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Some Factors Involved in the Assay of Renin.

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An assay of renin on anesthetized dogs has proved difficult because of the variable sensitivity of individual animals.[1, 2] Assays based on fewer than 10 tests have not always proved reliable. The factors underlying this difference in sensitivity are not wholly clear. Since only one test was made on an individual dog in one day, and an interval of 4-5 days allowed between tests, it is not plausible that a tachyphylaxis or a progressive resistance to the renin was contributing to the variability. Data has been assembled on several points which might bear on the inconstancy of assay results. Does the level of the starting blood pressure, which shows variation in anesthetized dogs, affect the magnitude of the response to renin? Would an assay based on a minimum number of dogs with repetition of tests on each be more reliable? Since it has been reported[3] that nembutal anesthesia reduces and even eliminates the response of the rabbit to renin, to what extent is the anesthetic affecting the results obtained with dogs?

I. *Influence of the Starting Pressure on the Response to Renin.* The results of 200 assay tests, covering some 20 lots of extract irrespective of differences in potency, have been grouped according to the initial blood pressures of the dogs used. Although these pressures varied from 80 to 160 mm Hg, over 80% were between the limits of 95 and 130 mm Hg, which can be considered the normal range.[4] All renin doses of 0.3 mg per kg body weight or more gave blood pressure responses not significantly different from each other. These maximal responses have been grouped together in the overdosage category.

Table I shows that an inverse correlation exists between the blood pressure rises evoked by overdosages of renin and the starting

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1 Swingle, W. W., Taylor, A. R., Collings, W. D., and Hays, H. W., *Am. J. Physiol.*, 1939, **127**, 768.

2 Wakerlin, G. E., and Chobot, G. R., *Am. J. Physiol.*, 1939, **126**, 646.

3 Pickering, G. W., and Prinzmetal, W., *Clin. Sci.*, 1938, **3**, 211.

4 These represent mean pressures, taken by the needle puncture method (Parkins, W. M., *Am. J. Physiol.*, 1934, **107**, 518).

TABLE I.
Correlation Between Initial Blood Pressure and Response to Renin in the Anesthetized Dog.

Initial blood pressure mm Hg	Overdosage			0.2 mg/kg body wt			0.1 mg/kg body wt		
	No. of tests	Blood pressure rise mm Hg	Blood pressure peak mm Hg	No. of tests	Blood pressure rise mm Hg	Blood pressure peak mm Hg	No. of tests	Blood pressure rise mm Hg	Blood pressure peak mm Hg
81- 90	7	125±3.7	211±3.8	1	93	184	2	68	157
91-100	19	119±3.2	209±3.1	8	84±7.6	177±5.6	11	53±2.8	150±3.3
101-110	30	104±2.5	212±2.4	10	72±3.2	177±3.0	9	51±5.4	159±5.3
111-120	32	92±2.4	213±2.3	8	68±2.9	183±2.8	7	42±3.3	159±2.9
121-130	12	79±4.5	207±3.4	6	67±3.5	192±3.3	11	46±2.7	169±2.3
131-140	4	77	213	9	46±2.8	183±2.8	6	31±3.1	165±2.9
141-150	1	60	208	4	43	187	3	37	180
Avg	105*	101±1.7	211±1.5	46*	65±2.2	183±1.7	49*	47±1.5	161±1.6

*Totals.

pressure levels. In other words, the maximum rise is limited by a fairly constant maximum peak pressure. Such a clear correlation between rise and starting pressures is not so evident with the lower doses of renin. There is some indication that the rise decreases as the initial pressure becomes greater, but within the normal starting pressure range the trend is not sufficient to warrant correcting assay results for these differences. In most groups the peak pressures obtained show less variability than do the rises. With an assay based on peak pressures, too, corrections probably need not be made for differences in initial pressures.

II. *Effect of Repetition of Renin Tests on the Same Dog.* A single lot of renin extract, containing 5.1 mg solids (0.8 mg N) per cc, was used for repeated tests on 6 dogs, An interval of at least 72 hours was allowed between tests under anesthesia, for an abnormal depression of the initial blood pressure has been sometimes observed with more frequent use of anesthesia. An interval of 24 hours was allowed between tests on control animals without anesthesia. These intervals seemingly had no effect on the results. The data given in Table II are indicative of the variation in response to be expected with a random selection of animals. In general, repeated tests on the same animal tend to be more consistent than tests on different dogs, but the results are still far from constant. There was no evidence that the use of an animal for assays over long periods of time affected the reliability of the results. For example, one dog was used for 18 tests in 8 weeks, and at no time showed evidence of the development of either a refractoriness or an increased sensitivity to the renin.

TABLE II.
Effect of Nembutal Anesthesia on the Blood Pressure Response of the Dog to Renin Injections.

Dose per kg body weight γ	Unanesthetized				Anesthetized			
	Dog	Initial blood pressure mm Hg	Peak blood pressure mm Hg	Rise in pressure mm Hg	Dog	Initial blood pressure mm Hg	Peak blood pressure mm Hg	Rise in blood pressure mm Hg
694*	1	96	175	79	1	94	257	163
		108	185	77		97	231	133
		100	185	85		102	229	127
		105	191	86		91	232	141
780*	2	113	215	102	2	130	191	61
		112	196	84		112	185	73
		122	206	84		116	215	99
		119	217	98		114	210	96
		112	199	87		109	204	95
50	1	100	170	70	1	99	145	46
		105	172	67		109	145	36
		106	168	62		110	153	43
		109	164	55		86	147	61
25	2	124	167	43	2	108	162	54
		120	151	31		102	143	41
		107	152	45		125	157	32
25	3	113	171	58	3	120	161	41
		126	172	46		131	165	34
		123	172	49		107	155	48
25	4	118	141	23	4	123	142	19
		118	148	30		129	154	25
		122	158	36		126	150	24

*1.5 cc extract (overdosage).

III. *Effect of Nembutal Anesthesia on the Response to Renin.* Five dogs in the previous series were given overdosages of renin both with and without anesthesia. Two of these (*e. g.*, Dog No. 1, Table II) showed significantly lower peak pressures and smaller rises when not anesthetized, a difference marked solely by a sudden interruption of the rise and a sustained holding of the pressure level at around 180 mm Hg in the unanesthetized tests. To check on this anesthesia difference, overdosages of the same extract were given 8 anesthetized and 7 unanesthetized dogs. The average blood pressure rises were 100 ± 3.4 and 70 ± 1.6, the peak pressures 211 ± 3.1 and 176 ± 2.3, respectively. This depression of the maximum peak pressure is present in enough dogs to make an assay of large doses of renin in the unanesthetized dog uncertain.

When smaller doses of renin were used, there was, in 2 dogs

(Nos. 1, 3, Table II) some indication of a depression of the rise with anesthesia. The other 3 showed no consistent difference between tests with and without anesthesia. Anesthesia has no definite effect on the variability of response in the dog.

For most routine testing, the anesthetized dog is the preferable test animal. If a sufficient time interval is allowed between tests, the repeated use of 2 or 3 dogs affords as accurate an assay as one based on 1 test each on a large series of dogs. Assays in both anesthetized and unanesthetized animals will show large variation, and a minimum of 10 tests is required for accuracy. Where the rise expected is clearly sub-maximal, the more rapid assay on unanesthetized dogs can be substituted.

Summary. The magnitude of the blood pressure rises given by large doses of renin must be corrected for differences in starting pressure levels, while those given by smaller doses need not be. An assay based on repeated tests on 2 or 3 dogs is as reliable as one based on a larger series of animals. Nembutal anesthesia may affect, in some dogs, the magnitude of the rise after large renin doses, but usually has little effect on the rise after small doses. For most routine testing, the anesthetized dog is the preferable test animal.

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Labyrinthine Disregard after Removal of the Caudate.*

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In an earlier research upon neostriatal function[1] the present authors observed that "It is difficult to escape the suspicion that animals (cats) with neostriatal injuries suffer from some variety of vestibular disturbance"; the literature relating to this observation was reviewed and it was noticed that Bergouignan and Verger[2] found that the ipsilateral circus movement (dogs) produced by unilateral caudate lesion was accentuated by rotation to the same side while rotation to the opposite side gave no circus movement. Cocainization of the labyrinth of either side always produced an accentuation of

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1 Mettler, Fred A., and Mettler, Cecilia C., *Role of the Neostriatum,* in press.

2 Bergouignan, M., and Verger, P., *Compt. rend. soc. de Biol.,* 1935, **118**, 1539.