In vitro Studies of the Mechanism of Polyuria Induced by Dextro Propoxyphene (Darvon).* (30472)

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In a recent case report of dextro propoxyphene (Darvon®) intoxication(1), polyuria was a prominent clinical feature. Urine volume in excess of 5 liters per 24 hours in the presence of both hypotension and hypernatremia suggested the syndrome of diabetes insipidus. The lack of response to large doses of exogenous antidiuretic hormone (Pitressin®) further implied antagonism of antidiuretic hormone at the renal tubular level. This possibility was examined further in the isolated surviving toad bladder, an *in vitro* analogue of the mammalian distal tubule-collecting duct complex that permits direct evaluation of such effects in the absence of other complicating variables.

Materials and methods. A single hemibladder from a freshly pithed *Bufo marinus* was suspended between the two parallel chambers of a Peterson-Edelman volume flow apparatus(2) which had been modified to permit monitoring of water flow across 2 adjacent areas of the membrane (Fig. 1). The serosal chambers were filled with normal Ringer solution (Na 113.4, K 3.4, Cl 120, HCO_3 2.4, Ca 2.7, glucose 5.5 millimoles per liter, pH 8.1, osmolality 240 milliosmoles per kilogram) and the mucosal chambers with Ringer solution diluted 1:10 with distilled water (osmolality 24 milliosmoles per kilogram). The serosal chambers remained open and the

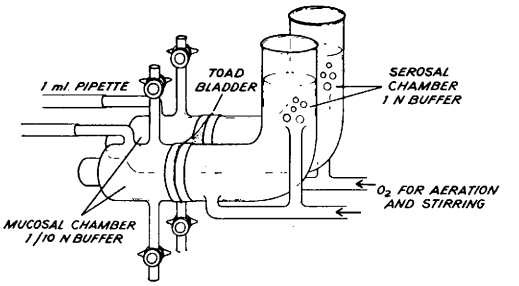

FIG. 1. Apparatus for measurement of net water movement (double chamber).

buffers were continuously stirred and oxygenated by the passage of a small stream of room air. After the mucosal chambers were filled, the stopcocks were closed and the net movement of water across the hemibladder from the mucosal to the serosal chamber was monitored by the movement of the maniscus in horizontally mounted 1 ml serologic pipettes attached to the 2 mucosal chambers. Volume changes were measured at 10-minute intervals and net water flow expressed in microliters per minute.

Low permeability to water under baseline conditions is indicated by the low volume flow in the control period (Fig. 2). When antidiuretic hormone (Pitressin®) was added to both the control and the experimental serosal chambers in a final concentration of 5 milliunits per milliliter, there was a marked

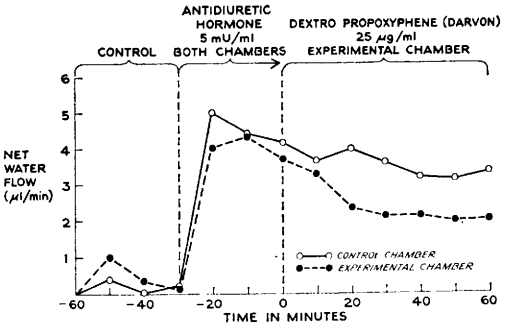

FIG. 2. Representative experiment using dextro propoxyphene (Darvon), 25 μg/ml.

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TABLE I. Results of Dextro and Levo Propoxyphene Studies *in vitro*.

	Conc (μg/ml)	Δ Flow rate (μl/min)	n	p
Dextro propoxyphene	25.0	$-.7 \pm .15$*	5	$<.01$
	10.0	$-.3 \pm .09$	6	$<.02$
	2.5	$-.1 \pm .10$	5	NS
Levo propoxyphene	25.0	$-1.5 \pm .36$	6	$<.01$
	2.5	$-1.1 \pm .20$	9	$<.01$

* $\pm$ S.E.

increase in membrane permeability to water and net water flow across the 2 areas of the hemibladder.

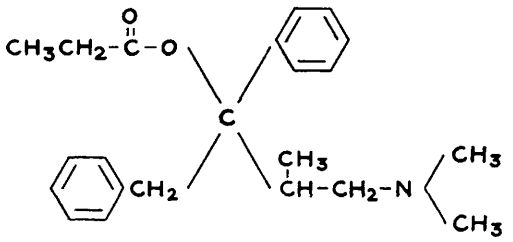
Although the absolute flow between the 2 sides differed, this difference remained relatively constant so that the curves of water flow remained parallel. When crystalline dextro propoxyphene§ was added to the serosal medium of the experimental chamber, inhibition of antidiuretic hormone action resulted in a decrease in flow on the experimental side. Inhibition was defined as the decrease in flow in the experimental chamber compared to the parallel control during the 30- to 60-minute period after addition of dextro propoxyphene compared to the 30-minute period immediately preceding its addition.

Results. Dextro propoxyphene, 25 μg per milliliter, significantly inhibited the net water flow induced by 5 milliunits per milliliter of antidiuretic hormone. Inhibition was also significant at a concentration of 10 μg per milliliter but none was apparent at lower concentrations (Table I).

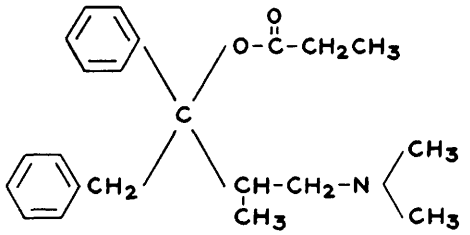
Levo propoxyphene, which differs from dextro propoxyphene only in the orientation of groups on its first asymmetric carbon (Fig. 3), significantly inhibited the net water flow induced by 5 milliunits per milliliter of antidiuretic hormone at concentrations of 25 and 2.5 μg per milliliter.

A β-diastereo isomer and several possible metabolic end products of dextro propoxyphene (α-d-4-dimethylamino-1,2-diphenyl-3-methyl-2-butanol HCl, α-d-1,2-diphenyl-3-methyl-4-methylamino-2-propionoxy butane maleate, α-d-1,2-diphenyl-3-methyl-4-methylamino-2-butanol HCl) did not produce any antagonism of antidiuretic hormone at similar concentrations.

§ Eli Lilly Co., Indianapolis, Ind.



DEXTRO PROPOXYPHENE



LEVO PROPOXYPHENE

FIG. 3. Molecular configuration of dextro and levo propoxyphene.

Fifty microliters of a concentrated solution of dextro propoxyphene added to the 20 ml of normal Ringer's solution and antidiuretic hormone in the experimental chamber gave a final concentration of dextro propoxyphene of 25 μg per milliliter (pH of 8.0, osmolality of 24.15 milliosmoles per liter). It was not felt that pH or osmolality changes would have any significant effect on the membrane.

Discussion. The case report(1) of marked polyuria after a fatal dose of dextro propoxyphene (Darvon) suggested a diagnosis of diabetes insipidus. Polyuria is not a common clinical finding in drug intoxication(3), particularly when associated with hypotension requiring correction with pressor amines. The absence of glycosuria, the presence of hypernatremia and the correction of the latter by large amounts of intravenous dextrose and water are evidence against osmotic diuresis as an explanation of the polyuria. The lack of response to exogenous antidiuretic hormone further suggested hormonal antagonism at the renal tubular level but the critical state of the patient made it impossible to exclude defective release of antidiuretic hormone from the hypothalamic-posterior hypophyseal axis. Transient hypokalemia and the electrocardio-

graphic changes of potassium depletion were noted(4), suggesting that the inhibition of antidiuretic hormone action might have occurred indirectly as a result of potassium depletion nephropathy(5). No information is available concerning the presence or absence of hypercalcemia, which also inhibits antidiuretic hormone action at the renal tubule(6).

The isolated surviving toad bladder has been shown to possess many physiologic features in common with the mammalian distal tubule-collecting duct complex. These include low permeability to water, which is increased by antidiuretic hormone(7), similar increases in permeability to urea and related small molecules(7), and sodium transport that is stimulated by mineralocorticoid(8). The use of the isolated surviving toad bladder in these experiments has permitted direct demonstration of dextro propoxyphene inhibition of antidiuretic hormone in a system that obviates considerations of direct drug effects on release of antidiuretic hormone by the pituitary, changes in glomerular filtration rate and excretion of solutes as well as indirect effects on antidiuretic hormone action through electrolyte imbalance.

It should be emphasized that the concentration of dextro propoxyphene used in these experiments (10 to 25 μg per milliliter) is similar to that noted in the kidney of experimental animals receiving the drug (11 to 62 μg per g wet weight)(9). The ability to demonstrate *in vitro* inhibition of antidiuretic hormone action at similar concentrations supports direct antagonism by dextro propoxyphene at the renal tubular level as an explanation of the observed polyuria.

In animal experiments it has been noted that levo propoxyphene also has diuretic properties and antagonizes the effect of antidiuretic hormone(9). In the isolated surviving toad bladder, antagonism of antidiuretic hormone action at a concentration even lower than that of dextro propoxyphene was demonstrated. Several chemical analogues of this series failed to do so.

Polyuria was not mentioned in other case reports of dextro propoxyphene ingestion(10-12) or in animal studies(9,13). However, on direct inquiry, polyuria was acknowledged to be present in one case(14), and the amounts of drug ingested were either trivial or unknown in several others. The absence of overt polyuria may be attributable to differences in the doses of the drug employed or reported, in metabolism of the drug or in compensatory release of pituitary antidiuretic hormone.

In view of the dearth of effective, nontoxic, peripheral antidiuretic hormone antagonists, further exploration of dextro and levo propoxyphene and their derivatives is being undertaken in the search for an agent to use in states of antidiuretic hormone excess(15).

Summary. The isolated surviving toad bladder was used to demonstrate direct peripheral antagonism between antidiuretic hormone and both dextro propoxyphene (Darvon®) and levo propoxyphene (Norvad®). These findings suggest that the polyuria accompanying a recent death after dextro propoxyphene ingestion was a result of drug-induced nephrogenic diabetes insipidus.

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