Effect of the vasopressin antagonist d/CH₂/5Tyr/Et/VAVP on the antidiuretic action of exogenous and endogenous vasopressin

F. A. László, S. Csáti and L. Baláspiri

Endocrine Unit, First Department of Medicine, and
Department of Medical Chemistry, University Medical School, Szeged, Hungary

Abstract. The effect of [1-(β-mercapto-β,β-cyclopentamethylene-propionic acid),2-0-ethyltyrosine,4-valine]-arginine vasopressin on the water metabolism was studied in rats. The compound decreases the antidiuretic action of exogenous vasopressin in Brattleboro rats; in rats without diabetes insipidus it causes temporary polyuria and eliminates the response of antidiuresis to an osmotic stimulus. The results indicate that this compound can block the antidiuretic action of both exogenous and endogenous vasopressin.

The attention of researchers dealing with the preparation of vasopressin analogues has recently turned to the synthesis of antagonists. Manning and his colleagues have described a number of compounds which behave as vasopressin antagonists (Manning et al. 1981a,b; Sawyer et al. 1981). From among these analogues we have examined the effect of [1-(β-mercapto-β,β-cyclopentamethylene-propionic acid),2-0-ethyltyrosine,4-valine]-arginine vasopressin (d/CH₂/5Tyr/Et/VAVP) on the water metabolism in rats. We studied how this compound influences the antidiuretic action of exogenous and endogenous vasopressin.

Material and Methods

The vasopressin antagonist d/CH₂/5Tyr/Et/VAVP was provided by Professor Manning (Toledo, Ohio); the details relating to its preparation are to be found in the publications by his team (Manning et al. 1981a,b). The chemical composition of this analogue is shown in Fig. 1.

Compared with the structure of the vasopressin molecule, differences may be observed at three sites. The hemicysteine at position 1 is replaced by cyclopentamethylene-propionic acid; the hydroxyl group on the tyrosine residue at position 2 contains an O-ethyl substituent; the glutamine at position 4 is replaced by valine. Previous experiments demonstrated that the analogue causes a considerable decrease in the antidiuretic action of vasopressin (Manning et al. 1981a,b; Sawyer et al. 1981). This property of the antagonist is characterized by the 'effective dose'; this is defined as the quantity of the substance which halves the antidiuretic effect observed 20 min after iv administration of 2 IU vasopressin. In the present case this proved to be 1.9 nmol/kg (1 nmol = 1.14 µg). The antagonism can be regarded as selective. The compound also displays a marked antipressor action: Manning et al. (1981b) found the 'effective antivasopressor dose' to be 0.49 nmol/kg.

We carried out three experimental series of examinations. It was investigated how this antagonist influences the antidiuretic action of an exogenously administered vasopressin preparation. Female homozygous rats of the Brattleboro strain, weighing 160–180 g, were used; these animals do not possess an endogenous ADH reserve. Doses of 10 µU or 30 µU arginine vasopressin (Organon, Oss) were injected iv into animals prehydrated by the method of De Wied (1960) and anaesthetized with ethanol. The antidiuretic effect was expressed as a percentage diuresis change; this was taken as the quantity of urine collected with a bladder catheter in a 10 min period before injection of the hormone, as a percentage of the corresponding quantity after the injection. The antagonist was administered iv in the 'effective dose' (1.9 nmol/kg = 2.16 µg/kg) 20 min before the injection of vasopressin.

We subsequently studied whether d/CH₂/5Tyr/Et/
VAVP is able to suspend the antidiuretic effect of endogenous vasopressin, and whether it can induce a state corresponding to diabetes insipidus. Male rats of the R-Amsterdam strain, weighing 180–200 g, were used in these experiments. The animals were placed individually in cages suitable for urine collection; the amount of urine was measured in 1 h periods for 4 h after administration of the antagonist, and the osmolality of the urine samples was determined with an Advance osmometer. Both before and during the study the rats received water ad libitum. The antagonist was injected iv in a dose of 10 or 30 µg/kg, dissolved in 0.2 ml physiological NaCl. Besides the rats of the R-Amsterdam strain, homozygous Brattleboro rats too were used as controls. Instead of the antagonist, the control groups received 0.2 ml physiological NaCl solution iv.

In the following experiments, physiological NaCl solution in a dose of 5% of the body weight was administered via a stomach tube to rats of the R-Amsterdam strain, and the urinary output was measured in 1 h periods for 4 h.

In the meantime the animals did not consume liquid. This method is suitable for the production of water retention by the increase of endogenous vasopressin excretion (Kovács et al. 1962; László & Kovács 1968). Instead of physiological NaCl solution, tap-water was administered to the control group. The vasopressin antagonist, in a dose of 30 µg/kg, was injected iv immediately before the loading with physiological NaCl or tap-water.

Results

Fig. 2 shows the data relating to diuresis inhibition, expressed as a percentage of the urinary output. Arginine vasopressin decreases the urinary output in a dose-dependent manner. Administration of the antagonist before the arginine vasopressin injection moderated the diuresis inhibition considerably.

The results of the experiments aimed at suppressing the antidiuretic effect of endogenous vasopressin are given in Figs. 3–5. It may be observed that a 10 µg/kg body weight dose of the antagonist caused hardly any change in the urinary output (Fig. 3), whereas the dose of 30 µg/kg body weight enhanced the diuresis appreciably. The maximum in the poluria is to be seen in the fraction from the second hour. In this period the quantity of urine attained the level for the homozygous Brattleboro rats with a total ADH deficiency. The diuretic action of the antagonist is a temporary one; after 4 h the urinary output has
returned to the normal level. The urinary osmolality varies accordingly (Fig. 4). It is decreased by the vasopressin antagonist in a dose-dependent way. With this method a considerable difference can be demonstrated between the osmolality values for the controls and even that following administration of the low dose of the antagonist. After water loading, the antagonist enhances the diuretic reaction only moderately (Fig. 5). The administration of physiological NaCl in place of tap-water led to a marked water retention in the control group. The antagonist averted this effect completely; the extent of urine excreted in the fraction from the second hour exceeded by far the mean measured after tap-water loading.

Discussion

Our results demonstrate that the vasopressin antagonist examined, d/CH₂5Tyr/Et/VAVP, is able to block the antidiuretic effect of exogenous vasopressin, and diminishes the effect of basal endogenous vasopressin, therapy inducing temporary polyuria and eliminating antidiuresis in response to an osmotic stimulus. Other investigators too have described the antidiuretic antagonist action of d/CH₂5Tyr/Et/VAVP (Manning et al. 1981a,b; Sawyer et al. 1981). These properties mean that the compound is of promise from the aspect of clinical application.
One of the main conditions for the practical introduction of vasopressin antagonists is that they should be highly potent and selective compounds. However, two problems must be considered: a) the strength of the effect of the antagonist is much lower than that of the naturally occurring hormone agonist, arginine vasopressin. In human (Edwards 1977) and in rat (Dogterom et al. 1978; Moore et al. 1977) the effective circulating hormone levels is in the pg/ml order, whereas the antidiuretic effect of endogenous vasopressin could be blocked with a 30 µg/kg dose. The dose difference is even more striking in the experiment aimed at averting the antidiuresis caused by exogenous vasopressin. To halve the antidiuretic effect of 10 pg (3 µU) arginine vasopressin, 4.0–4.3 µg antagonist had to be administered to the homozygous Brattleboro rats, with no endogenous vasopressin reserve and b) d/CH2/5-Tyr/Et/VAVP possesses an appreciable antipressor effect. The ‘effective antipressor dose’ of the compound (0.49 nmol/kg = 0.56 µg/kg) is lower than the effective antidiuretic dose. When the drug is used, therefore, the blood pressure-decreasing effect must be taken into account.

Recently, D-tyrosine has been built into the molecule in place of the L-tyrosine at position 2 (Manning et al. 1982a). The ‘effective dose’ of the antidiuretic antagonist d/CH2/5-D-Tyr/Et/VAVP is 1.1 nmol/kg, but the strength of its antipressor effect is the same as that of the L-tyrosine analogue. The most potent antidiuretic antagonist compound found so far is d/CH2/5-D-Phe-VAVP (‘effective dose’ = 0.67 nmol/kg); here too the antipressor action is marked (‘effective dose’ = 0.58 nmol/kg) (Manning et al. 1982b). The most promising compound appears to be d/CH2/5-D-Leu-VAVP, which primarily exerts antidiuretic antagonist action (‘effective dose’ = 1.2 nmol/kg) (Manning et al. 1982b), while its antipressor effect is more than 20 times weaker (‘effective dose’ = 26 nmol/kg).

Research on the synthesis of vasopressin analogues has therefore not been completed. For the treatment of hyperadiuretinism, the primary need is for an antagonist of high efficacy and selectivity, which is available in sufficiently large amount for human therapy.

References


Received on October 4th, 1983.