± 210.6	± 19.8	± 19.1
s.e.	± 15.5	± 20.8	± 24.8	± 52.6	± 4.22	± 4.77
<i>t</i>	1.430		0.331		0.737	
<i>P</i>	> 0.05		> 0.05		> 0.05	

DISCUSSION

The fact that there is a much closer relationship between the total amine in the two glands of any one cat, when expressed as per gland rather than as per 100 mg of gland, shows that in this animal all results should be calculated per gland and not per unit weight of gland. In some experiments the method of calculation makes little difference to the answer but in certain cases, e.g. either where the difference in weight of the two glands is large or where the degree of depletion or of resynthesis is small, it can make a considerable difference. It was found that if the two glands differed in weight by 10% or more, then calculating the answer per gland instead of per 100 mg of gland,

TABLE 4. Relationship between the amine content of the left and right adrenal gland of the cat

Difference in weight of glands (mg)	Increase in weight of glands (%)	Noradrenaline (%)		Total amine in left Total amine in right	
		Left gland	Right gland	Per 100 mg	Per gland
(a) Males					
0	0	63.8	63.8	1.00	1.00
1	0.5	13.8	17.4	1.03	1.03
3	2.3	95.4	95.5	1.17	1.14
5	2.6	0	0	1.00	1.03
10	5.8	59.2	60.7	1.41	1.33
11	6.6	43.1	36.1	1.19	1.12
10	9.1	81.5	85.0	0.92	1.00
23	9.8	77.5	84.3	1.14	1.04
51	15.4	74.5	68.8	0.86	0.99
32	16.4	57.2	61.2	1.55	1.33
32	20.5	53.7	49.4	1.16	0.96
35	25.9	68.8	65.7	1.32	1.05
(b) Castrated males					
0	0	52.9	52.9	1.00	1.00
3	1.1	36.6	37.3	1.00	1.02
12	5.3	49.8	43.6	1.25	1.19
10	6.3	76.3	61.7	1.57	1.48
12	7.0	32.2	31.8	1.05	0.98
21	8.4	0.9	14.7	1.23	1.14
18	9.8	49.3	46.9	1.16	1.05
16	11.4	76.1	75.5	1.03	0.92
23	12.4	45.8	52.9	1.09	0.97
24	12.6	85.9	81.9	1.08	0.96
29	15.2	47.0	53.5	0.83	0.96
52	19.1	46.0	53.1	0.82	0.97
(c) Females					
1	0.5	5.5	0.7	1.05	1.05
1	0.6	2.3	0	1.01	1.00
2	1.0	3.8	0.9	0.97	0.98
2	2.0	38.3	40.8	0.80	0.81
6	2.5	3.2	6.8	0.94	0.96
5	4.2	40.9	38.9	1.12	1.08
17	8.1	9.8	0	1.26	1.17
15	8.3	46.9	41.7	1.05	0.97
10	9.8	29.4	27.3	1.23	1.12
38	18.1	53.4	53.3	0.99	0.84
35	21.2	40.7	43.4	1.57	1.30
40	36.4	40.0	38.1	0.71	0.97
Mean		44.49	44.04	1.099	1.053
No. of observations		36	36	36	36
Variance		—	—	0.043	0.019
s.d.		± 26.5	± 26.4	± 0.207	± 0.138
s.e.		± 4.41	± 4.39	± 0.034	± 0.023
		$t = 0.072$ $(P > 0.05)$		Variance ratio (F) = 2.25	

reduces the mean difference between the amine contents from 25 to 10%. The fact that the percentage of noradrenaline is so similar in both the glands of the animal is also of importance because otherwise the use of one gland of each animal as a control would not be valid.

TABLE 5. The relationship between the amine content of the left and the right adrenal gland of the cat
(Data derived from Table 4.)

	Males	Castrated males	Females	Total
Closer relationship per gland than per 100 mg of gland	9	9	10	28
Same answer per 100 mg and per gland (weight of glands same or 1 mg different).	2	1	1	4
Closer relationship per 100 mg than per gland	1	2	1	4
Total number in group	12	12	12	36

Although there is no significant difference between the percentages of noradrenaline in the glands of the male and the female cats there is a wide variation in this percentage between individual animals, in both the sexes. The least amount of noradrenaline found was 13.3% and the greatest 90.7% (Table 3). This wide range is in agreement with the work of others (Vogt, 1952; West, 1955). The somewhat lower percentages of noradrenaline in the results of Table 4, as compared with those of Table 3, may be explained by the fact that in Table 4 the glands were removed at the completion of the experiment, by which time there was inevitably some depletion of the glands. This was immaterial in these experiments since it was reasonable to assume that both glands were affected equally.

Percentage differences and not absolute differences in weight have been used in determining whether there is any significant difference between the weights of the two glands of each animal. Obviously this is necessary because a few milligrams difference in weight of the two glands is negligible if the glands are large but is important if they are small. No attempt has been made to correlate the weight of the glands with the age of the cat. Although it might be expected that the age of the cat is proportional to the weight of the animal, this is not necessarily so since many factors, such as the health and diet of the animal, affect the weight. In any case, the weight of almost all the cats used in these experiments only varied between two and four kilograms.

SUMMARY

1. There was no significant difference between the two sexes with respect to the weight of the adrenal glands, the weight of the glands per kilogram of body weight, the total amine content of the glands and the percentage of noradrenaline in the glands.

2. There was a closer relationship between the total amine content of the two glands of any one animal when expressed as per gland than when expressed as per 100 mg of gland.

3. There was no significant difference between the percentage of noradrenaline in the left and in the right adrenal glands.

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