Nephrogenic Diabetes Insipidus

Effects of 3,5, Cyclic-adenosine Monophosphate

N. F. JONES, M. A. BARRACLOUGH, N. BARNES, and D. G. COTTOM*

From St. Thomas' Hospital and The Hospital for Sick Children, Great Ormond Street, London

Jones, N. F., Barracough, M. A., Barnes, N., and Cottom, D. G. (1972). *Archives of Disease in Childhood*, 47, 794. **Nephrogenic diabetes insipidus: effects of 3,5, cyclic-adenosine monophosphate.** In view of the evidence that the actions of vasopressin are mediated by the release of adenosine 3,5, cyclic monophosphate ('cyclic-AMP'), the effects of this nucleotide were studied in 3 children with nephrogenic diabetes insipidus (NDI) who were insensitive to vasopressin.

Intravenous injections of cyclic-AMP reduced urine flow in 2 of the 3 patients, but did not increase urinary osmolality nor the urinary concentrations of sodium, creatinine, and urea. In these respects the antidiuresis differed from that normally produced by vasopressin.

The reduction in urine flow produced by cyclic-AMP was probably related to a fall in glomerular filtration rate associated with the prominent circulatory effects of the nucleotide. These effects appear to preclude the use of cyclic-AMP as an antidiuretic agent in the treatment of NDI.

Nephrogenic diabetes insipidus is an hereditary disorder of the kidney characterized by insensitivity to the antidiuretic action of vasopressin (Waring, Kajdi, and Tappan, 1945; Forssman, 1956). Evidence for the current belief that vasopressin is present in this disorder but that the nephron cannot respond to it has been summarized by Orloff and Burg (1966). The basis for the failure to respond to vasopressin is unknown.

There is evidence that vasopressin influences the metabolism of the renal medulla *in vivo* (Jones and Welt, 1967). Vasopressin is believed to act at cellular level by increasing the formation of adenosine 3,5, cyclic monophosphate ('cyclic-AMP'), which then increases epithelial permeability. Both cyclic-AMP and vasopressin increase the epithelial permeability to water and stimulate active sodium transport in the isolated toad bladder (Orloff and Handler, 1962; Edelman, Petersen, and Gulyassy, 1964). Both substances increase the permeability to water of isolated rabbit collecting tubules (Grantham and Burg, 1966). The concentration of cyclic-AMP in toad bladder cells is increased by vasopressin, and this hormone

also stimulates production of cyclic-AMP by homogenates of dog kidney (Handler *et al.*, 1965; Brown *et al.*, 1963).

Cyclic-AMP is present in human urine (Butcher and Sutherland, 1962) and its excretion may increase in response to vasopressin (Pawelson *et al.*, 1970). Moreover, the administration of cyclic-AMP causes antidiuresis (Levine, 1968; Barracough and Jones, 1970). There is thus considerable evidence that vasopressin's actions on the kidneys are mediated by cyclic-AMP. It is possible that in NDI the defect at cellular level involves a failure to generate cyclic-AMP in response to vasopressin. If this were true, then the nucleotide might retain its antidiuretic action thereby offering an approach to therapy. The effects of cyclic-AMP were therefore tested in patients with NDI.

Patients and Methods

The relevant features of the 3 patients studied are shown in Table I. In each patient the diagnosis was established in infancy by the absence of a detectable response to vasopressin after correction of the initial fluid deficit and at a time when the blood urea level was normal. There was no evidence of other defects in tubular function. Case 2 was complicated by dilatation of the urinary tracts. This patient was studied after bilateral ureterostomies had been performed, when the

Received 6 March 1972.

*Dr. Cottom died on 13 August 1971. For a brief obituary note see Volume 46, 728, 1971.

TABLE I
Clinical Features of the Patients Studied

Case No.	Sex	Age (yr)	Family History of NDI	Creatinine Clearance ml/min per 1.73 m ²	Blood Urea mg/100 ml
1	M	15	—	115	12
2	M	6	+	82	32
3	M	6/12	—	105	30

blood urea was normal and the creatinine clearance was 82 ml/min per 1.73 m².

The studies were performed only when the children were well hydrated and this was supported by a normal plasma osmolality in each case. Fluid was infused intravenously at a rate to match the urine output. 2.5% dextrose was used in Cases 1 and 2, but Case 3 received 4.8% dextrose in 0.18% saline as this solution had been used immediately before the study when the infant had been vomiting about once a day.

Urine was collected by spontaneous voiding in Case 1, via the ureterostomy tubes draining each kidney in Case 2, and by bladder catheter in Case 3. Venous blood was sampled only 3 times (Cases 1 and 2) or twice (Case 3), as it was expected that close matching of fluid input to urine output should lead to steady concentrations of plasma solutes. Changes in the concentrations of these solutes varied by under 4% in all studies.

Intravenous injections of vasopressin ('Pitressin', Parke Davis), cyclic-AMP, and normal saline were given into the tubing of the intravenous drip. The cyclic-AMP used in Cases 1 and 2 was supplied by Sigma

Chemicals. That used in Case 3 was donated by Koch-Light Chemicals Ltd.

Osmolality was measured with an Advanced Osmometer, creatinine and urea by Autoanalyser (Technicon), and sodium by flame photometer.

The following formulae were used in the calculations:

$$\text{Osmolal clearance: } C_{\text{Osm}} = \frac{U_{\text{Osm}} V}{P_{\text{Osm}}}$$

$$\text{Free water clearance: } C_{\text{H}_2\text{O}} = V - C_{\text{Osm}}$$

where U_{Osm} = Urinary osmolality, mOsm/kg
 P_{Osm} = plasma osmolality, mOsm/kg
 V = urine flow, ml/min.

Results

Urine flow fell briefly after each dose of cyclic-AMP in Cases 1 and 3 but did not change appreciably in Case 2 (Fig. 1 to 3). Urinary osmolality and urinary sodium concentration were not altered detectably by cyclic-AMP (Table II). Whenever cyclic-AMP reduced urinary flow there were associated falls in osmolal clearance, free water clearance, and in creatinine and urea clearances. All these clearances returned to control levels concurrently with the return of urine flow to control. When cyclic-AMP did not reduce urine flow (Case 2) no significant change occurred in these renal clearances.

Injections of vasopressin (Cases 1 and 3), or of saline (Case 2), were without apparent effect. In the doses used cyclic-AMP always caused a brief tachycardia lasting 2 to 10 minutes. In Case 1, the first dose of cyclic-AMP (40 mg) caused a

TABLE II
Details of Urine Flow and Urinary Solute Concentrations Before and After Injections of Cyclic-AMP

Case No.	Period*	Urinary Concentration				
		Urine Flow ml/min	Osmolality mOsm/kg	Na mEq/l.	Urea mg/100 ml	Creatinine mg/100 ml
1	Control	12.25	52	18	58	5.1
	C-AMP (1 mg/kg)	3.30	47	14	76	9.2
	Control	10.67	38	12	68	6.8
	C-AMP (1.5 mg/kg)	2.27	39	12	73	9.2
2	<i>Left Kidney</i>					
	Control	3.80	68	10	175	5.6
	C-AMP (1.2 mg/kg)	3.56	71	12	173	5.1
	Control	3.83	73	15	154	5.1
	C-AMP (3 mg/kg)	3.90	66	13	153	5.3
	<i>Right kidney</i>					
	Control	1.52	69	14	152	5.1
	C-AMP (1.2 mg/kg)	1.56	71	15	149	4.5
3	Control	1.50	74	19	135	4.5
	C-AMP (3 mg/kg)	1.40	69	17	135	4.8
	Control	3.89	95	43	142	3.2
	C-AMP (1 mg/kg)	2.50	93	43	144	3.4
	Control	4.03	90	40	167	3.3
	C-AMP (2 mg/kg)	2.32	100	42	168	3.4

*The 'control' period values are the means of 3 consecutive 5 to 12 minute collection periods immediately before cyclic-AMP was given. The values shown in each 'C-AMP' period refer to the period in which urine flow was lowest after cyclic-AMP. In every case this was the period immediately after injection of cyclic-AMP.

FIG. 1.—Effect of vasopressin and of cyclic-AMP on renal function in Case 1.

FIG. 2.—Effect of NaCl and of cyclic-AMP on the function of the right kidney in Case 2. Effects on the left kidney were similar.

FIG. 3.—Effect of cyclic-AMP and of vasopressin on renal function in Case 3.

transient fall in blood pressure from 104/72 to 98/50 mmHg, but no change in blood pressure was detected after the second dose. In Case 2, cyclic-AMP 25 mg produced no detectable change in blood pressure, but this changed from 105/65 to 105/45 mmHg after 62.5 mg. Blood pressure was not measured in Case 3.

Case 1 felt 'light-headed' and faint for about 5 minutes after each dose of cyclic-AMP. Facial flushing was observed in Cases 2 and 3 after each dose.

Discussion

Though cyclic-AMP transiently reduced urine flow in Cases 1 and 3, it did not increase urinary osmolality nor the urinary concentrations of sodium, creatinine, or urea. In these respects its actions differ completely from those of vasopressin. In view of the observed circulatory changes and because of the known cardiovascular actions of cyclic-AMP it is likely that a fall in glomerular filtration rate caused antidiuresis.

It is possible that continued exposure to cyclic AMP might allow the development of a tubular

response to this nucleotide, or that use of the more permeant dibutyryl cyclic-AMP, might produce an antidiuresis resembling that normally caused by vasopressin. The present results do not therefore establish firmly that the tubular defect in NDI is after the stage at which cyclic-AMP is formed. Any attempt at circumventing this problem was prevented by the unacceptable circulatory effects already caused by the dosage used and it is concluded that cyclic-AMP has no therapeutic value in NDI.

We thank Koch-Light Chemicals Limited for supplying some of the cyclic-AMP used.

REFERENCES

- Barraclough, M. A., and Jones, N. F. (1970). Effects of adenosine 3',5'-monophosphate on renal function in the rabbit. *British Journal of Pharmacology*, **40**, 334.
- Brown, E., Clarke, D. L., Roux, V., and Sherman, G. H. (1963). The stimulation of adenosine 3',5'-monophosphate production by antidiuretic factors. *Journal of Biological Chemistry*, **238**, 852.
- Butcher, R. W., and Sutherland, E. W. (1962). Adenosine 3',5'-phosphate in biological materials. I. Purification and properties of cyclic 3',5'-nucleotide phosphodiesterase and use of this enzyme to characterize adenosine 3',5'-phosphate in human urine. *Journal of Biological Chemistry*, **237**, 1244.
- Edelman, I. S., Petersen, M. J., and Gulyassy, P. F. (1964). Kinetic analysis of the antidiuretic action of vasopressin and adenosine-3',5'-monophosphate. *Journal of Clinical Investigation*, **43**, 2185.
- Forssman, H. (1956). On hereditary diabetes insipidus. *Acta Medica Scandinavica*, **121**, Suppl. 159, 1.
- Grantham, J. J., and Burg, M. B. (1966). Effect of vasopressin and cyclic-AMP on permeability of isolated collecting tubules. *American Journal of Physiology*, **211**, 255.
- Handler, J. S., Butcher, R. W., Sutherland, E. W., and Orloff, J. (1965). The effect of vasopressin and of theophylline on the concentration of adenosine 3',5'-phosphate in the urinary bladder of the toad. *Journal of Biological Chemistry*, **240**, 4524.
- Jones, N. F., and Welt, L. G. (1967). Adenosinetriphosphate in rat renal papilla: effects of vasopressin and of ischemia. *American Journal of Physiology*, **212**, 939.
- Levine, R. A. (1968). Antidiuretic responses to exogenous adenosine 3',5'-monophosphate in man. *Clinical Science*, **34**, 253.
- Orloff, J., and Burg, M. B. (1966). Vasopressin-resistant diabetes insipidus. In *The Metabolic Basis of Inherited Disease*, 2nd ed., p. 1247. Ed. by J. B. Stanbury, J. B. Wyngaarden, and D. S. Fredrickson. McGraw-Hill, New York.
- Orloff, J., and Handler, J. S. (1962). The similarity of effects of vasopressin, adenosine 3',5'-phosphate (cyclic AMP) and theophylline on the toad bladder. *Journal of Clinical Investigation*, **41**, 702.
- Pawson, L. G., Taylor, A., Mintz, D. H., Field, J. B., and Davis, B. B. (1970). Effect of vasopressin on renal cyclic-AMP generation in potassium deficiency and patients with sickle hemoglobin. *Metabolism*, **19**, 694.
- Waring, A. J., Kajdi, L., and Tappan, V. (1945). A congenital defect of water metabolism. (Abst.) *American Journal of Diseases of Children*, **69**, 323.

Correspondence to Dr. N. F. Jones, St. Thomas' Hospital, London S.E.1.