A PRELIMINARY REPORT

on

THE ACTION OF PITUITRIN ON THE KIDNEY

by

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Introduction.

In reviewing the literature it was found that a large amount of work had been done on the Action of Pituitrin on the Kidney. The results obtained by different workers were conflicting. One group claimed that an injection of the Extract of the Posterior Lobe of the Pituitary Body gave a diuretic effect, while the other group obtained an antidiuresis. It was this disagreement that led Dr. Stoland to suggest the problem, and my taking up this particular work.

Historical

The fact that extracts of the Posterior Lobe of the Pituitary Body had an effect upon the kidney action was first observed in 1901 by Magnus and Shäfer(10). These investigators injected the extract intravenously in anaestheized animals. They observed that there was a marked increase in the flow of urine in dogs, cats, monkeys, and rats. Their method was to place a canula in the bladder or ureter and record the number of drops of urine excreted both under normal and injective conditions. The diuresis that they obtained lasted from twenty to thirty minutes. Upon receiving an increased flow without vasodilation they concluded that the extract acted directly on the renal epithelium in bringing about the increased flow of urine.

In 1906 Shäfer and Herring (16) repeated the same work and arrived at the same conclusions. Their results were not so constant as those of Magnus and Shafer(10). In nineteen dogs they found that twelve showed a duirectic effect, in the other seven the flow was diminished. Houghton and Merrill(8) in 1908 found that in dogs the increased flow lasted on the average of nineteen minutes. They concluded that since there was a rise in the general blood pressure the increase in the flow of urine was dependent on that factor. Halliburton, Candler, and Sides(5) in 1909 found that an injection of the extract

in cats cause an increase in the flow of urine. In 1910 Ott and Scott(13) confirmed their results. Dale(3) in 1909 working with perfused kidneys of the dog and cat found that pituitary extracts caused a vaso-constriction of the renal vessels. Pentimalli and Quercia(14) in 1912 working with the isolated kidney of the rabbit found that in perfusion when the extract was added there was a diminished flow from both the ureter and renal vein. Hoskins and Means (7) in 1912 working with dogs under narcosis found an antidiuretic effect and concluded that is was not due to the rise in general blood pressure. King and Stoland (9) in 1913 working with etherized dogs, obtained upon intravenous injection of pituitrin a rise in the general blood pressure which lasted for ten minutes accompanied by a swelling of the kidney and an increased flow of urine fro twenty minutes. They concluded that the effect was due to vaso-dilation. Gabriels(4) in 1913 perfusing the isolated kidney of the dog found an increased flow from the ureter without vaso-dilation. Herring(6) in 1913 found that the extracts of the posterior lobe of the pituitary body had a diuretic effect on all animals, which those of the pars intermedia and anterior lobe did not possess. Cow(2) in 1914 found that in cats the diurectic effect lasted for fifteen minutes. In 1916 Von Myenbery(11) reports a decreased flow for eight to ten hours. In 1917 Motzfeldt(12) finds that under all conditions the subcutaneous injection of pituitrin checks

flow of urine, and concludes that this antidiuretic action is due to the stimulation of the Sympathic Nervous System, and that the renal vaso-motor system is in this respect of chief importance. In 1918 Addis, Barnett and Shevky(1) found that subcutaneous injections of pituitrin decrease the amount of urea that is excreted by the kidney, although the blood urea concentration is higher. In the same year Rees(15) found that injections of the extract cause no increase in the twenty-four hour output of urine, and concluede that the diuresis was offset by a later diminished flow, and that there was no variation from the daily output.

Discussion of Literature

It will be seen from the work done that the investigators used three different methods to arrive at their conclusions. The first class used anaesthetized animals; the second, perfused kidneys, and the third, animals without anaesthesia or surgical interference. The first method was the original one and was used by Magnus and Shafer(10). They used anaesthetized animals and inserted a canula directly into the bladder. Later investigators used this same method, except they placed the canula directly into the ureter. They then recorded the drops of urine that fell from the canula in a given length of time both under normal and injected conditions. In this method there are two factors which might influence the

action of the extract on the kidney. First, we do not know to what extent narcosis will effect the Sympathic Nervous System, and second, operations on the bladder or ureter as reflex action may interfere seriously with the normal activity of the kidney. Motzfeldt(12) in his work has shown that the action of pituitrin on the kidney is chiefly through its action on the Sympathic Nervous System. The second method, that of perfusion, has serious objections on account of abnormal conditions, and some experimentors (Pentinialli and Quercia(14), Gabrils(4)) have excised the kidney, thereby taking it away from all nervous control. The third class collected the urine from animals without anaesthesia or surgical intervention. The animals were placed in metabolism cages and the urine collected in that way, or they were catheterized at regular intervals. In placing animals in metabolism cages evaporation of urine and contamination with feces may bring in uncontrollable errors. In catheterization it is not always certain that all of the urine is expelled, since the position of the animal and the tonicity of the bladder wall may differ at different times. There may also be some reflex inhibition on the activity of the kidney when the catheter is inserted or withdrawn. The first investigators worked over an interval of several hours, They found a diuresis lasting about twenty minutes, and concluded that it was due to the rise in general blood pressure. Their injections were all intravenous.

Motzfeldt(12) worked over periods of six to eight hours and found an antiduresis, and concluded that it was due to the inhibition of the kidney action through the Sympathetic Nervous System. In 1918 Rees(15) collecting the urine from animals over periods of twenty-four hours found the daily output was not altered by injection of pituitrin, and concluded that the antidiuresis found by Motzfeldt(12) was due to intestinal staces brought about by the contraction of the smooth muscles of the intestine, which lasted from six to eight hours and delayed the absorption of water.

Method

In the experiments reported in this paper we have used a method which we believe permits of the collection of the urine as it flows from the kidney without contamination. In this method we are also quite certain that there are no reflexes that will interfere with the normal activity of the kidney. In all our work we used female dogs. A bladder fistula was prepared by stitching an opening in the bladder to the abdominal wall. The urethra was cut and both ends closed. It required about two weeks for the fistula to heal and the opening would diminish until the size of a lead pencil. This opening remained approximately this size during the life of the animal and all the urine flowed out through it. These animals were placed in a stand and sling and were allowed to

remain for twelve hours. A special catheter was placed in the fistual for collection of the urine. In this way the urine drained out as it was excreted. The urine was collected in a container packed in ice. A standard diet was fed to the dogs at a definite time every day. As long as the diet was constant the excretion remained the same within narrow limits. Dogs become accustomed to staying in the sling and allow themselves to hand limp and sleep most of the time. The normal position for a dog to rest is lying down. When in the sling they are on their ventral side with legs hanging down. It was found best to allow them to remain twelve hours in the sling, followed by twelve hours out. The twelve hours that were run could be either day or night. Since on a constant diet the excretion day after day was practically the same, experiment for the twenty-four hour period could be made by using twelve hours of one day and twelve hours of the following night. By this method we were able to get twenty-four hours samples without keeping the dogs suspended for more than twelve hours period. In order to arrive at definite conclusions concerning the activity of the kidney we concluded that we must make quanitative determinations of the nitrogenous compounds of both the blood and urine. We, therefore, made a quanitative determination of the total nitrogen, urea, and ammonia content of the urine and total nitrogen and urea content of the blood every four hours. The total nitrogen, urea,

and ammonia determinations were made by Folin Macro-Kjeldhl. Our blood analyses were made by the method outlined by Folin and Wu. In all cases the nitrogen was distilled over as ammonia, collected in N/50 acid and titrated with N/50 alkali. Duplicates were always run on all determinations and results checked in that way.

Results

Since we found that the normal urinary output of the same animal was practically the same when under uniform conditions we have in making our tables taken the average of the number of days run. When one cubic centimeter of Pituitrin "S" was injected subcutaneously it will be seen that the largest output of urine was during the first four hours. Here the volume was very much increased. The amount of total nitrogen, urea, and ammonia output were also much higher. The diuresis continued throughout the twelve hours, coming back more nearly to normal the last four hours. When one cubic centimeter of Pituitrin "S" was injected intravenously the diuresis did not go as high as when injected subcutaneously. The diuresis lasted throughout the first eight hours. During the last four hours the flow was practically normal. When one cubic centimeter of Pituitrin "S" was injected at seven a.m. and seven p.m. it caused a marked increase in both twelve hour intervals. During the next twelve hours, however, the nitrogenous content of the urine was much below normal,

although the volume excreted was normal. This shows that following a pituitrin diuresis the nitrogenous body content is below normal. When repeated injections were made at twenty-four hour intervals for three days, the volume of urine excreted was always very much above normal, while the nitrogenous content slowly dropped each day. As we have already shown that twenty-four hours after injection of Pituitrin "S" the nitrogenous content is far below normal, it goes to show that the injection of Pituitrin "S" will always cause an increased output of the urinary nitrogenous constituents, over that which it would have been had pituitrin not been injected.

In the following tables the grams of nitrogen indicated is the total content excreted in the urine over the period of four hours. The nitrogen content of the blood is expressed in grams per one hundred cubic centimeters.

Table 1., Dog 1.- Average of eight normal days.

	Urine				Blood	
Time	Volume	Total N.	Urea N.	AmmoniaN.	Total N.	Urea N.
11 a.m.	80	.7278	.3545	.1806	2.856	1.764
3 p.m.	34	.4586	.1594	.1403	3.444	1.541
7 p.m.	27	.4651	.1746	.1470	4.228	1.624

Table 2., Dog 1.- Average of three days, one cubic centimeter of Pituitrin "S" was injected subcutaneously at seven a. m.

	Urine				Blood	
Time	Volume	Total N.	Urea N.	Ammonia N.	Total N.	Urea N
11a.m.	115	1.1894	.8337	.2113	2.436	1.635
3p.m.	63	.7959	.4536	.2192	2.160	1.520
7p.m.	32	.5556	.2108	.2106	2.962	1.544

Table 3., Dog 1.-Average of two days, one cubic centimeter of Pituitrin "S" was injected intravenously at seven a. m.

	Urine				Blood	
Time	Volume	Total N.	Urea N.	Ammonia N.	Total N.	Urea N
11a.m.	113	.9723	.6561	.1869	2.240	1.4360
3p.m.	34	.4856	.1746	.1836	2.856	1.548
7p.m.	27	.4651	.1747	.1770	3.743	1.634

Table 4., Dog 1.- Average of two days, one cubic centimeter of Pituitrin "S" was injected subcutaneously at seven a. m., and seven p. m. the day before.

	Urine				Blood	
Time	Volume	Total N.	Urea N.	Ammonia N.	Total N.	Urea N.
11a.m.	80	.5297	.2663	.1617	2.453	1.544
3p.m.	38	.4138	.1358	.1344	3.256	1.502
7p.m.	26	.4056	.1468	.1470	3.842	1.624

Table 5., Dog 2.- Average of eight days normal.

	Urine				Blood	
Time	Volume	Total N.	Urea N.	Ammonia N.	Total N.	Urea N.
11a.m.	42	.7483	.4662	.1155	3.163	1.873
3p.m.	15	.3648	.2153	.0529	3.271	1.652
7p.m.	13	.4313	.2155	.0882	3.876	1.693

Table 6., Dog 2.- Average of three days, one cubic centimeter of Pituitrin "S" injected subcutaneously at seven a. m.

	Urine				Blood	
Time	Volume	Total N	Urea N.	Ammonia N	Total N.	Urea N
11a.m.	87	1.0617	.8626	.09828	2.734	1.743
3p.m.	33	.5170	.3889	.06930	2.342	1.601
7p.m.	19	.3088	.2016	.05460	3.163	1.586

Table 7., Dog 2.- Average of two days, one cubic centimeter injected intravenously at seven a. m.

	Urine				Blood	
Time	Volume	Total N	Urea N.	Ammonia N	Total N.	Urea N
11a.m.	58	.9450	.8295	.1087	2.364	1.563
3p.m.	19	.4830	.3158	.1101	2.458	1.628
7p.m.	12	.4013	.2436	.09468	2.830	1.673

Table 8., Dog 2.- Average of two days, one cubic centimeter of Pituitrin "S" injected subcutaneously at seven a. m. and seven p. m. the day before.

	Urine				Blood	
Time	Volume	Total N.	Urea N.	Ammonia N.	Total N.	Urea N.
11a.m.	50	.6948	.3946	.0834	2.050	1.352
3p.m.	18	.3428	.2056	.0910	2.263	1.264
7p.m.	14	.3163	.1653	.0923	2.181	1.392

Figure 1. Volume of urine plotted in cubic centimeters as the ordinate, and time in four hour periods as the absissa. The system of broken lines used in this and all following figures are thus explained:

———— Normal

-------- One c.c. of pituitrin "S" injected subcutaneously at seven a.m.

-.-.-.-.-. One c.c. of Pituitrin "S" injected intravenously at seven a.m.

– – – – – – Day following subcutaneous injection of pituitrin "S".

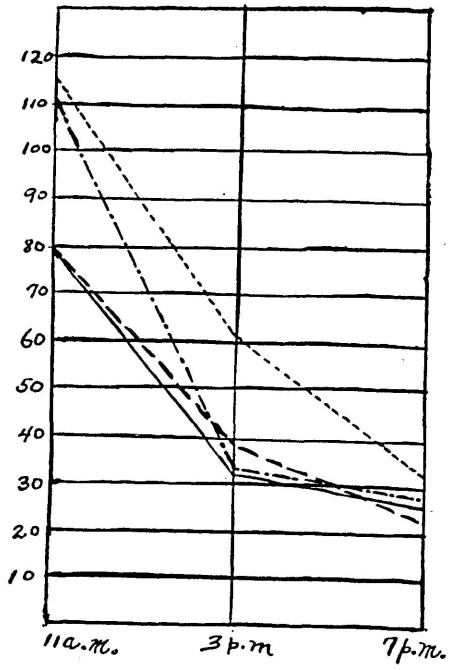


Figure 2. Total Nitrogen contentof blood and urine plotted with total nitrogen in grams as the ordinate and time in four hour intervals as the absissa.

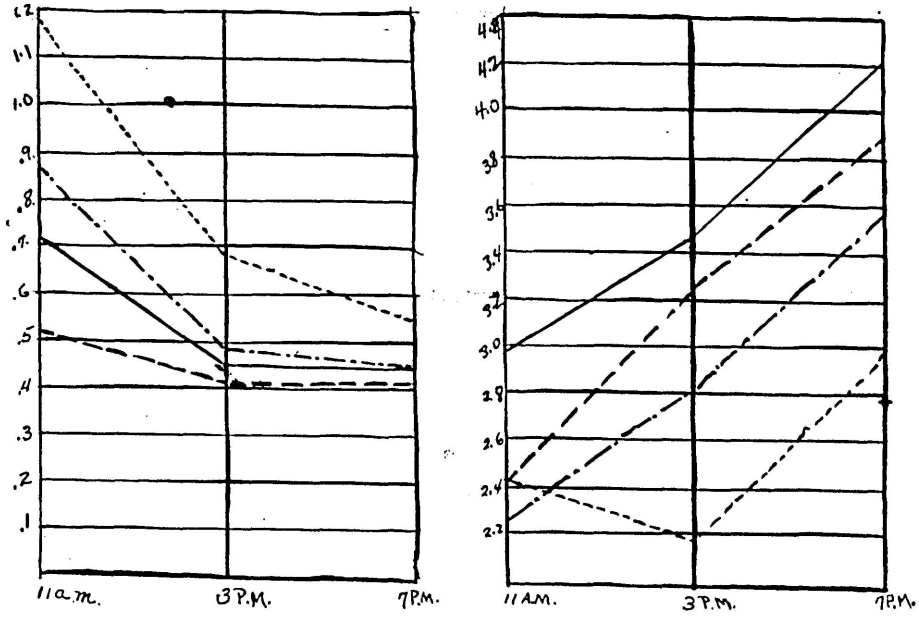


Figure 3. Urea Nitrogen content of the blood and urine plotted with urea nitrogen in grams as the ordinate and time in four hour intervals as the absissa.

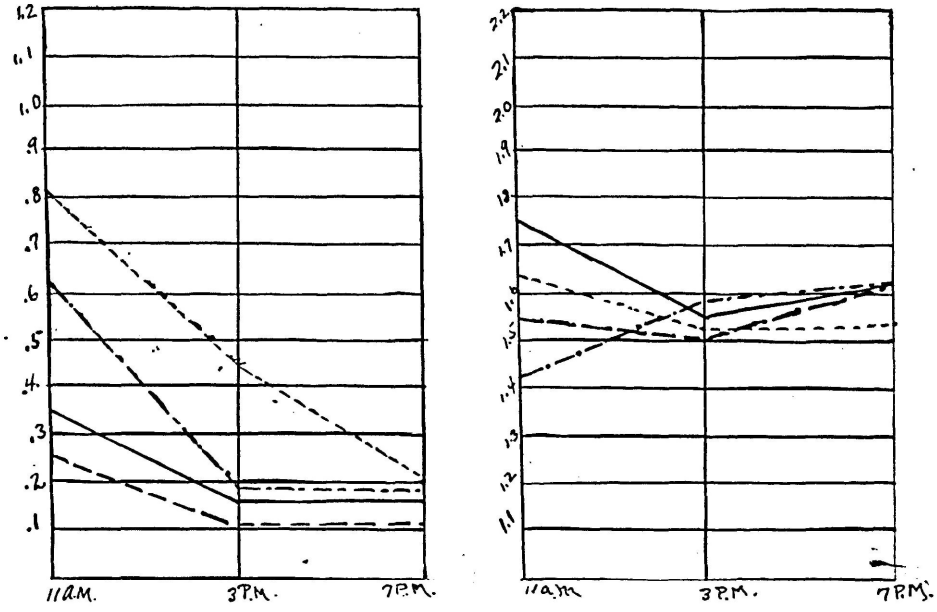
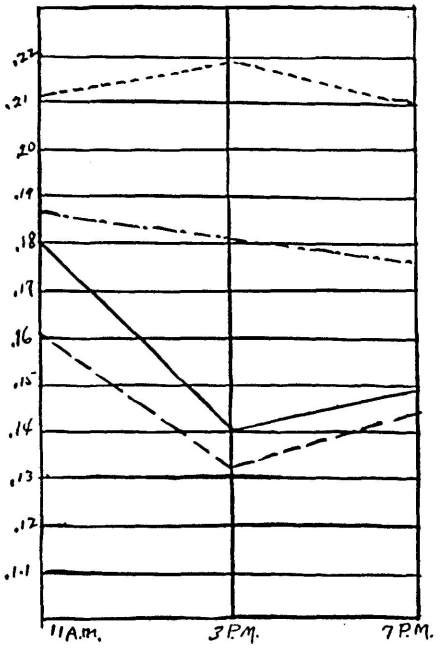


Figure 4. Amonia Nitrogen content of the urine plotted with amonia nitrogen in grams as the ordinate and the time in four hour intervals as the absissa.



Conclusion

1. The extract of the Posterior Lobe of the Pituitary Body always caused a diuresis whether injected subcutaneously or intravenously. The diuresis caused by subcutaneous injection lasted throughout the entire twelve hours. That of the intravenous injection came back to normal within eight hours and started to drop during the last four hours.

2. The nitrogenous content of the blood is decreased after injection.

3. The volume of urine excreted twenty-four hours after the injection is as great as normal but the nitrogenous content of both blood and urine is far below normal.

4. When injections were made at twenty-four hour intervals for three consecutive days the volume was far above normal and remained the same, while the nitrogenous content of the blood and urine gradually decreased, showing that pituitrin will always cause the same increased output in volume of urine over what it would have been had pituitrin not been injected, but not as high as the preceding injected day.

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