Effect of Veronal Anaesthesia upon the Epinephrine Secretion and the Blood Sugar Concentration.

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The velocity with which the epinephrine is liberated from the suprarenal gland of dogs, non-anaesthetized is not subject to change towards a particular direction by applying various kinds of anaesthetics, such as ether, urethane, morphine, chloralose and avertin.¹⁾

In this experiment veronal was tested in the same manner.

In the dogs, in which particular regions were previously deafferented, that is D_{10} - L_3 dorsal roots severed, the lumbar route operation was carried out for collecting suprarenal vein blood samples without anaesthetizing, evoking any pain and opening the abdominal cavity. The epinephrine concentration was estimated by means of the rabbit intestine segment method and the blood sugar by the Hagedorn and Jensen method.

„Veronal-Natrium“ of E. Merck, was dissolved freshly before use in 0.9% NaCl-solution so as to contain 15–17%, and intravenously injected in doses of 0.16, 0.25, 0.3 and 0.47 grm. per kilo of body weight.

The poisoning symptoms corresponded to the amount given, that is in Exp. 1 (0.16 grm. per kilo) and Exp. 4 (0.25 grm. per kilo) poisoning was not so profound; in Exp. 1 the reflexes never disappeared, the animal never slept profoundly. The nictating membrane protruded largely about one hour after veronal, but two hours later it was apt to recover almost completely, and in Exp. 4 the reflexes and the muscle tone also did not wholly disappear, but the sleep was deeper com-

1) Kodama, J. of Biophysics, 1924, 1, 79; Sato and Aomura, Tohoku J. of Exp. Med., 1930, 15, 17; Sato and Ohmi, Ibid., 1933, 21, 411, 433; Sato, Satow and Degti, Ibid., 1934, 24, 485.

TABLE.

The epinephrine output, blood sugar content, etc. after intravenous injection of veronal in de-afferented dogs.

Number of specimen	Time of collection (min.)	Blood flow (c.c.)				Epinephrine (mgrm.)			Blood sugar (%)	Body temperature (°C.)	Room temperature (°C.)	Heart beat per minute	Frequency of respiration per minute	
		quantity	duration (sec.)	quantity per minute	quantity per kilo per min.	content in 1 c.c. blood	output							
							per minute	per kilo per minute						
Dog 1. 15.2 kilos ♀. 11. IX. 1933. (2. V. 1933. 11.7 kilos. D ₁₀ -L ₃ dorsal spinal roots severed.) 10:00-11:00 a.m. Left lumbar route preparation. 1:20 p.m. Experimented on as follows.														
I	before	10	2.1	60	2.1	0.14	0.000075	0.00016	0.000010	0.128	39.5	19.5	144	24
II		9	2.3	60	2.3	0.15	0.000075	0.00017	0.000011	0.134				
1:30 p.m. 16 c.c. veronal solution (3 grms. veronal in 20 c.c. of 0.9% NaCl-solution) injected into v. saphena dextra. (About 0.16 gm. per kilo) Soon later the animal quiet.														
III	after	15	1.9	60	1.9	0.13	0.000075	0.00014	0.000009	0.121	39.0	19.5	180	24
IV		16	2.1	60	2.1	0.14								
V		50	1.7	60	1.7	0.11	0.0001	0.00017	0.000011	0.135				
VI		51	1.8	60	1.8	0.12					37.7	20	180	25
VII		90	1.1	60	1.1	0.07	0.0001	0.00011	0.000007	0.126				
VIII		91	1.1	60	1.1	0.07					39.5	20	235	38
IX		130	1.0	60	1.0	0.07	0.0001	0.00010	0.000007	0.126				
X		131	0.8	60	0.8	0.05					39.7	20	235	33
XI		160	1.2	60	1.2	0.08	0.0001	0.00012	0.000008	0.114				
XII		161	1.3	60	1.3	0.09					39.3	19	228	48
XIII		190	1.4	60	1.4	0.09	0.0001	0.00014	0.000009	0.123	39.6	19	234	48
4:50 p.m. Shed about 80 c.c. blood from lumbodorsal artery. 5:00 Cut off the left splanchnic nerv. The wound sewed up. Anaesthesia shallow. 1:45 Reflexes +; nictating membrane was stretched. 2:25 Muscle tone +; shivering; reflexes +. 3:0 and 4:10 Almost as above.														
Dog 2. 14.0 kilos ♂. 18. X. 1933. (11. V. 1933. 12.8 kilos. D ₁₀ -L ₃ dorsal spinal roots severed.) (Both limbs quite normal.) 10:00-11:00 a.m. Left lumbar route preparation.														
I	before	15	2.2	40	3.3	0.24	0.00005	0.00016	0.000012	0.102				
II		14	1.7	30	3.4	0.24	0.00005	0.00017	0.000012	0.102	38.8	21.5	138	24
12:15-12:17 25.2 c.c. veronal solution (30 c.c. of 0.9% NaCl-solution containing 5 grms. veronal) injected into the right v. saphena (0.3 gm. per kilo). 5 minutes later the animal quiet. Reflexes +. Nictating membran was stretched. Muscle tone +.														
III	after	15	2.0	30	4.0	0.29	0.00005	0.00020	0.000014	0.086				
IV		16	2.0	30	4.0	0.29					37.5	22	108	32
V		30	1.6	30	3.2	0.23	0.000075	0.00025	0.000018	0.091				
VI		31	1.8	30	3.6	0.26					37.1	22.5	138	30

Number of specimen	Time of collection (min.)	Blood flow (c.c.)				Epinephrine (mgrm.)		Blood sugar (%)	Body temperature (°C.)	Room temperature (°C.)	Heart beat per minute	Frequency of respiration per minute	
		quantity	duration (sec.)	quantity per minute	quantity per kilo per min.	content in 1 c.c. blood	output per minute						per kilo per minute
	after												
VII	60	1.4	30	2.8	0.20	0.000075	0.00021	0.000015	0.093	36.1	22.5	138	30
VIII	61	1.4	30	2.8	0.20								
IX	90	1.2	30	2.4	0.17								
X	91	1.3	30	2.6	0.19	0.000075	0.00019	0.000014	0.100	35.9	23	138	20
XI	120	1.2	30	2.4	0.17								
XII	121	1.0	30	2.0	0.14								
XIII	180	0.7	30	1.4	0.10	0.000010	0.00012	0.000009	0.102	35.6	22.5	144	21
XIV	181	0.6	30	1.2	0.09								
XV	181	0.5	30	1.0	0.07								
XVI	240	2.1	60	2.1	0.15	0.000010	0.00020	0.000015	0.100	35.4	22.5	156	27
XVII	241	1.0	30	2.0	0.14								
4:23 Shed about 80 c.c. blood from the lumbo-dorsal artery.													
4:30 The wound sewed up.													
12:30 Quiet, reflexes +, anaesthesia somewhat deep, pupils small, nictating membran was stretched, respiration deep.													
1:15 Slept. 1:40 Muscle tone ±. 2:15 Muscle tone —, reflexes ±.													
2:45 Shivering. 3:15 Muscle tone +. 4:15 Pupils small; reflexes +; anaesthesia became somewhat shallow.													
Dog 3. 11.0 kilos ♀. 15. XI. 1933.													
(26. IV. 1933. 13.0 kilos. D ₉ -L ₃ dorsal spinal roots severed.)													
(VIII. '33 6 youngs were born.)													
9:45-10:40 Left lumbar route preparation; both hind legs quite normal.													
12:20 Experimented on as follows:—													
I	before												
II	5	2.8	30	5.6	0.51	0.00005	0.00028	0.000025	0.112				
	4½	3.0	30	6.0	0.55	0.00005	0.00030	0.000026		39.1	18	108	20
12:25-12:30 c.c. veronal solution (35 c.c. of 0.9% NaCl-solution containing 6 grms. veronal) introduced into v. saphena dextra. (About 0.47 gm. per kilo). Nictitating membrane was stretched.													
III	after												
IV	30½	1.8	30	3.6	0.33	0.00005	0.00018	0.000016	0.108				
V	60	1.8	40	2.7	0.25	0.00005	0.00013	0.000012	0.112	36.9	19.5	135	10
VI	61	1.2	30	2.4	0.22					36.0	19	105	8
VII	90	2.2	40	3.3	0.30				0.119				
VIII	91	1.8	30	3.6	0.33	0.00005	0.00018	0.000016		35.3	18	90	8
IX	120	2.4	30	4.8	0.44	0.00005	0.00024	0.000022	0.114				
X	121	2.2	30	4.4	0.40					34.2	18.5	96	8
3:00 Died. Indifferent blood collected from v. cava. inf.													
Before the veronal injection, heart rate irregular.													
5 minutes after the injection, laid down weakly. 1:00 Heart rate regular, pupil small, corneal reflex ±, muscle tone —. 1:30 Lay quietly, the other conditions same. Colour of specimen V.-X. dark.													
2:30 Respiration irregular.													
Intestine tracings reproduced for example.													
Dog 4. 16.8 kilos ♀. 13. XII. 1933.													
(2. V. 1933. 11.7 kilos. D ₁₀ -L ₃ dorsal spinal roots severed.													
11. IX. " 15.2 " Left lumbar route preparation for veronal experiment: Dog 1. L splanchnicotomy.)													
10:00-11:30 a.m. Right lumbar route preparation.													

Number of specimen	Time of collection (min.)	Blood flow (c.c.)				Epinephrine (mgrm.)			Blood sugar (%)	Body temperature (°C.)	Room temperature (°C.)	Heart beat per minute	Frequency of respiration per minute
		quantity	duration (sec.)	quantity per minute	quantity per kilo per min.	content in 1 c.c. blood	output						
							per minute	per kilo per minute					
		1:55 p.m. Experimented on as follows:—											
		before											
I	15	2.8	40	4.2	0.25	0.0002	0.00084	0.000050	0.110				
II	14	2.2	30	4.4	0.26	0.0002	0.00088	0.000052	0.110	39.6	20	126	21
		2:08-2:10 p.m. 25.2 c.c. of veronal solution (5 grms. veronal sodium in 30 c.c. of 0.9% NaCl-solution) injected into v. saphena sin. (0.25 gm. per kilo).											
		after											
III	15	1.5	60	1.5	0.09	0.000075	0.00010	0.000006	0.117	39.1	21	150	36
IV	16	1.3	60	1.3	0.08								
V	35	0.5	60	0.5	0.03								
	36	0.5	60	0.5	0.03	0.000010	0.00005	0.000003	0.116	38.8	22	198	36
VII	75	0.7	60	0.7	0.04								
VIII	76	0.5	60	0.5	0.03								
IX	100	0.6	60	0.6	0.04	0.000010	0.00006	0.000004	0.101	39.5	22	222	54
X	101	0.6	60	0.6	0.04								
XI	140	1.5	60	1.5	0.09								
XII	141	1.4	60	1.4	0.08	0.000030	0.00043	0.000026	0.116	40.0	20	228	60
	152	(Arterial blood.)											
	4:42	Final bleeding from the femoral artery.											
	4:45	Died.											
		Soon after the injection, the animal laid down. 15 minutes after the injection, somewhat excited, nictating membran was stretched, corneal reflex +, pupil reflex +, pupils large.											
	2:30	The animal laying quietly; muscle tone +.											
	3:0	Shivering on the whole body, slept.											
	3:30	Reflexes +, muscle tone +. Short inspiration and slow expiration.											
		Specimen IX and X a little dark.											
	4:30	Reflexes +, muscle tone +, moved the head and paw.											
	4:34	Looked a little excited.											

pared with Exp. 1. A short time after injecting veronal solution the animal was slightly excited, 2 hours after veronal the animal appeared somewhat excited, that is the recovery already had set in at this time.

When 0.3 gram. veronal per kilo was dosed, the poisoning was more severe; the reflexes became very stupid, the muscle tone disappeared and sleep was profound. 4 hours after the injection some recovery was noted. 0.47 gram. veronal per kilo produced a too severe poisoning which resulted in the death of the animal.

The blood flow from the cannula inserted into the lumbo-supra-renal vein diminished gradually on receiving veronal in the doses above given. The initial values were 0.15 c.c. per kilo per minute, 0.24 c.c., 0.55 c.c. and 0.26 c.c. After veronal it was reduced to 0.05 c.c. per kilo per minute (2 hours), 0.07 c.c. (3 hours), 0.22 c.c. (1 hour) and 0.03 c.c. (half an hour). The figures in parentheses refer to the

Intestine tracings for Dog 3.(Reduced to $\frac{2}{3}$)

In all the intestine tracings, at the mark "X" atropine-Tyrode's solution, in which the rabbit intestine segment was beating rhythmically, was replaced by indifferent blood solution, and at the "numeral" the indifferent blood solution to which a certain quantity of adrenalin chloride of Sankyo Co. was added, or by the blood specimen solution. All the blood solutions were prepared by diluting with 4 volumes of Tyrode's solution, and the quantity of the blood employed for one assay was 0.5 c.c.

The numeral of specimens and the quantity of adrenaline solution which is showed in c.c. and in concentration were added to each observation. For example, "0.1 1/2000" shows "0.1 c.c. of adrenaline solution with the concentration of 1/2000 mgrm. in 1 c.c." i.e. 0.00005 mgrm. adrenaline. To show the I specimen, we used the numeral "I"

In all tracings, time intervals are 30 seconds.

Fig. a. I: Weaker than 0.00005 mg. and a little weaker than 0.000025 mg. II: As strong as 0.000025 mg. and a little stronger than I. IV: About the same with 0.000025 mg. and weaker than 0.00005 mg.

Fig. b. V: Weaker than 0.00005 mg. and a little weaker than or almost as strong as 0.000025 mg. VIII: As strong as 0.000025 mg. IX: As strong as VIII (0.000025 mg.) and a little weaker than 0.00005 mg.

To sum up:—

I: Weaker than 0.00005 mg. and a little weaker than 0.000025 mg. It was assayed at 0.00005—mg. per 1 c.c. II: As strong as 0.000025 mg. and a little stronger than I. It was taken as 0.00005 mg. per 1 c.c. IV: About the same with 0.000025 mg. and weaker than 0.00005 mg. It was taken as 0.00005 mg. in 1 c.c. V: Weaker than 0.00005 mg. and a little weaker than or almost as strong as 0.000025 mg. It was assayed at 0.00005 mg. per 1 c.c. VIII: As strong as 0.000025 mg. That is 0.00005 mg. per 1 c.c. IX: As strong as VIII (0.000025 mg.) and a little weaker than 0.00005 mg. It was taken as 0.00005 mg. in 1 c.c.

time needed to arrive at the minimum rate after the veronal application. Thereafter the rate tended again to recover.

The concentration of the suprarenal blood remained during veronal narcosis nearly unaltered; in Exps. 1 and 2 it increased but only

very insignificantly, in Exp. 3 it remained entirely unaltered, and in Exp. 4 it underwent small variations, sometimes a small reduction, sometimes a small augmentation.

Consequently the epinephrine output per minute was measured reduced by the veronal anaesthesia though in a small degree.

0.00001 mgrm. per kilo per minute before veronal and 0.000007–0.000011 mgrm. after it in Exp. 1; 0.000012 mgrm. before and 0.000009–0.000018 mgrm. after in Exp. 2; 0.000025 mgrm. before and 0.000012–0.000022 mgrm. after in Exp. 3, and 0.00005 mgrm. before and 0.000003–0.000026 mgrm. after in Exp. 4.

The epinephrine secretion was found insignificantly reduced during the veronal anaesthesia. The poisoning symptoms in these dogs were of various degree according to the dose given, but no relation can be detected between the degree of poisoning and the rate of epinephrine secretion, because there was not detectable any definite difference in the latter in these four experiments.

The blood sugar concentration was not altered by the veronal administration in a dose varying from 0.16–0.47 grm. per kilo of body weight.

In Exp. 1, it was measured as 0.128% 10 minutes before veronal, and on receiving the veronal it fluctuated between 0.114–0.135% in a lapse of 3 hours; in Exp. 2 a small reduction such as from 0.102% to 0.086% and 0.091% occurred within half an hour after veronal injection, but one hour later a complete recovery was estimated in the sugar concentration. In Exp. 3 no variation was witnessed in the blood sugar content throughout the whole experimental course. About the same was noted in Exp. 4. Some variations, strictly speaking, were measured in the blood sugar concentration during the veronal anaesthesia, but they were of an insignificant degree and of various signs.

During the veronal anaesthesia a body temperature fall was brought about and its magnitude depended upon the amount given. When 0.16 grm. per kilo (Exp. 1) and 0.25 grm. (Exp. 4) were given, the fall occurred only temporarily such as 1 hour or 1.5 hours, and its degree was never large, while 0.3 grm. veronal per kilo produced a large and long lasting decrease, and 0.47 grm. per kilo was capable of lowering the body temperature to 34° within two hours and half an hour later the animal eventually died. In spite of detrimental conditions of the animal, such as a profound body temperature fall, relaxed muscles, weakened reflexes, disturbed circulation (blood very

dark), etc., the blood sugar concentration was not altered at all and the epinephrine output rate was never measurable as increasing. Only a little decrease was noted in the latter. Before veronal the rate of secretion was 0.000025 mgrm. per kilo per minute and the minimum during the anaesthesia 0.000012 mgrm. These figures are very much like those obtained in this Laboratory when a dog was sleeping,²⁾ 0.00001–0.000012 mgrm. during sleep against 0.000018–0.00003 mgrm. during quiet but not a sleeping period.

The heart rate decreased for a while when small doses were given (Exps. 1 & 4), but later it increased and finally far exceeded the initial rate, seemingly followed by an increasing of the body temperature. No large increase of the heart rate was noted in Exps. 2 and 4, in which the drug was given in somewhat large doses and the body temperature continued to fall. The respiration rate inclined to increase in Exps. 1 and 4, while it remained nearly unaltered in Exp. 2 and distinctly decreased in Exp. 3; such variations of the respiratory frequency correspond well to the general condition of animals and to the bearing of the body temperature.

SUMMARY.

Veronal was injected intravenously into dogs, non-anaesthetized in doses varying from 0.16 to 0.47 grms. per kilo of body weight. The suprarenal vein blood collected without and with narcosis, fastening or laparotomy was tested for epinephrine by means of the rabbit intestine segment method.

The rate of epinephrine secretion from the suprarenal gland showed a tendency to reduce on poisoning, but only very slightly. And the blood sugar concentration remained practically unaltered.

2) Satake, Watanabé and Sugawara, *Tohoku J. of Exp. Med.*, 1927, 9, 7.