PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

On the Action of the Venom of Bungarus coeruleus (The Common Krait)

R. H. Elliot, W. C. Sillar and George S. Carmichael

Phil. Trans. R. Soc. Lond. B 1905 **197**, 327-345
doi: 10.1098/rstb.1905.0011

XI. *On the Action of the Venom of* Bungarus Cœruleus (*the Common Krait*).

By Major R. H. Elliot, *I.M.S.*;* W. C. Sillar, *M.B.*, *B.Sc.*; *Lecturer on Experimental Pharmacology and Senior Assistant in the Materia Medica Department of the University of Edinburgh; and* George S. Carmichael, *M.D., University Assistant in the Materia Medica Department of the University of Edinburgh.*

Communicated by Professor Sir Thomas R. Fraser, *M.D., F.R.S.*

(From the Pharmacological Laboratory of the University of Edinburgh.)

Received May 12,—Read June 9, 1904.

Contents.

	Page
Previous Work recorded	327
Determination of M.L.D. Frog	328
,, ,, Rabbit	329
,, ,, Rat	330
Protective Power of Calmette's Serum	330
Effect upon Nerve-endings	331
Examination of Blood	333
Perfusion of Frog Vessels	333
,, ,, Ventricle	333
Kymographic Experiments	335
Summary of Conclusions	344

The following experimental research upon the pharmacological action of the venom of the common Krait (*Bungarus cæruleus*), was performed in the Pharmacological Laboratory of the University of Edinburgh, and we are greatly indebted to Sir Thomas R. Fraser, not only for facilities in the performance of that work, but also for the encouragement he has uniformly given us.

Previous examination of the action of this venom appears to be limited to the work recorded in the papers recently published by Captain Leonard Rogers, I.M.S.† He states that the symptoms following poisoning by this venom are identical with those following poisoning by Cobra venom, whereas our results, as shown below, seem to indicate that while these symptoms are similar, still they differ so much in relative degree as to render it doubtful if they can in future be spoken of as identical.

* On special duty for Snake Venom research under the orders of the Secretary of State for India.

† 'Indian Medical Gazette,' vol. 39, No. 1, January, 1904, p. 30; 'Lancet,' February 6, 1904, p. 350.

For the Krait venom used in the experiments we are greatly indebted to the Madras Government, who placed Dr. P. P. PINTO on special duty for the purpose of collecting venom. We take this opportunity of expressing our thanks to this gentleman, for we are aware how difficult it is to obtain specimens of this poison.

The total amount available was 0·077 of a grm., which was the venom furnished by expression from the dissected glands of two snakes. This gives a total of 0·0385 of a grm. per snake and corresponds very closely with what one of us has found to be the ordinary yield of this species. It is obviously much less than the average yield from a large Cobra.

Experiments were performed :—

(1) To determine the minimum lethal dose of the dried venom for frogs and small mammals—rats and rabbits being chosen.

(2) To determine the protective power of CALMETTE'S anti-venomous serum against Krait venom compared with its power of antagonising Cobra venom.

(3) To determine whether any intravascular clotting or hæmolysis occurred in the circulating blood, following a lethal dose.

(4) To determine the action of the venom on the isolated vessels of the frog.

(5) To determine the action of the venom on the isolated ventricle of the frog.

(6) To ascertain generally the causes which influence the vital functions of mammals exposed to the action of the venom and which bring about death.

EXPERIMENTS to determine the Minimum Lethal Dose of Krait Venom for Frogs (*Rana esculenta*). Solution used was 0·0001 grm. in 1 cub. centim. of RINGER'S Fluid.

No.	Dose per kilo.	Weight of frog.	Remarks.
	grm.	kilo.	
1	0·002	0·047	3 hours after injection, frog prone; respiration very shallow; lower eyelids fully and permanently raised. Died 22 hours after injection.
2	0·0015	0·044	2 hours after injection, frog prone; respiration very irregular; eyelids fully raised. Died within 22 hours.
3	0·001	0·041	4 hours after injection, respiration normal; eyelids three-quarters raised. 22 hours after injection respiration ceased; eyelids fully raised. Died in 3 days.
4	0·0005	0·039	3 hours after injection, eyes open; respiration easily visible. 22 hours after injection, eyelids raised; frog prone; can turn over; reflexes present; respiration shallow. Died in 3 days.

EXPERIMENTS to determine the Minimum Lethal Dose by Subcutaneous Injection of Krait Venom for Rabbits. The solution used was 0·0001 grm. in 1 cub. centim. of RINGER'S Fluid.

No.	Dose per kilo.	Weight of rabbit.	Remarks.
	grm.	kilo.	
1	0·00125	1·420	Died in 4 hours. The phrenic nerves responded to an induction current with the coils distant 450 millims. The vagi responded at 140 millims., and the left sciatic at 220 millims.
2	0·001	1·580	Died in 6 hours. Phrenics responded at 220 millims. Right vagus at 20 millims.; left vagus at 100 millims. Right sciatic at 220 millims.
3	0·0009	1·630	Died in 5 hours. Phrenics failed to respond with coils separated more than 60 millims. Vagi gave no complete response to maximal stimulus.
4	0·00075	1·590	Died in 2 hours.
5	0·0007	1·680	Died in 5 hours. Phrenics responded at 420 millims.; vagi at 100 millims.
6	0·0006	1·625	Died in 6 hours.
7	0·0005	1·975	Died in $4\frac{1}{2}$ hours. Right phrenic responded at 420 millims.; left phrenic at 220 millims.
8	0·0004	2·220	Died in $5\frac{1}{2}$ hours. Phrenics responded at 390 millims.; vagi at 80 millims.
9	0·0002	2·460	Died within 15 hours.
10	0·00015	1·375	Died within 21 ,,
11	0·0001	2·120	Died within 21 ,,
12	0·00009	2·110	Died within 21 ,,
13	0·00008	1·960	Died within 21 ,,
14*	0·00007	1·860	Recovered; moderately ill.
15	0·00006	1·840	Recovered. Very ill for 2 days; extremities paralysed.
16	0·00004	2·120	Recovered.

* The solution of venom used in Experiment 14 was 1 day old.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

EXPERIMENTS to determine the Minimum Lethal Dose by Subcutaneous Injection of Krait Venom for Rats. The solution used was 0·0001 grm. in 1 cub. centim. of RINGER'S Fluid.

No.	Dose per kilo.	Weight of rat.	Remarks.
	grm.	kilo.	
1	0·004	0·087	Died in 3 hours.
2	0·003	0·053	Died in 1½ „
3	0·002	0·079	Died in 7–9 hours.
4	0·001	0·072	Died in 3½ hours. The phrenic nerves at death failed to respond to an induction current with the coils distant more than 220 millims. The right vagus partially responded at 40 millims. Left vagus nil.
5	0·0009	0·244	Recovered after being very ill.
6	0·0008	0·153	Recovered.
7	0·0007	0·157	Recovered.
8	0·0006	0·155	Recovered.

A point of special interest is the rise of the lower eyelids. This is due to an elastic recoil of the lids when the depressor of the lower eyelids is paralysed. The interest attached to this point lies in the fact that Cobra venom and another Colubrine venom produce the same symptom, while it is invariably absent in the course of Daboia poisoning.

We have therefore concluded that the minimum lethal dose of our specimen of dried Krait venom was, in the case of the frog, about 0·0005 grm. per kilo.; in the case of the rat, 0·001 grm. per kilo., and in the case of the rabbit as low as 0·00008 grm. per kilo. Our supply of the venom was too small to allow us to determine the lethality in other animals.

The relatively great lethal action of Krait venom for the rabbit as compared with the rat is remarkable and shows a great divergence from the relative lethality of Cobra venom for the same animals as determined by one of us, who found the M.L.D. of a specimen of Cobra venom for rats 0·0005 grm. per kilo., and for rabbits 0·0006 grm. per kilo.

EXPERIMENTS to demonstrate the Protective Power of CALMETTE'S Antivenomous Serum against (1) Cobra Venom and (2) Krait Venom.

The solution of dried Cobra venom in RINGER'S fluid used was of a strength of 0·0001 grm. in 10 cub. centims.; and the solution of Krait venom used was of a

strength of 0·0005 grm. in 1 cub. centim. Equivalents to 10 M.L.D. were mixed with antivenomous serum for 30 minutes before injection into animals. Rats were used and the M.L.D. of Cobra venom taken was 0·0005 grm. per kilo. and of Krait venom 0·0009 grm. per kilo.

Krait venom experiments.				Cobra venom experiments.			
Weight of rat in kilos.	Venom per kilo.	Serum per kilo.	Results.	Weight of rat in kilos.	Venom per kilo.	Serum per kilo.	Results.
	grm.	cub. centims.			grm.	cub. centims.	
0·108	0·009	2	Dead in 4 hours.	0·183	0·005	2	Dead in 6 hours.
0·129	0·009	3	Dead in $2\frac{3}{4}$ „	0·175	0·005	3	Dead in 45 „
0·120	0·009	4	Dead in $1\frac{3}{4}$ „	0·188	0·005	4	Dead in 45 „
0·112	0·009	5	Dead in $2\frac{1}{2}$ „	0·166	0·005	5	Remained well.
				0·087	0·005 (Control.)	nil	Dead in $1\frac{3}{4}$ hours.

Calmette's antivenomous serum therefore, in the amounts effectual against Cobra venom, appears to be ineffectual against Krait venom.

The integrity of the endings of the phrenic, vagi and sciatic nerves was examined in the majority of animals which died, the various nerves being exposed just at death. Induction stimuli were applied, and invariably there was noted definite diminution in the response to such stimuli—fibrillary twitchings of the diaphragm, for example, occurring with a stimulus to the phrenics where in the normal animal a complete contraction of that muscle would occur; and the vagal stimulation required to stop the heart was almost invariably greater than that required in the case of normal animals, whilst in some even the maximal stimulus failed to do more than slow the heart. In the case of rabbit No. 15, which recovered, there was definite evidence of paralysis of the extremities. This was at first more noticeable in the fore limbs, and the paralysis seemed to pass off in the same order, the hind limbs being the last to recover.

The comparative effect on skeletal muscle and nerve of the action of the venom was investigated on the frog's gastrocnemius with the sciatic nerve attached. The sensitiveness to stimulation by a Faradic current was ascertained in the case of the nerve and the muscle respectively, and the nerve was then placed in a fresh solution of the Krait venom dissolved in 0·65 per cent. tap-water saline where it remained for 22 hours, the muscle being carefully kept from contact with the poison and placed in normal saline. The strength of the venom solution used was 1 : 10,000. At the end of 14 hours the excitability of the nerve was unimpaired and at the end of 22 hours was only very slightly impaired.

At the same time, with a similar nerve-muscle preparation, the muscle was placed

in the venom solution, and in 2 hours and 20 minutes it had become much less excitable, though the excitability of the nerve was unimpaired. At the end of $3\frac{3}{4}$ hours no response could be elicited by stimulating the nerve, and it required strong direct stimulation of the muscle to cause any contraction. After $5\frac{1}{4}$ hours no contraction of any part of the muscle occurred with the strongest direct stimulation. It will be observed therefore, that the muscle was gradually poisoned. The ends of the nerve within the muscle appeared to have been poisoned before the muscle had lost its power to contract and before the nerve trunk was appreciably affected.

In order to define more nearly the loss of excitability of the nerve and muscle a similar experiment was performed with more frequent observations. The same characteristics in the action were observed. After 2 hours the excitability of the nerve was unimpaired, but during the next 20 minutes it diminished rapidly and was entirely lost. The muscle had gradually become less responsive, and finally ceased to contract even on the strongest stimulation, 1 hour after the nerve ends had been poisoned and $3\frac{1}{2}$ hours after the muscle had been placed in the poison. Immediately after death the muscle was found when tested by litmus paper to be acid in reaction.* Our previous conclusions are therefore confirmed.

EXPERIMENTS to determine the Minimal Stimulation required to produce Muscular Contraction, measured by the Distance between the Secondary and Primary Coils, expressed in millimetres.

Time.	Poisoned muscle.	Protected nerve of poisoned muscle.	Poisoned nerve of protected muscle.
Before poisoning	230	390	390
hrs. m.			
0 30 after poisoning . . .	230	385	405
1 0 ,, ,, . . .	210	380	—
1 30 ,, ,, . . .	210	400	—
2 0 ,, ,, . . .	170	420	410
2 5 ,, ,, . . .	160	440	—
2 10 ,, ,, . . .	140	170	—
2 25 ,, ,, . . .	130	0	415
2 50 ,, ,, . . .	100	0	—
3 5 ,, ,, . . .	70	0	—
3 10 ,, ,, . . .	50	—	—
3 15 ,, ,, . . .	30	—	—
3 20 ,, ,, . . .	0	—	—
14 0 ,, ,, . . .	...	...	400
22 0 ,, ,, . . .	...	...	305

The intermediate superfluous readings are omitted.

* The indication given of a direct action on muscle structure by the early occurrence of an acid reaction was pointed out by FRASER, "On the Kombé Arrow Poison (*Strophanthus hispidus*)," 'Roy. Soc. Edin. 'Soc.,' 1869–70, p. 102.

The blood was examined in several cases immediately after death and in no case was any appearance of ante-mortem clotting found. Blood films from blood taken from the left side of the heart of rats immediately after death following a lethal dose of Krait venom showed, on examination of the red blood corpuscles, no evidence of hæmolysis having occurred. The corpuscles showed no abnormality, and serum separating spontaneously from blood in capillary tubes showed no hæmoglobin staining. The blood even from the left side of the heart was extremely venous in appearance.

Action on Blood Vessels.

One of us has shown that Cobra venom produces a marked constriction of the blood vessels of the frog when perfused through them in very dilute solution, the limit of this being reached at a strength of 1 : 10,000,000. It is of interest, therefore, to compare a similar action which we have found to be produced by Krait venom.

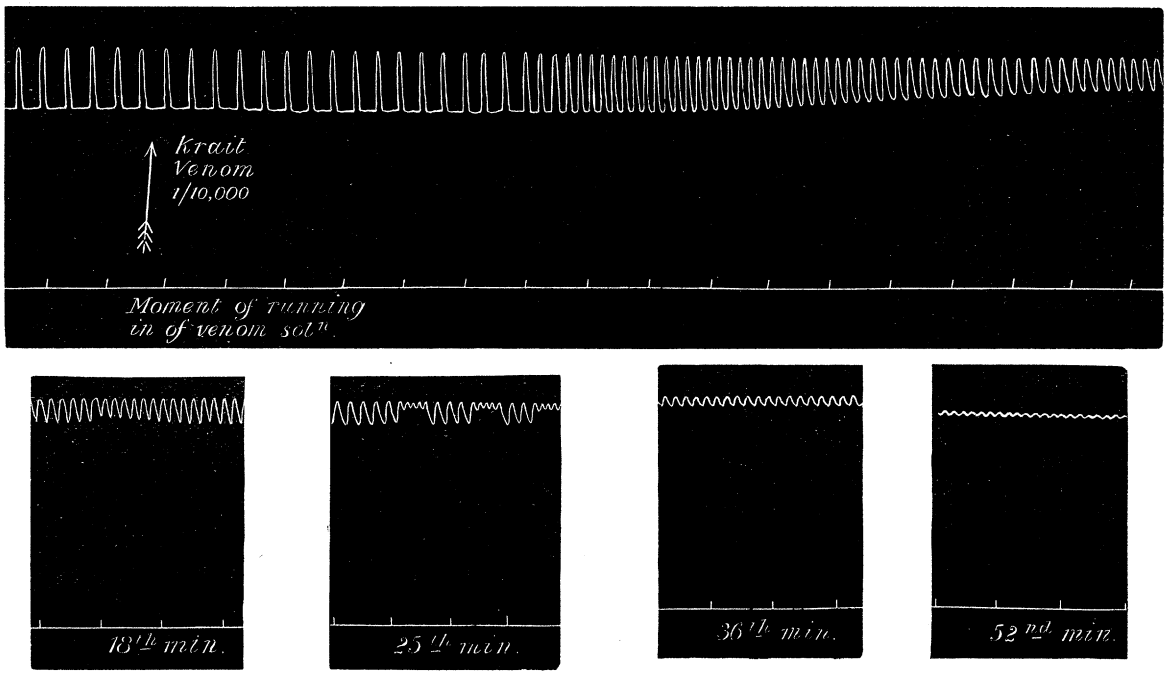
The brain and spinal cord of a frog having been carefully destroyed from half an hour to an hour before the beginning of the experiment, the thorax was opened and a cannula placed in one aorta (the other being ligatured). The venæ cavæ were cut and the cannula was attached by glass tubes to a series of MARRIOTTE'S flasks, by which the level of the fluids to be perfused was maintained at a constant height of 17 centims. The normal calibre of the vessels was ascertained by causing 0·65 per cent. sodium chloride solution to flow through them for from 10–20 minutes until the flow was uniform, the outflow being collected in graduated measures and the amount noted every 60 seconds. The venom dissolved in the same strength of saline solution was then perfused for from one hour to one hour and a-half. We had, unfortunately, too small a quantity of the venom to define the limits of its action with accuracy, but our experiments show that it produced an effect similar to that of Cobra venom but very much less in degree. In the strength of 1 : 500,000 we were unable to find any certain evidence of constriction, but in the strength of 1 : 250,000 the flow diminished to $\frac{1}{2}$ in 23 minutes and to $\frac{1}{4}$ in 69 minutes. There was no recovery when the normal saline was again perfused. With a strength of 1 : 100,000 the flow diminished to $\frac{1}{2}$ in 35 minutes, and with a strength of 1 : 50,000 the flow diminished to $\frac{1}{3}$ in 38 minutes.

Perfusion of the Frog-ventricle with Krait Venom.

This was carried out in a modification of SCHÄFER'S plethysmograph. Blood mixture was used for the normal, and as a vehicle for the venom. It consisted of 1 part of ox blood diluted with two parts of RUSCH'S modification of RINGER'S fluid. *Rana esculenta* was the animal used to provide the heart.

Experiment 1.—A frog-ventricle was perfused with a solution of Krait venom of a strength of 1 : 10,000. The following table gives the rate and the amplitude of beats in successive minutes :—

Minutes *before* venom solution was run in.	Number of beats per minute.	Amplitude of excursus of beat.	Minutes *after* venom solution was run in.	Number of beats per minute.	Amplitude of excursus of beat.
7	9·5	8·25	1	10	8
6	10·25	8·25	2	15	8
5	9·0	8·25	3	21·5	7·5
4	9·0	8·25	4	17·5	6·5
3	10·25	8·25	5	19·5	4·5
2	9·0	8·25	6	17·5	4·5
1	9·0	8·25	7	18	4·25
			20	18	3·25
			30	20	2·0
			40	25	1·0
			50	28·5	0·3



29.1.04. Perfusion of Frog-heart with Krait Venom Solution, 1 : 10,000. Read from left to right; systole upwards; time = 15 seconds. The minutes marked date from the running in of venom. The extracts are from a continuous tracing.

Experiment 2.—A frog-ventricle was perfused with a solution of Krait venom of a strength of 1 : 100,000. The tracing was continued for 52 minutes after the venom perfusion began. The result was entirely negative.

One of us* has shown that Cobra venom acts powerfully on the frog-ventricle, killing it in systole, when used in strong solutions, and acting as a tonic and stimulant

* Captain R. H Elliot, "A Contribution to the Pharmacology of Cobra Venom;" communicated to the Royal Society, January, 1904; abstract at 'Roy. Soc. Proc.,' vol. 73, p. 183.

when the strength of the venom was weaker. Thus a 1 : 100,000 solution of Cobra venom killed the frog-ventricle in 20 minutes, in a position of tight systole, whilst a 1 : 10,000,000 solution distinctly quickened the beat. We have here a similar though far weaker action.

It would have been desirable to test the action of stronger solutions of the venom. This we were prevented from doing by the smallness of our stock of venom.

Kymographic Experiments.

Experiment 1.—A rabbit, weighing 3·75 kilos., was etherised; blood pressure was taken by the carotid, and respiratory movement was registered by means of a double stethograph. 0·004 grm. of Krait venom was dissolved in 1 cub. centim. of RINGER'S fluid and injected into the left external jugular vein, the injection lasting 40 seconds. The blood pressure before the injection registered 126 millims. of mercury; 1 minute after injection it had fallen to 92; it then recovered steadily and stood at 116 millims. in the 5th minute after injection. Meanwhile respiration had been getting slower and shallower, and practically ceased in the 8th minute. An asphyxial rise of blood pressure then took place, a rise to 128 millims. being registered. A drop in pressure followed, the level reaching 82 millims. in the 10th minute. There was no evidence of cardiac inhibition on the blood-pressure tracing to explain the early fall, and it was only when asphyxiation was well marked that the heart-beat slowed. Under artificial respiration the blood pressure rose, and attained its former high level. When the artificial respiration was temporarily stopped, at intervals, a steep rise of pressure invariably occurred; a resumption of artificial respiration brought the pressure back to its former level again at once. Slowly and steadily the pressure fell, though the heart-beat remained comparatively rapid. After 1 hour 20 minutes of artificial respiration the experiment was stopped. In the meantime the stoppage at intervals of the artificial respiration had clearly shown that no respiratory effort whatever was being made by the animal. Whilst it was still under the anæsthetic the chest was opened and the phrenic, vagal, and sciatic nerves were tested with the secondary current. Neither the phrenic nor sciatic nerves responded to even maximal stimuli by so much as the production of a muscular twitch. Maximal stimulation of the vagi slowed, but did not stop the heart.

Since the M.L.D. of Krait venom for rabbits is 0·00008 grm. per kilo., this animal received over thirteen times the subcutaneous M.L.D. intravenously.

Experiment 2.—A dog, weighing 9 kilos., received an intravenous injection of 0·005 grm. of Krait venom dissolved in 5 cub. centims. of RINGER'S fluid. The tracings in order from above are: (1) Intestinal volume recorded by a plethysmograph (SCHÄFER'S); (2) blood pressure taken in the carotid; (3) respirations recorded by a double stethograph; (4) 10 seconds time-marks; (5) signal. The injection was made by the left external jugular, and took 72 seconds. There was an immediate fall of blood pressure, accompanied at first by a fall and later by a distinct rise in intestinal volume. There is no slowing of the beat, so we apparently have not got cardiac inhibition to deal with. The rise in intestinal volume will explain the fall in general blood pressure, if we may suppose that the first fall in intestinal volume (a matter of 20 seconds only) was due to a dilatation in another portion of the splanchnic area which had not yet reached the loop of gut under observation. Another possible explanation will be given presently. An extract from this portion of the tracing is here shown. The presence of Traube-Hering curves in the blood-pressure tracing supports the view that we have to do with a central vaso-motor dilatation. This dilatation continued throughout the experiment, and was increased when a second injection of 0·005 grm. of the venom was given 30 minutes after the first injection. In spite of this the general blood pressure remained high throughout the experiment. This is apparently to be explained

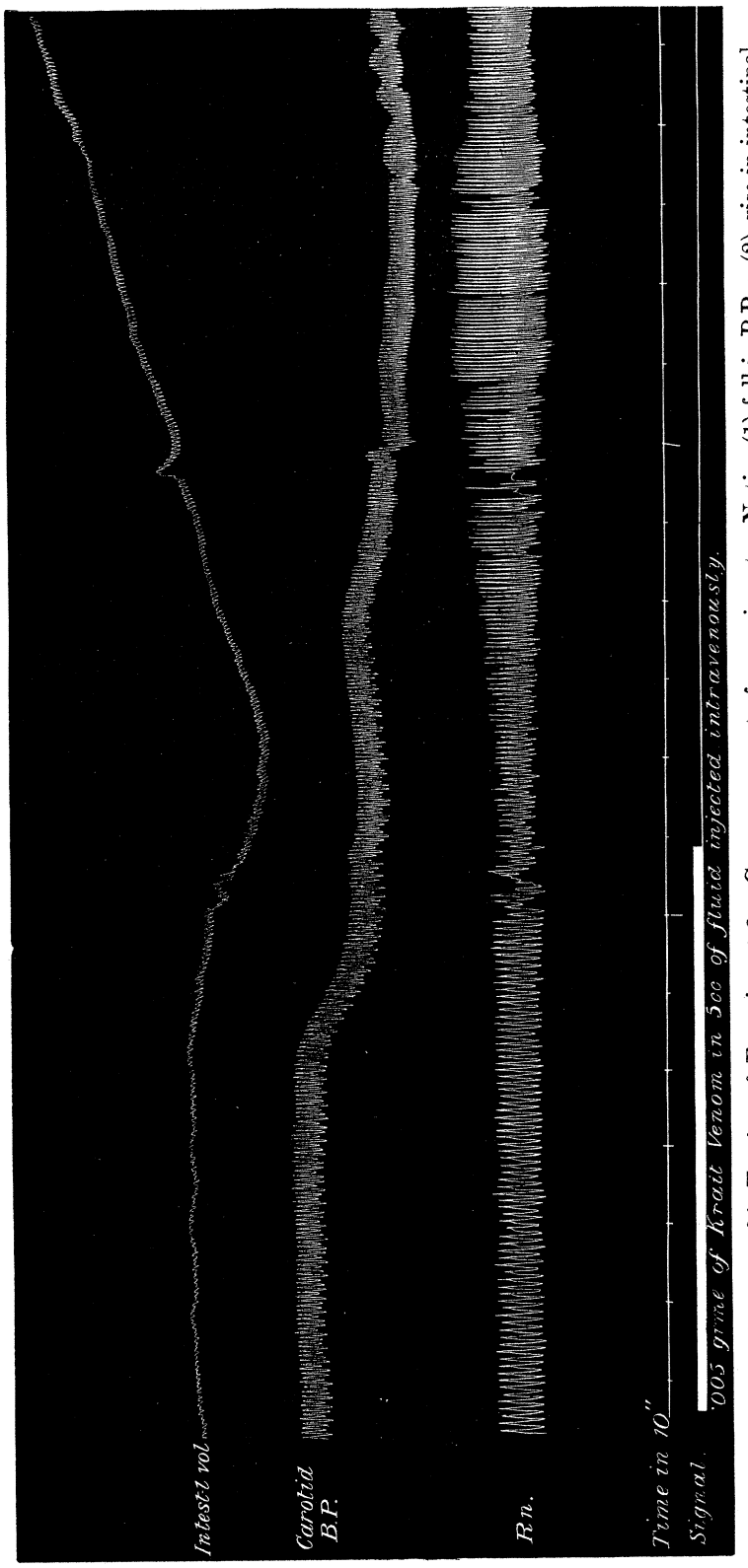
by the direct constriction of the systemic arterioles by the circulating venom. We have already dealt with this action.

Extracts from the tracing are given on pp. 337 and 338.

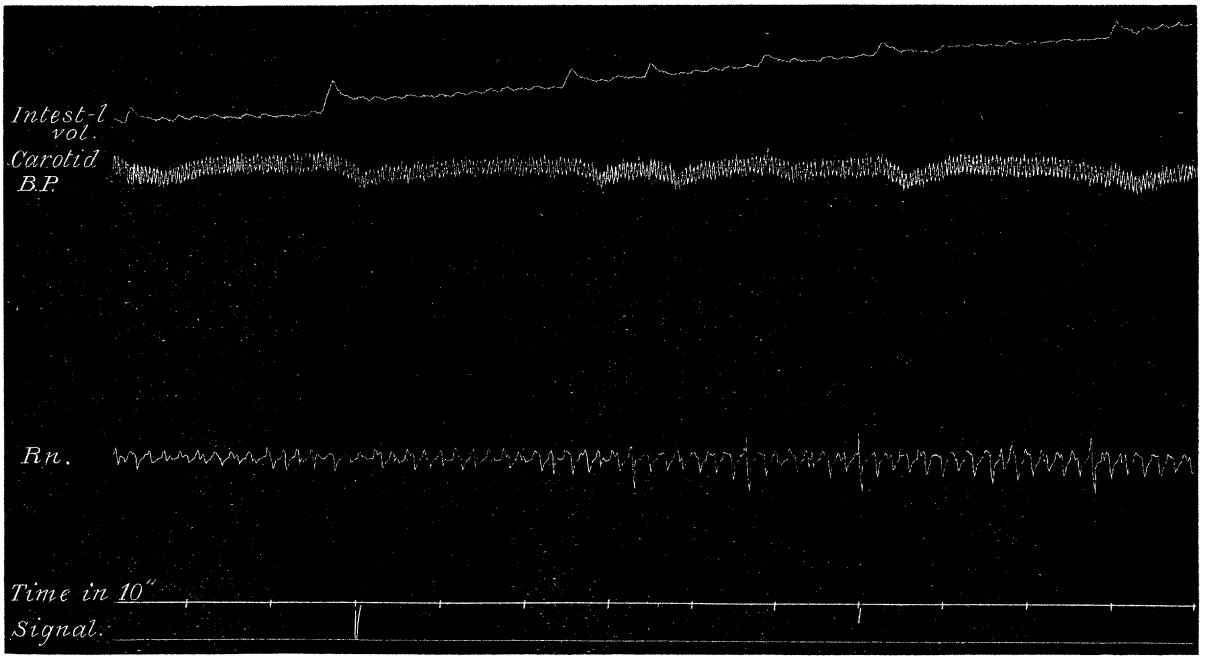
The accompanying table gives a short analysis of the tracing. Respiration was first excited and then gradually paralysed, just as occurs in Cobra poisoning. The experiment was stopped after 55 minutes, and the nerve-ends were tested by the secondary coil. The phrenics responded with the coil at 300 millims. from the primary coil, the sciatics at 280 millims., and vagal inhibition was got at 120 millims. The two first were, therefore, distinctly blunted in sensitivity; in the last this is doubtful:—

Time in minutes.	Number of heart-beats in 10 seconds.	Number of respirations in 10 seconds.	Amplitude of respiratory movement as shown by stethograph.	Blood pressure in millimetres of mercury.
			millims.	
1 before first injection .	18	11	10	140
1 after commencement of first injection . .	19	12	9	118
2 ditto	21	19	18	105
5 ditto*	16	11	6	136
10 ditto*	13	9	11	155
15 ditto*	14	11	4	164
20 ditto	20	10	21	162
25 ditto	17	9	9	168
30 ditto	17	9	3	151
A farther injection of 0·005 grm. of Krait venom was now given, in the same way as the last dose.				
31 ditto	17	8·5	3	148
32 ditto	18	9	4	144
35 ditto	19	7	2·5	167
40 ditto	19	7	1·75	162
45 ditto	19	6·75	3	164
50 ditto	20	6	2	174
55 ditto	20	5	3·5	172

* It is possible that the anæsthetic was to blame in these three readings.



Extract from Kymographic Tracing of Experiment 2. Commencement of experiment. Notice (1) fall in B.P.; (2) rise in intestinal volume; and (3) changes in respiration.

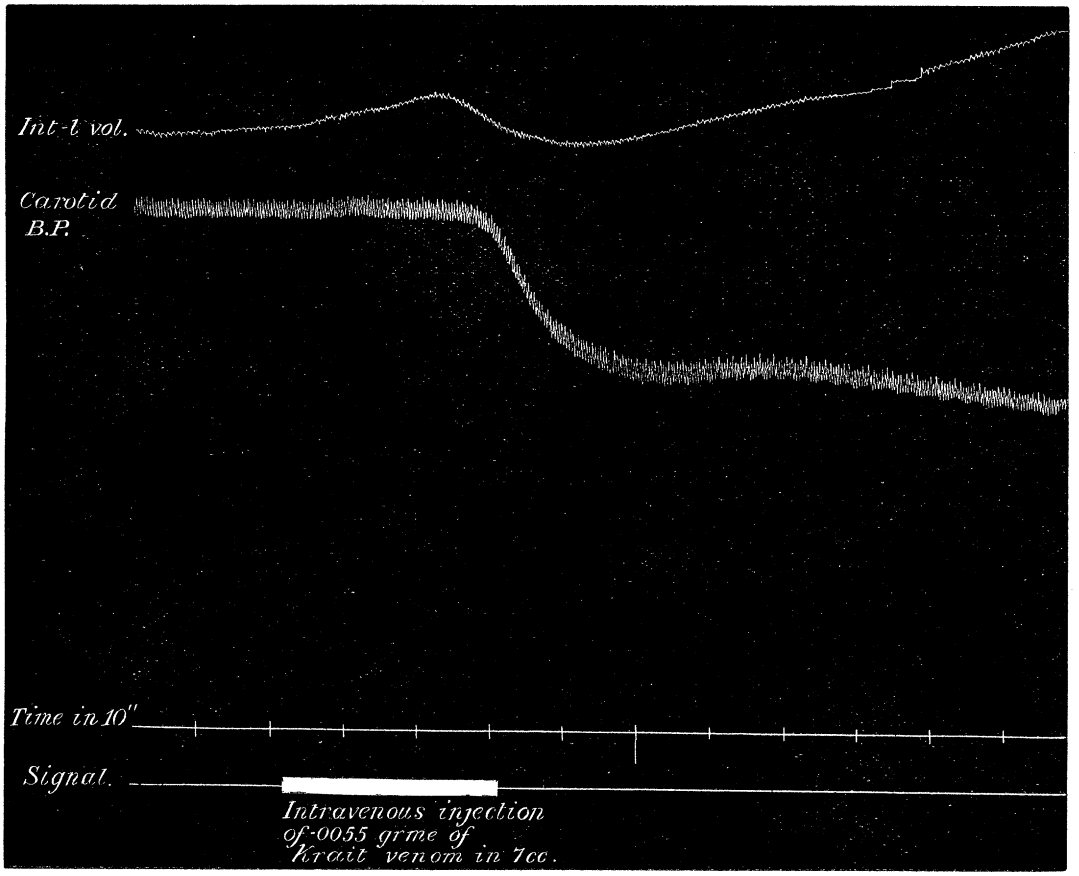


Extract from Kymographic Tracing of Experiment 2. The end of the tracing just before the experiment was stopped. Notice (1) the high B.P.; (2) the rising intestinal volume (the lever had been lowered several times); and (3) the irregular respiration.

Experiment 3.—The previous experiment was repeated on a dog weighing 5·5 kilos. The dose of venom injected was 0·0055 grm. (Krait venom), dissolved in 7 cub. centims. of RINGER'S fluid. There was a speedy fall of blood pressure. Slightly preceding this there was a rise in the plethysmographic lever denoting a rise in intestinal volume; then followed a passive fall of intestinal volume, and immediately afterwards a great rise thereof. This rise in intestinal volume continued during the 17 minutes the tracing was taken, but the blood pressure recovered in spite of it. The following table gives the results recorded:—

Time in minutes.	Number of heart-beats.	Blood pressure in millimetres of mercury.
Before injection . . .	26	128
1 after	26	86
2 „	26	79
3 „	27	73
4 „	27	75
5 „	27	76
10 „	25	94
15 „	23	108

An extract from the tracing is furnished on p. 339.



Extract from Kymographic Tracing of Experiment 3.

Experiment 4.—A rabbit, weighing 5 kilos., was etherised; blood pressure was taken in a carotid artery, respiratory movement was recorded by means of a double stethograph, and injections of Krait venom were made through the marginal vein of one ear.

The first injection consisted of 0·0006 grm. of Krait venom dissolved in 1 cub. centim. of normal saline solution, and took 1 minute to pass in. The second was given 22 minutes later, and consisted of 0·001 grm. of Krait venom in 1 cub. centim. of normal saline solution; it took 25 seconds to pass in.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

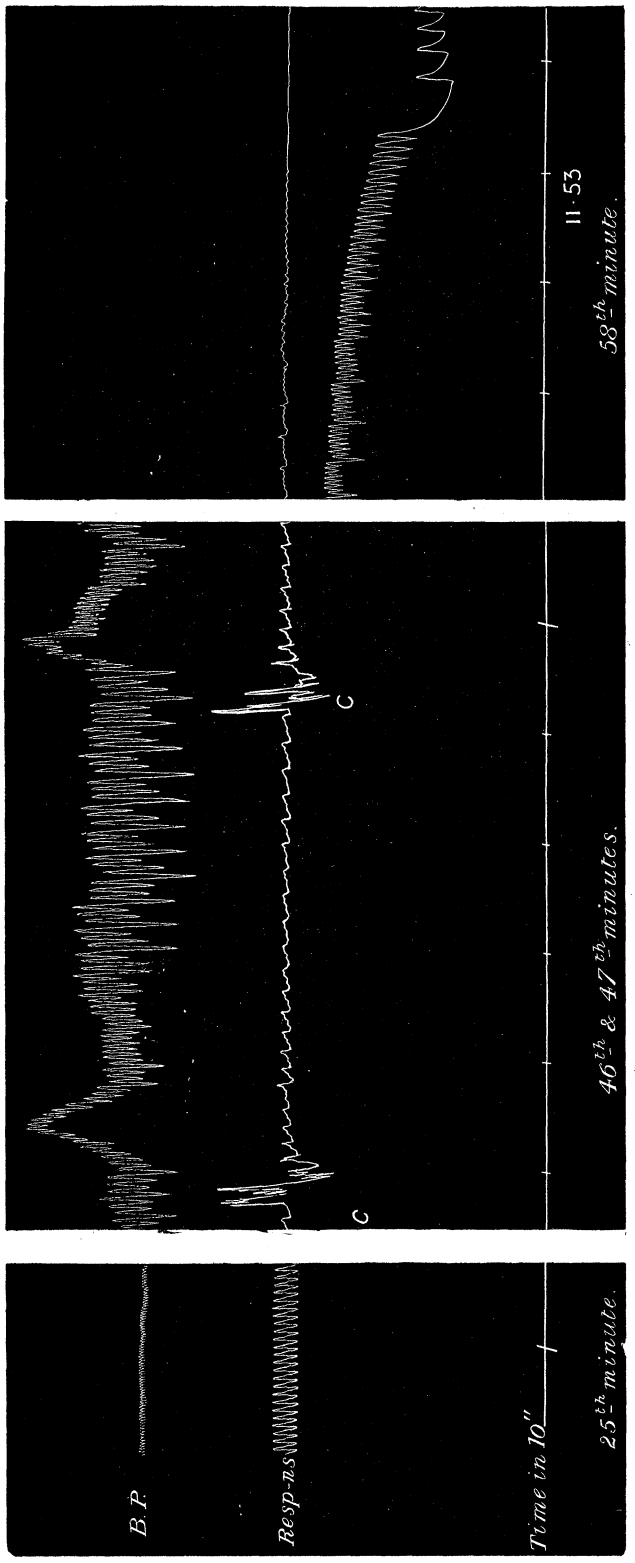
The following table shows the results obtained :—

Time in minutes.	Heart-beats in 10 seconds.	Respirations in 10 seconds.	Respiratory excursus.*	Blood pressure in millimetres of mercury.
			millims.	
Before injection	47	19	6	136
1 after ditto	43	14	5·5	132
2 ditto	43	14	5·25	129
3 ditto	44·5	13·5	5	126
4 ditto	44	14	5·5	124
5 ditto	44	14	5·5	122
6 ditto	42·5	15·5	5·25	124
10 ditto	42	17·5	5·5	126
15 ditto	41	17	4·5	121
20 ditto	39	15	4·75	119
22 ditto	40	16	5	121
The second injection was given just before the last reading was taken.				
23 after first injection	42	15	4·5	123
24 ditto	42·5	14·5	4·5	115
25 ditto	42	13·5	4	112
26 ditto	42	13	4·25	112
30 ditto	40	12	4·75	111
35 ditto	38	10·5	5	113
40 ditto	35	10	4·75	112
45 ditto	17	6·5	2	114
50 ditto	13	3	1	124
55 ditto	29	4·5	2	65
58 ditto	18	0	0	38
60 ditto	Dead	—	—	—

Experiment 5.—A rabbit, weighing 3·5 kilos., was prepared in all respects like that in the previous experiment, and received 0·002 grm. of Krait venom, dissolved in 1 cub. centim. of RINGER'S fluid, injected into the marginal vein of one ear; the injection took 40 seconds. The result is shown in the following table :—

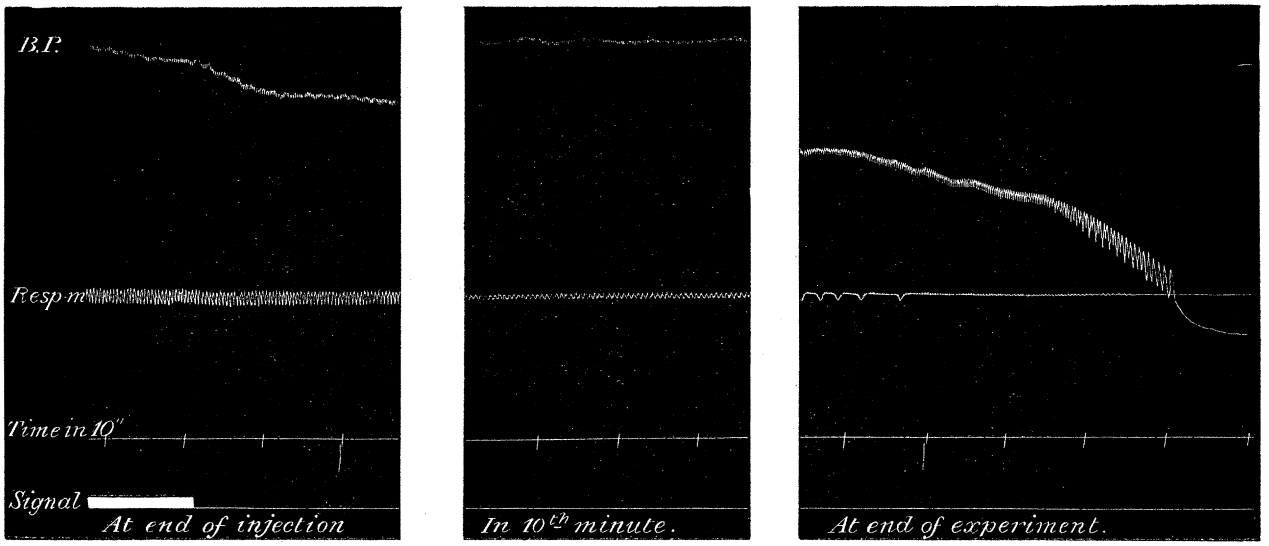
Time in minutes.	Heart-beats in 10 seconds.	Respirations in 10 seconds.	Respiratory excursus.*	Blood pressure in millimetres of mercury.
			millims.	
Before injection	53·5	24	2	80
1 after ditto	54	23	2	70
2 ditto	54	22	2	64
5 ditto	52	21	1·5	70
10 ditto	51	16	1	86
15 ditto	49	15·5	0·5	80
16 ditto	41	9	0·3	64
17 ditto	42	4·5	1	50
18 ditto	Dead	—	—	—

* *Note.*—The respiratory excursus given in Experiment 4 and 5 is the excursus in millimetres of the lever point of the double stethograph.



Extracts from Kymographic Tracing of Experiment 4.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES



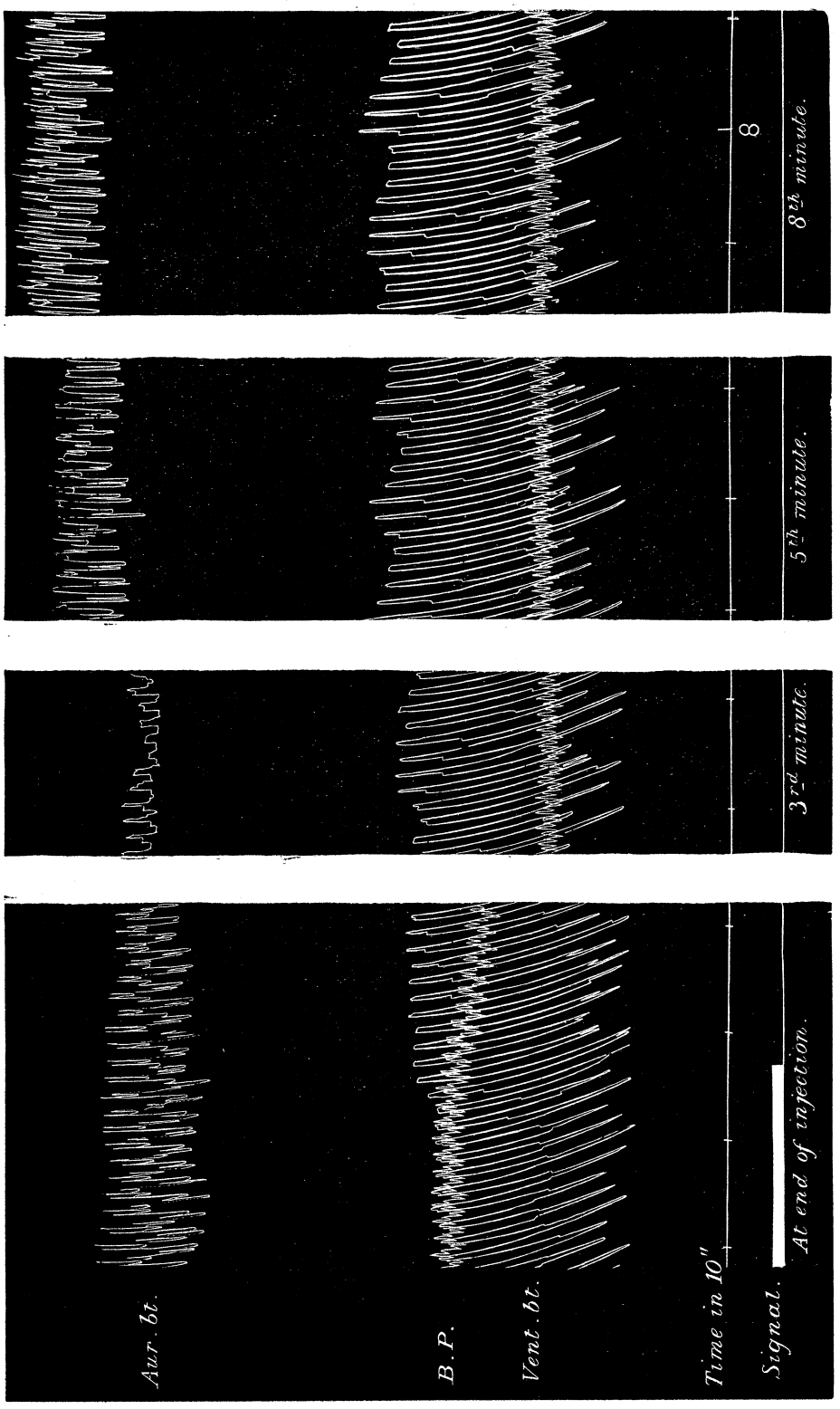
Extracts from Kymographic Tracing of Experiment 5.

Experiment 6.—A cat, weighing 2·2 kilos., was etherised; blood pressure was taken in a carotid artery; the chest was opened and the movements of the auricle and ventricle were recorded by means of levers attached to their walls by silk threads; artificial respiration was maintained throughout; the injection was made through an external jugular vein, time was registered in 10 seconds.

After obtaining a normal, 0·002 grm. of Krait venom was injected, in 1 cub. centim. of RINGER'S fluid; the injection took 18 seconds. There was an immediate fall of blood pressure, and at the same time there was clear evidence of a weakening of the auricular beat, accompanied by some slowing of the heart. The ventricle did not appear to be affected. The force of the auricular beat recovered rapidly, was normal in the fifth minute, and during the next 4 minutes was decidedly in excess of what it had been before the injection. Extracts from the tracing are given; the following table shows the changes that occurred in heart-rate and blood pressure:—

Time in minutes.	Number of heart-beats.	Blood pressure in millimetres of mercury.
Before injection	27	121
1 after ditto	26	106
2 ditto	22	87
3 ditto	20	86
4 ditto	21	88
5 ditto	21	88
6 ditto	21	89
7 ditto	20	90
8 ditto	20	90
9 ditto	20	90

Experiment 7.—The previous experiment was repeated on a cat weighing 2 kilos., a dose of 0·003 grm. of Krait venom being injected. The results yielded bore out those of the previous experiment, and need not therefore be given in detail.



Extracts from Kymographic Tracing of Experiment 6.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Summary of Conclusions.

We are now in a position to consider the very diverse influences which act upon the blood pressure in Krait poisoning, and we find a very different state of affairs from that which holds in Cobraism.

(1) The heart-muscle, or its nerve-end mechanism, is acted on directly by Krait venom. This action is strictly comparable to that of Cobra venom, but is much less strong. The tracings, taken with the mammalian heart exposed and with levers attached to the auricular and ventricular walls, show, however, that an important increase of cardiac force is in this way brought about. A decided increase of heart rate was also observed in several tracings.

(2) There are indications of a feeble cardio-inhibitory action. This supervenes so rapidly, when it is seen at all, that it must almost certainly be of central origin. The action of this venom is thus again brought into line with that of Cobra poison, and is much more feeble than that of the latter in this particular phase of its activity. So much so indeed, that the inibition is quickly and easily masked by the direct effect of the venom on the heart, the latter action is manifested more slowly. It is probable that inhibition of the heart is responsible for the early fall in blood pressure, which is sometimes noticed before the plethysmographic lever has recorded any change in intestinal volume. The slight extent of the inhibition, even when it is observed, may be judged from the fact that it is only the auricle which shows the change, the ventricle curve not being perceptibly affected in our tracings. Moreover slowing of the heart was never well marked and was often absent.

(3) The direct action of the venom on the arterial muscle, or on its nerve-end mechanism is, as we have shown above, such as to produce vaso-motor constriction. This effect can be clearly got with solutions of 1 : 250,000 and below, but we failed to get it at 1 : 500,000. It is therefore much weaker than the corresponding action of Cobra venom, which can be demonstrated in a solution of 1 : 10,000,000.

(4) The injection of Krait venom is followed by a very early and decided fall of blood pressure. This fall has been shown to be mainly due to a dilatation of the vessels of the splanchnic area, and is almost certainly central in origin.

It is obvious that the direct tonic effect of Krait venom on the heart and vessels tends to raise the general blood pressure. On the other hand, the slight cardio-inhibition and the powerful action on the central vaso-motor (leading to dilatation of the splanchnic area vessels), are responsible for the marked early fall so constantly met with. The two latter forces come into action before their opponents have time to manifest themselves.

There is yet another factor to be taken into account, viz., the influence of the poison on the respiratory mechanism. We regard this as the most important element in the whole case.

Our experiments have shown that death in Krait poisoning is due to failure of the respiratory mechanism and, moreover, that when this failure occurs, the phrenic and other motor nerve-ends are still active, though their sensitivity is always more or less impaired, and often very decidedly so. We, therefore, conclude that the interference with respiration, which culminates in the death of the subject, is due to a direct action of the venom on the respiratory centre. This view is borne out by the rapidity with which respiration is often affected in these experiments. The limited quantity of poison at our disposal prevented us from pushing this question farther, as we would like to have done. We hope at a later date to apply Krait venom directly to the exposed medulla oblongata with a view to ascertain whether the centre can be directly poisoned in this way without the nerve-ends being at all affected. One of us has shown that such is the case with Cobra venom. We are, however, distinctly of the opinion that the blunting of nerve-end sensibility, which we have observed, is sufficient to add materially to the embarrassment of an already damaged respiratory centre.

Respiratory failure, slowly developed, brings in its train all the phenomena of slow asphyxiation. The circulation of venous blood through the medulla oblongata tends to slow the heart by acting on the vagal cardio-inhibitory centre. It must also stimulate the vaso-motor centre, and so tend to cause arteriolar constriction, and a consequent rise in blood pressure. These two latter forces will, of course, act in opposition to each other, whilst both are superadded to those which we have already discussed.

It is, therefore, very evident that the problems which lie hidden in a blood-pressure tracing taken from a Krait poisoned animal are very complex. The main features are, however, obvious enough. The early fall of pressure is due to a central vaso-moter action on the splanchnic area, assisted and sometimes probably preceded by a small measure of cardio-inhibition of central origin. The recovery of pressure which follows is attributable (1) to an increase in cardiac force brought about by the direct action of the venom on the heart, and (2) to the direct constrictive action of the venom on the vessel walls. Later, as respiration progressively fails, the asphyxiation of the medulla oblongata is thrown into the scale in favour of vaso-constriction. Nevertheless, the direct action of the venom on the vaso-motor centre is so powerful that one never sees the sustained high levels and the final great rises of blood pressure which are a common feature of a tracing in Cobra poison experiments.

Most of the kymographic work of this paper has been carried out in the Physiological Laboratory of Edinburgh University. For the permission to use his apparatus, and for his valuable advice and assistance, we are deeply indebted to Professor E. A. Schäfer.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES