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THE ANTIDIURETIC RESPONSE TO AND EXCRETION OF PITUITARY (POSTERIOR LOBE) EXTRACT IN MAN, WITH REFERENCE TO THE ACTION OF NICOTINE

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The work described in this paper is a continuation of observations already made in this laboratory on the antidiuretic action of nicotine (Burn, Truelove, and Burn, 1945; Walker, 1948). Taylor and Walker (1951) have recently shown that the action is followed by the appearance of an antidiuretic substance in the urine of man resembling in its properties the posterior lobe hormone and not nicotine.

If a man drinks water, the onset of diuresis indicates the arrest of the normal secretion of antidiuretic hormone. When posterior lobe extract is injected intravenously at this point, inhibition of the diuresis occurs and lasts for a time depending on the amount injected. We have determined the dose-response relationship in three subjects. We have also estimated the proportion of antidiuretic hormone excreted in the urine after the intravenous injection of posterior lobe extract in varying amounts. A few experiments have been done in which the antidiuretic response to nicotine (either inhaled in tobacco smoke or injected) has been determined, and the amount of antidiuretic hormone excreted has been measured.

METHODS

The observations were made on three male subjects in the afternoon. Each of them emptied his bladder every 15 minutes and recorded the volume. In certain experiments in which the antidiuretic effect was expected to be small, this period was reduced to 5 minutes. At the beginning of an experiment, the subject was required to show that he was not already very hydrated by producing two volumes, each of less than 25 ml. Then he drank one litre of water in 1–2 minutes. About 45 minutes later, when the volume of urine produced in the previous 15 minutes was more than 70 ml., the injection was given, or, in some experiments, the smoking was begun. The diuresis was then usually inhibited for a varying time, after which it increased to reach a second peak. The urine output was recorded until this peak was passed.

The pituitary (posterior lobe) extract (PLE) was prepared by British Drug Houses, Ltd. Nicotine was injected as a solution of nicotine hydrogen tartrate in saline. This salt contains one-third by weight of nicotine base. All injections were made intravenously. The doses were usually contained in 0.5 ml., except the large doses of PLE and nicotine, which were in 1 ml.

The method of extraction of antidiuretic hormone from urine was that described by Noble, Rinderknecht, and Williams (1939). Our practice was to reduce the volume of solvent as much as possible, using each portion twice if need be, so as to shorten the distillation.

TABLE I

TIMES TO 50 PER CENT EXCRETION DETERMINED IN CROSS-OVER TEST IN 16 RATS

Exp.	Standard (min.)	Test (min.)	Percentage recovered
1	169	160	78
2	133	128	87
3	156	147	77

The urine passed before injection, and that passed after, were extracted separately, giving two solutions, a blank and a test. A suitable amount of PLE was added to the blank solution to make it comparable with that present in the test solution. The blank plus the known amount of PLE then became the Standard, and these two solutions were assayed by the method of Burn (1931). All results were obtained by carrying out the full cross-over test in 16 rats.

RESULTS

Efficiency of extraction.—Three experiments were performed to test the method of urine extraction. On each occasion normal urine was obtained and divided into two parts. To one part PLE was added in the proportion of 100 milliunits (mU.) to 100 ml., and both parts were then extracted. The first extract was the test solu-

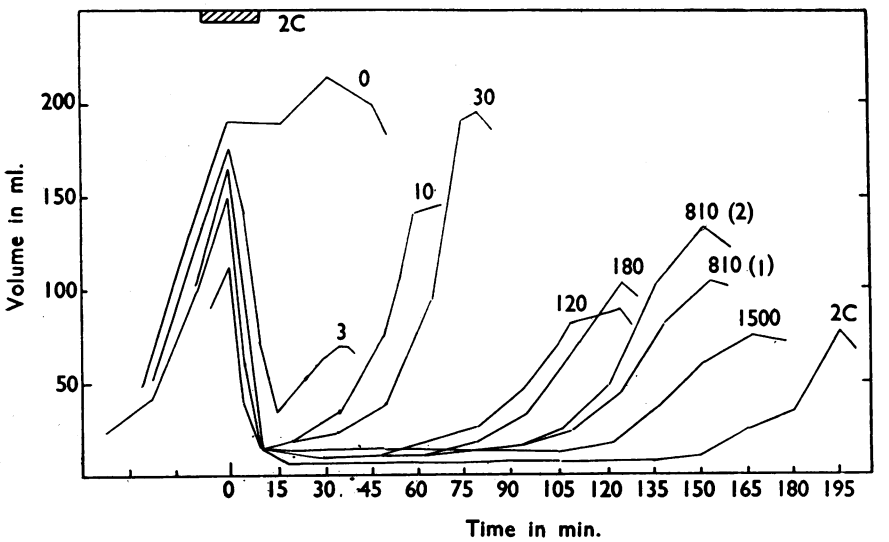

FIG. 1.—Relation between the dose of PLE and the urinary excretion. Ordinates: volume of urine in ml. per 15 min. period. Abscissae: Time in min. An intravenous injection of the dose, in mU. PLE, stated on each curve was made at zero time. The curve showing the excretion after the smoking of two cigarettes (2C) has been adjusted so that the onset of inhibition coincides with that for the other curves.

TABLE II

RELATION OF DOSE OF PLE TO PERIOD OF ANTIDIURETIC ACTION

Dose (mU.)	Time (min.) from injection to peak of diuresis		
	A	B	C
3	35	—	—
10	70	68	67
30	82	103	81
120	128	113	98
180	123	123	118
180	—	—	113
270	113	125	110
405+405*	155	—	123
810	137	158	158
810	153	—	138
1,500	166	—	150

* These doses were given at an interval of 15 minutes.

tion. A Standard solution was prepared by adding 100 mU. PLE to an amount of second extract corresponding to 100 ml. of urine. Thus any substances in the urine extract which might augment the effect of the antidiuretic hormone were equally present in both solutions. The results are given in Table I.

The doses of the Test solution given were chosen on the assumption that 100 per cent recovery would be obtained. Table I shows that this introduced no serious error. The mean percentage recovery was 81, a result which is in accord with those of Noble *et al.* (1939), who recovered 70–90 per cent of the original dose.

Relation of dose of PLE to inhibition.—We next observed the inhibition produced by injecting PLE. A series of results in one subject (A) is shown in Fig. 1 in which the curves of diuresis have been superimposed so that the time of injection is taken as zero in each experiment. For each dose the time elapsing between the injection and the peak of diuresis after the inhibition was determined. The results

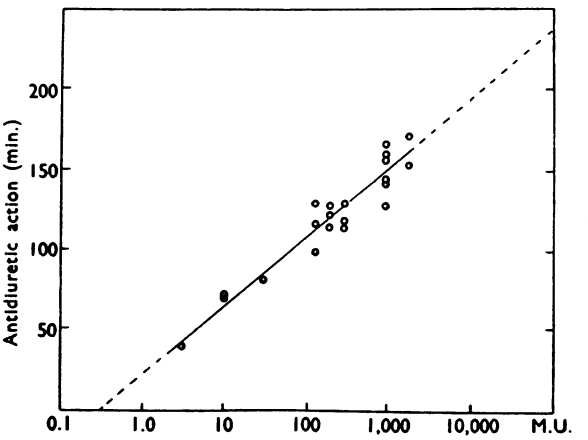

FIG. 2.—Relation between the dose of PLE and the time elapsing between the injection of the dose and the peak of diuresis which terminates the inhibition. A regression line has been drawn through the experimental points. Ordinates: Time in min. Abscissae: Dose in mU. PLE on a logarithmic scale.

are shown in Table II. The periods for all three subjects are plotted against the dose in milliunits on a logarithmic scale in Fig. 2. A regression line has been drawn through these points, and the equation of the line is

$$\text{Time in minutes} = 23.0 + 42.2 \log \text{dose}$$

where the zero of log dose is taken at 1 mU. For comparison with later theory, it is convenient to write this equation as

$$\text{Dose} = 0.285e^{0.0545t} \text{ milliunits} \quad \ldots\ldots\ldots\ldots\ldots (1)$$

an equation which relates the dose injected and the consequent average time to the peak.

Proportion of antidiuretic hormone excreted.—The samples of urine obtained in the above experiments were extracted, and were then examined by the rat method. The urine extract obtained after the injection until the time when inhibition was disappearing was the Test solution, and the Standard solution was prepared by adding a suitable amount of PLE to the extract of urine collected before the injection. It was found that, when the dose of PLE injected into the human subject

TABLE III

ANTIDIURETIC HORMONE IN URINE AFTER INTRAVENOUS INJECTION WITH PLE DURING WATER DIURESIS

Dose PLE injected per subject (mU.)	No. of subjects	Quantity recovered (mU.)	Percentage recovery
120	3	0	0
180	3	30	5.5
270	3	21	2.6
810*	2	60	3.7
810	3	180	7.4
810	2	120	7.4
1,500	2	95	3.1
			Mean 5.0

* In this experiment the dose was given in two equal parts at an interval of 15 minutes.

was 120 mU. or less, insufficient hormone appeared in the urine for it to be detected. The results are given in Table III, in which it can be seen that the mean recovery was 5 per cent, and the individual figures varied from 2.6 to 7.4 per cent.

Action of inhaled nicotine.—Subjects A and B smoked cigarettes on two occasions, and subjects C and D were given injections of nicotine acid tartrate, C on three occasions and D on two occasions. The duration of inhibition of diuresis was recorded and the amount of antidiuretic hormone excreted in the urine was estimated. The results are shown in Table IV. When A and B smoked cigarettes their urines were combined for the estimation of antidiuretic hormone on each occasion. In Exp. 4 of Table IV the inhibition in subject A lasted for 195 minutes. Reference to Table II shows that an inhibition of this duration would be caused by the injection of a quantity greater than 1,500 mU. PLE, which was the largest amount actually injected. In Exp. 5 of Table IV the inhibition in subject B lasted for 105 minutes, which would be caused by the injection of only 85 mU. In the

combined urines of A and B, the antidiuretic hormone found present must have come almost entirely from subject A. The amount found was 95 mU., and, on the assumption that this was 5 per cent of the amount liberated in the blood, the amount liberated in the blood of subject A was 1,900 mU.

TABLE IV

ACTION OF NICOTINE

Exp.	Subject	Nicotine	Inhibition (min.)	Antidiuretic hormone excreted (mU.)
4	A	2 cigarettes	195	95
5	B	2 cigarettes	105	
6	A	ł cigarette	105	None
7	B	2 cigarettes	105	
8	C	0.8 mg. base i.v.	180	75
9	C	0.8 mg. base i.v.	60	None
10	C	0.8 mg. base i.v.	105	Not determined
11	D	1.1 mg. base i.v.	60	Not determined
12	D	2.7 mg. base i.v.	75	Not determined

In Exps. 6 and 7 of Table IV, the inhibition in each subject was equal to that which would be caused by 85 mU., so the total liberated in the blood of both would be 170 mU. If 5 per cent of this was excreted, the total excreted in the two urines would be 8.5 mU., which is a quantity too small to be estimated by the cross test, and in fact no antidiuretic substance was found.

Action of injected nicotine.—The results of injecting 0.8 mg. nicotine (base) into subject C in Exp. 8 of Table IV differed greatly from the result in Exps. 9 and 10, when the same injection was given. In Exp. 8 the period of inhibition was 180 minutes, corresponding to the release of more than 1,500 mU. In the urine 75 mU. was found, corresponding to a release of 1,500 mU. in the blood.

In Exps. 9 and 10 the injection of the same amount of nicotine both times produced a much smaller effect equivalent to that produced by the injection of 7 and 85 mU. respectively. The most likely explanation of this difference between the result of Exp. 8 and those of Exps. 9 and 10 seems to be that subject C was an apprehensive person. During Exp. 8 he experienced unpleasant effects of nausea and sweating, and when he learnt that the injection was to be repeated on the day of Exp. 9 and again in Exp. 10 he was alarmed. O'Connor and Verney (1945) observed that the antidiuretic response to an irritating faradic stimulus was irregular and that the irregularity depended on the presence of the sympathetic chain ; further, they found that the antidiuretic effect of such a stimulus was abolished by the intravenous injection of adrenaline given shortly before the stimulus was applied. Kelsall (1949) obtained a similar result. It may therefore be that the reduced effect of nicotine in subject C in Exps. 9 and 10 was due to inhibition of the action of nicotine by fear.

There is, however, a great variation in the effect of nicotine in different persons ; this is illustrated by the small effect of 1.1 mg. and 2.7 mg. in subject D.

Effect of smoking.—The observations made in our experiments, together with those previously recorded by Burn *et al.* (1945) and by Walker (1948), make it possible to describe the quantity of antidiuretic hormone which is released by smoking. The observations are set out in Table V, and the figures in the last column fall into two groups, namely, those in which the equivalent amount of PLE is greater than 1,000 mU., and those in which the amount ranges from 3 to 190 mU. In the experi-

TABLE V

AMOUNTS OF ANTIDIURETIC HORMONE RELEASED BY SMOKING

Authors	Subject	No. of cigarettes	Inhibition (min.)	Equivalent amount PLE (mU.)
Burn *et al.* (1945) ..	L.H.T.	3	210	>1,000
	L.H.T.	3	120	190
	E.M.V.W.	3	210	>1,000
	E.M.V.W.	1	105	90
	J.H.B.	1	195	>1,000
	T.H.C.L.	2	180	>1,000
	J.R.W.G.	2	195	>1,000
Walker (1948)	A (N)*	1	165	>1,000
	B (N)	1	165	>1,000
	C (N)	1	120	190
	D (N)	1	105	90
	E (S)	1	75	18
	F (S)	1	60	7
	G (S)	1	60	7
	H (S)	2	180	>1,000
	J (S)	2	120	190
	K (N)	2	90	40
	L (S)	2	90	40
	M (S)	2	60	7
	N (S)	2	45	3
This paper	A	2	195	>1,000
	A	1	105	85
	B	2	105	85
	B	2	105	85

* S = smoker, N = non-smoker.

ments of the first group the subjects experienced nausea, sweating, sometimes diarrhoea, and occasionally vomiting. Sufficient nicotine was absorbed to produce toxic effects not seen when cigarettes are ordinarily smoked. In the experiments of the second group these toxic effects were inappreciable, and it is fair to take the figures of 3 to 190 mU. as indicating the amounts of antidiuretic hormone which are usually released.

The amounts vary greatly in the same individual at different times ; thus L. H. T., the first subject in Table V, smoked three cigarettes on two occasions ; on the first occasion at a time when he was not smoking, because he was in training, the amount released was more than 1,000 mU. ; on the second occasion, at a time when he was smoking about 15 cigarettes a day, the amount released was 190 mU.

So far as it is justifiable to speak of an average amount released, the mean figure is 75–100 mU. for one or two cigarettes.

Discussion

The rate of destruction of antidiuretic hormone.—During the time that elapses between the injection of PLE and the subsequent peak of diuresis which marks the end of the antidiuretic effect, the hormone is destroyed. The equation of the regression line (1) shows that the logarithm of the fraction of the injected dose that is left at any time is proportional to the time that has elapsed. A simple explanation of this is that the rate of destruction of antidiuretic hormone (ADH) is proportional to the concentration. If this is true, then, when the urinary flow increases, it should do so at an exponential rate.

Now, in the experiments described, it is seen that even with the largest doses of PLE, or the most prolonged inhibitions with smoking or nicotine, the urinary output never fell to zero. There was always some excretion, amounting to about 10 ml. per 15-minute period. It seems that this minimal excretion is not under the control of ADH, and, in order to find that part which is so controlled, this residual excretion must be subtracted from all the observed readings. This has been done in Fig. 3, in which zero time is the time of injection, and the volumes plotted refer to the urinary output in the 15-minute periods, 10 ml. having been subtracted from each

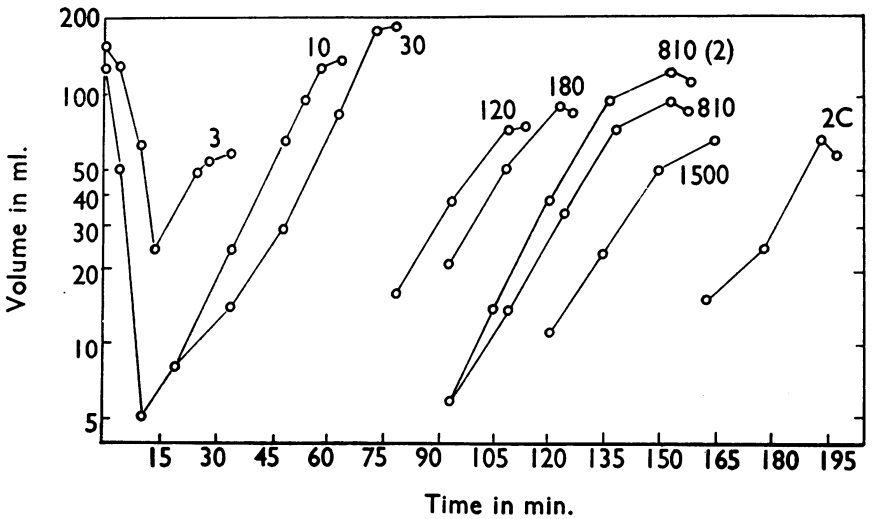

FIG. 3.—Rate of increasing urinary excretion. Ordinates: Urinary output in ml. per 15 min. 10 ml. has been subtracted from the output actually measured, and the remainder has been recorded. Abscissae: Time in min. The figures on the curves relate them to the dose in mU. PLE or to the smoking of two cigarettes.

figure recorded. In some experiments only two points are available, but where more points were obtained the resemblance to an exponential curve is unmistakable. Naturally this rate of increase of urine flow can only be maintained up to the maximum at which the kidney can work; also, it will only continue as long as the subject is hydrated; when the surplus water has been lost, the subject's own pituitary will liberate ADH and stop the diuresis. So the curves are rounded off at the top, and the observed peak of diuresis may occur at any point in its course.

If destruction of ADH proceeds at the same rate as the urinary flow increases, then the fraction of any injected dose that is left, at a time "t" is given by the expression

$$y_t = y_o e^{-kt} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (2)$$

where "y" is the fraction of the dose at the relevant time ($y_0 = 1$); and k is equal to the slope of the curves in Fig. 3. Actually these slopes are not all the same; k varies from 0.071 for the small doses to 0.048 for the largest dose.

The rate of inhibition of diuresis.—It has been noticed for some time that a finite period must elapse before the maximum degree of inhibition is obtained. Indeed, Kelsall (1949) used this phenomenon to compare the effect of injections of PLE with the effect of pain.

The length of the period given by Kelsall was an average figure of 14 minutes with limits of 11 and 23 minutes. The longest period that we observed was 14 minutes for the dose of 3 mU., and always shorter periods for larger doses. A dose of 120 mU., for instance, appears to have an immediate effect. A similar result is described by Verney (1947) in his Croonian lecture. In Fig. 20*b*, page 64 in his paper, there are three assay curves on the dog "Nicky." The time from injection to maximum inhibition is there seen to be 10 minutes for 1 mU., about 8 minutes for 2 mU., and 5–6 minutes for 3 mU.

It would be expected that the blood concentration would rise to a maximum very shortly after the injection of a dose of PLE. The circulation time in man is in the region of 10–30 seconds and mixing of the dose with all the blood should be obtained in about two minutes. Before this time, some blood will contain a higher concentration than the average, so the kidney ought to inhibit diuresis very quickly. A mechanism is therefore needed which will provide a delay of some 10 minutes before maximum inhibition is obtained.

Theory.—If we suppose that the ADH which inhibits diuresis is present in the kidney, and not in the blood, then, as the dose of PLE is injected into the blood, it must first be transferred into the kidney.

At a time $t = 0$, a dose x is given. Let X be the quantity remaining in the blood after time t, and suppose that in a short time dt a quantity $\alpha X dt$ transferred from the blood to the kidneys.

Then $dX = -\alpha X dt$.

Suppose further that the hormone present in the kidneys is destroyed at a rate proportional to Z, the quantity actually present in the kidneys; this rate is $\beta Z dt$ in time dt.

Then $dZ = \alpha X dt - \beta Z dt$

Hence $\dfrac{dX}{dt} = -\alpha X$ and $\dfrac{dZ}{dt} = \alpha X - \beta Z$

with conditions that at $t = 0$, $X = x$, $Z = 0$.

The solutions are

$$X = xe^{-\alpha t}$$

$$Z = \frac{\alpha x}{\alpha - \beta}(e^{-\beta t} - e^{-\alpha t}) \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (3)$$

It is reasonable to suppose that there is some quantity Z_0, such that excretion is inhibited (partially or wholly) as long as $Z > Z_0$. As t increases from zero to moderate values and to infinity, Z increases from zero to a maximum and then decreases again to zero. If the maximum exceeds Z_0, there will be inhibition. In general, there will be two times, t_1 and t_2, at which $Z = Z_0$, and inhibition will continue in the interval between them. Now t_1 and t_2 are the roots of the equation

$$Z_0 = \frac{\alpha x}{\alpha - \beta}(e^{-\beta t} - e^{-\alpha t})$$

or

$$x = \frac{(\alpha - \beta)Z_0}{\alpha} \cdot \frac{e^{\beta t}}{1 - e^{-(\alpha - \beta)t}} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (4)$$

In fact, t_2, the time to the peak, is the largest root of this equation. For "large" x, t will be large and $e^{-(\alpha-\beta)t}$ small relative to unity. If this term is ignored, then approximately

$$x = \frac{(\alpha - \beta)Z_0}{\alpha}\beta t_2 \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (5)$$

The regression equation (1) is presumably an approximation to this. It suggests that $\beta = 0.0545$. Actually β might be a little larger to allow for the neglected term, but even if β were 0.06 (and $\alpha = 0.2$), $1 - e^{-0.14t}$ would be so near to unity after 50 minutes that it could be safely ignored.

The considerations leading to $\alpha = 0.2$ are as follows: the blood flow through the kidneys is 1.2 l. per minute. As the blood volume is 6 l., a fifth of the blood volume passes through the kidneys per minute. It is supposed that the kidneys extract all the hormone from the blood that passes through them, and that this cleared blood immediately dilutes the remainder. This will not be exactly true in practice, as a finite time must elapse before mixing is obtained and before the cleared portion can again enter the kidney. This will result in an increase of the true value of α.

Let $\alpha = 0.2$, $\beta = 0.055$. Then equation (5) also suggests that

$$\frac{0.2 - 0.055}{0.2} \times Z_0 = 0.285$$

or $Z_0 = 0.4$ mU.

Fig. 4 shows

$$\frac{Z}{x} = \frac{0.2}{0.2 - 0.055}(e^{-0.055t} - e^{-0.2t}) \quad \text{(from equation 3)}$$

that is, the proportion of the applied dose in the kidneys, undestroyed, at time t. The proportion reaches a maximum of about 0.61 at $t = 9$ minutes, and thereafter decays to zero. After $t = 20$, the decay is almost entirely exponential. The smallest dose that produces any inhibition is x_0, such that $0.61x_0 = Z_0$, or $x_0 = 0.66$ mU.

It is now possible to see what happens when a small dose is given: 0.66 mU. leads to a maximum value of 0.4 mU. in the kidney in 9 minutes. A slightly smaller dose will not produce enough ADH in the kidney to inhibit diuresis and so will be recorded as having no effect. A slightly larger dose will cause a perceptible effect, and will be recorded as having inhibited diuresis for 9 minutes or more. This is the explanation of the term $e^{-(\alpha-\beta)t}$ in equation (4), for when t is small this expression is large relative to unity and cannot be ignored. It can also be seen that equation (1)

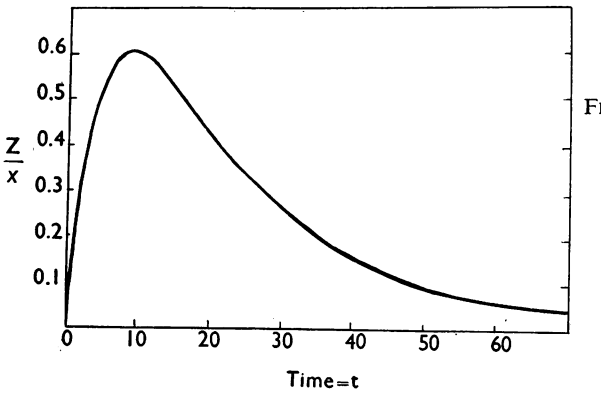

FIG. 4.—Relation between the fraction of an injected dose that is in the kidney and the time that has elapsed since injection. Ordinates: The proportion Z/x: Equation (3). Abscissae: Time in min. The fraction reaches a maximum of 0.61 of the injected dose, 9 min. after injection.

is only true when t is large. When t is less than 50 minutes, there ceases to be a linear relation between the time to the peak of diuresis following the inhibition, and the logarithm of the dose injected. Indeed, in reality, there is no linear relation at all; it is only seen because, when t is large, the difference between the theoretical relation and a linear relation is imperceptible.

Some evidence in favour of a non-linear relationship between the time to the peak and the logarithm of the dose, when the dose is small, is given by Verney (1947). Fig. 28 on page 75 of his paper shows some assay curves on the dog "Sally." The times to the peak (which are not, however, very clearly defined) with the doses 0.5, 1.0, and 2.0 mU. are evidently very similar, although the actual curves relating urinary excretion to the time elapsing since the injection are quite separate.

ADH in the urine.—It is possible that some ADH is filtered by the glomerulus and washed away in the profuse diuresis which is in progress when the injection is given. This would cause the rate of urine flow to change more slowly at the beginning of the onset of the inhibition than is to be expected from a consideration of the amount of hormone present.

The delay in the excretion of ingested water.—The peak of the diuresis which follows the ingestion of water is often hard to define. The peaks recorded in Table II, and to which Fig. 2 and equation (1) refer, related to a rate of urinary flow of about 150 ml. per 15 min. This rate of flow is generally first reached about 45 minutes after the ingestion of water. If at this time 0.4 mU. remains in the kidney (Z_0), equation (2) can be used to find out how much ADH was present when the water was ingested. According to the theory, k in equation (2) is equal to β in equation (3). As in fact k has a range of values, and the value 0.055 is within this range, no great error is introduced by assuming that k is equal to β. Then equation (2) shows that 4.8 mU. must have been present, some in the kidney and some still circulating in the blood.

Before the water was ingested, when the amount of hormone in the kidney was presumably constant, the rate of destruction of the hormone equalled the rate at which new hormone was brought by the blood. This gives 3.8 mU. in the kidney and 1.0 mU. in the whole of the blood. The rate of destruction is 0.21 mU. per minute in the kidney, or 3.5 μU. per second. The order of magnitude of these figures will vary considerably with the degree of hydration in a particular case.

It the time to a urinary flow of 150 ml. per 15 min. was 30 minutes instead of 45, the quantity present would be 2.1 mU., corresponding to a rate of destruction of 1.5 μU. per second.

The antidiuretic hormone liberated by nicotine.—The amount of antidiuretic hormone liberated by nicotine has been calculated from the period of inhibition produced, and in Table IV the results in smoking are tabulated. When the antidiuretic action lasted more than 120 minutes the amount of hormone liberated has been recorded as greater than 1,000 mU. If the amount is determined by extrapolation in Fig. 2 it would be much greater than 1,000 mU., and then would not agree with the amount liberated as calculated from the amount excreted in the urine (see Table IV). It was observed by Burn *et al.* (1945) that the injection of 100 mU. PLE caused an inhibition of 107 minutes, but that when this amount was given in two doses of 50 mU. each, separated by a period of 45 minutes, the period of inhibition was 140 minutes, which is an inhibition shown by the regression line to be caused by a single dose of 600 mU. When nicotine provides a prolonged stimulus to the pituitary as in smoking, then the antidiuretic hormone is likely to be released in a continuous stream throughout the smoking, and when it arrives at the kidney in this way a very much smaller total dose will provide an inhibition of a given duration. Thus, in Exps. 4 and 5, Table IV, the amount of hormone liberated in the blood when calculated from the amount found in the urine was 1,900 mU. If this amount was liberated all at once, it would cause an inhibition of 161 minutes. Actually the inhibition was 195 minutes, which corresponds to the liberation of 11,000 mU. all at once as determined by extrapolation in Fig. 2. If, however, 1,900 mU. were liberated, not all at once but during 23 minutes, it would certainly prolong the inhibition well beyond 161 minutes. Thus the discrepancy between the amount of hormone liberated (1) calculated from the duration of the antidiuretic action, and (2) calculated from the amount excreted in the urine, is probably explained.

SUMMARY

1. The antidiuretic response of a man to a range of doses of posterior lobe extract given intravenously has been measured in three subjects. A regression line relating dose to antidiuretic effect has been obtained. The action of the antidiuretic hormone has been considered mathematically.

2. The excretion of the antidiuretic hormone in the urine after intravenous injection of posterior lobe extract has been measured. For doses in the range of 180 to 1,500 milliunits the amount excreted was found to vary from 2.6 to 7.4 per cent, the mean figure being 5.0 per cent.

3. Some observations have been made of the amount of antidiuretic hormone excreted in the urine after the subjects had smoked cigarettes, and on other occasions after they had been injected with nicotine. When smoking is carried out so intensely that side-effects such as pallor, sweating, nausea, and giddiness are experienced, the amount of antidiuretic hormone liberated in the blood, as calculated from that which is excreted in the urine, is in the range of 1,000 to 2,000 milliunits. When smoking is carried out ordinarily, the amount liberated by one or two cigarettes, according to the sensitiveness of the subject, varies from 3 to 190 milliunits.

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