Heparin Mediates Transmembrane Potassium Transfer in Hyperkalemic Dogs (43141)

NATHAN HIATT AND JONATHAN R. HIATT

Department of Surgery and the Medical Research Institute, Cedars-Sinai Medical Center, Los Angeles, California 90048 and Department of Surgery, University of California at Los Angeles School of Medicine, Los Angeles, California 90024

Abstract. **Unheparinized, ureter-ligated control dogs that are potassium loaded, i.e., infused with 2 mEq of KCl/kg until prelethal electrocardiographic changes of hyperkalemic cardiotoxicity appear (end point), transfer 57 ± 4% (1.7 ± 0.1 mEq/kg) of administered potassium to intracellular fluid. Heparinized controls transfer 73 ± 1% (3.2 ± 0.2 mEq/kg); with simultaneous α-adrenoreceptor blockade, that proportion increases to 81 ± 2% (4.80 ± 0.7 mEq/kg) and with simultaneous β-receptor blockade it is 58 ± 3% (1.1 ± 0.1 mEq/kg). In potassium loaded, ureter-ligated dogs, heparin increases transmembrane potassium transfer as effectively as does a dosage of atropine large enough to cross the blood-brain barrier and its influence on potassium transfer, like that of atropine, is suppressed by β-adrenoreceptor blockade.** [P.S.E.B.M. 1990, Vol 195]

This investigation was prompted by the serendipitous finding that heparin improved potassium transfer from extracellular fluid (ECF) to intracellular fluid (ICF) in ureter-ligated (UL) dogs; it was made in the course of a totally unrelated investigation—potassium transfer in cross-circulated animals loaded with KCl. In subsequent studies of heparinized UL dogs continuously infused with 2 mEq of KCl/kg body wt until prelethal electrocardiographic (ECG) changes of hyperkalemic cardiotoxicity appeared, we found that transfer was greater than that of unheparinized controls, and we discovered that α-adrenoreceptor blockade enhanced the improvement and that β-adrenoreceptor blockade prevented it. Findings will be discussed.

Materials and Methods

Data were gathered from 21 mongrel dogs of either sex that weighed between 15.9 and 21.8 kg. After an 18-hr fast, they were anesthetized with intravenous sodium pentobarbital (30 mg/kg), ventilated with a Harvard respirator, and infused with about 25 ml/hr of 0.15 *M* NaCl via an intravenous drip. In all 21 dogs, the abdomen was opened by a midline abdominal incision and both ureters were ligated. Twelve dogs were heparinized by rapid infusion of 60 mg (6 ml–10,000 units) of commercial heparin; in two instances, this was followed by an intravenous injection of 30 mg 2 hr later. In four of the heparinized dogs, β-receptors were blocked with propranolol 15 min before the start of KCl administration by immediately following a priming intravenous injection of 0.3 mg/kg with an infusion (at 30 ml/hr) of a solution that contained 0.3 mg/kg. In four unheparinized and four heparinized dogs, α-adrenoreceptors were blocked in exactly the same way, by substituting 2.5 mg of phentolamine for the 0.3 mg/kg propranolol. Animals were potassium loaded by the 0.15 *M* NaCl drip being replaced with a Harvard peristaltic pump that delivered 30 ml/hr and a KCl solution; the concentration of the solution was such that each animal received 2 mEq/kg/hr. Protocols are given in the first column of Table I.

During potassium loading, lead II of an ECG (Hewlett-Packard Co., Palo Alto, CA) was monitored frequently; infusion was continued to the end point, i.e., until prelethal ECG changes of hyperkalemic cardiotoxicity appeared—ventricular bradycardia of <5 beats in 15 sec, ventricular flutter (no isoelectric phase), or a bizarre QRS pattern (W or sine wave shaped) (1). At the end point, KCl infusion was discontinued immediately.

Blood specimens from a femoral vein were obtained at the start of KCl infusion, at 30-min intervals thereafter, and at the end point. Serum potassium was

Received November 14, 1989. [P.S.E.B.M. 1990, Vol 195]
Accepted June 21, 1990.

0037-9727/90/1952-0237$2.00/0

Table I. UL Dogs Infused to End Point with 2 mEq of KCl/kg Body Weight

Group (no. of dogs)	Potassium (mEq/kg)	ΔK[a] (mEq/kg)	Potassium trans[b] (mEq/kg)	P[c] (%)	Ins[d] (microunits/ml)
1: Control, unheparinized (5)	3.0 ± 0.3[e]	1.3 ± 0.3	1.7 ± 0.1	57 ± 4	44 ± 6
2: Control, heparinized (4)	4.3 ± 0.2	1.2 ± 0.3	3.2 ± 0.2	73 ± 1	31 ± 9
P	<0.05	NS[f]	<0.001	<0.05	NS
3: Heparinized, phentolamine (4)	5.9 ± 0.7	1.1 ± 0.1	4.8 ± 0.7	81 ± 2	33 ± 8
P versus Group 2	NS	NS	<0.05	<0.05	NS
4: Heparinized, propranolol (4)	1.9 ± 0.1	0.9 ± 0.1	1.1 ± 0.1	58 ± 3	9 ± 9
P versus Group 1	<0.05	NS	<0.05	NS	<0.01
5: Unheparinized, phentolamine (4)	3.5 ± 0.7	1.4 ± 0.1	2.1 ± 0.1	59 ± 1	48 ± 4
P versus Group I	NS	NS	NS	NS	NS

[a] (End point − preinfusion mEq/liter serum) × 0.23.
[b] Potassium in −ΔK.
[c] $\frac{\text{Potassium trans}}{\text{Potassium in}} \times 100$.
[d] Maximum − preinfusion serum IRI.
[e] Mean ± SE.
[f] $P > 0.5$ by Student's *t* test.

determined by an Instrumentation Laboratory Flame Photometer (Lexington, MA) with a lithium internal standard, hematocrit was determined by the routine laboratory method, and serum-immunoreactive insulin (IRI) was determined by radioimmunoassay (2). Arterial blood pH was not measured since it has proved noncontributory in potassium-loaded dogs (3).

On average, an increase of serum potassium by 1 mEq/liter is equivalent to the addition (potassium increment) of 0.23 mEq of potassium/kg body wt (0.2 mEq of potassium/kg to 200 ml/kg of ECF (4) and 0.03 mEq of potassium/kg to 30 ml/kg of red blood cells [RBC] (5). When KCl infusion is halted, the total potassium increment to ECF and RBC is 0.23 times ΔK (end point minus preinfusion mEq/liter of serum potassium). Potassium transferred to ICF is equal to KCl infused minus total potassium increment. In dog RBC, sodium not potassium, is the main intracellular ion; the potassium concentrations of RBC hemolysates and ECF are similar (6). The amount of KCl that can be infused before prelethal ECG changes appear is dependent on ΔK and the quantity of potassium transferred to ICF. The percentage of infused potassium transferred to ICF not only measures the quantity of potassium transferred to ICF but also permits the comparison of transfer capacities. All values are reported as mean ± SE. Data were analyzed by the two-tailed unpaired Student *t* test, and the null hypothesis was rejected at the 0.05 level.

Results

Mean preinfusion serum potassium was 4.0 ± 0.3 mEq/liter and that of serum IRI was 10 ± 3 microunits/ml. During the course of infusion, average serum potassium concentration rose to 9.1 ± 0.3 mEq/liter and in all but one instance (Group 4) serum IRI also increased; the hormone level was usually maximal at the end point. Since the greatest volume of solution administered was less than 5 ml/kg (<1% total body water) and the maximum hematocrit change was +5 vol % (in a single unheparinized control), it was held that there were no unusual changes in ECF and ICF volumes.

Group 1 consisted of control, unheparinized dogs (Table I). Mean potassium transfer measured less than 60% of that infused, and the average increase of serum IRI concentration was approximately 45 microunits/ml.

Group 2 was control, heparinized dogs (Table I). The proportion of the potassium load and the actual quantity of potassium transferred to ICF were greater than that of unheparinized controls; the increase of serum IRI concentration was not significantly different from that in the dogs of Group 1.

Group 3 consisted of dogs treated with heparin and phentolamine (Table I). The proportion of the potassium load and the actual quantity of potassium transferred to ICF were greater than that of heparinized controls; the increase of serum IRI was statistically similar to that in Groups 1 and 2.

Group 4 was one of dogs treated with heparin and propranolol (Table I). Potassium transfer was like that of unheparinized controls; serum IRI concentration was similar to that of potassium-loaded dogs with β-receptor blockade (3).

Group 5 consisted of unheparinized dogs treated with phentolamine (Table I). Potassium transfer and the increase of serum IRI concentration were both within the range of that in unheparinized controls.

Discussion

Heparin improves potassium transfer in control potassium-loaded UL dogs; α-adrenoreceptor blockade

increases the improvement and β-adrenoreceptor blockade prevents it (Table I). In unheparinized animals, α-receptor blockade does not influence potassium transfer (Table I) and β-receptor blockade reduces it (3).

In control, unheparinized potassium-loaded UL dogs, when serum potassium reaches about 7.2 mEq/liter, potassium transfer capacity changes. Below that level, potassium transfer and the straight line that marks the increase of serum potassium concentration are like those in control preparations with intact renal function. Above that level, in those with UL, there is a reduction of potassium transfer; the line that marks the increase of serum potassium skews upward, and prelethal ECG changes of hyperkalemic cardiotoxicity quickly appear. Thus, although end point serum potassium concentration is alike in both controls (9–10 mEq/liter), *overall* potassium transfer in the UL potassium-loaded dogs is significantly less (7). In heparinized potassium-loaded UL controls, when serum potassium reaches about 7.2 mEq/liter, there is no reduction of potassium transfer capacity; the linear increase of serum potassium concentration continues unchanged to the same end point as before, and overall potassium transfer increases, i.e., becomes similar to that of controls with intact renal function (8) (Table I).

In potassium-loaded UL dogs, afferent impulses in the cervical vagi and certain muscarinic receptors in the brain are part of a renal-adrenal reflex that modulates neural traffic to the adrenal medulla. Treatment with atropine, *in dosages large enough to cross the blood-brain barrier* (and thus influence central modulation of potassium transfer), eliminates the fall of potassium transfer at 7.2 mEq/liter (9). Heparin penetrates the tight endothelial junctions and crosses the blood-brain barrier in perfused rat brains, but there is no evidence of entry into brain tissue (10). Heparin, like atropine, eliminates the reduction of potassium transfer capacity when serum potassium reaches about 7.2 mEq/liter and, in both, that effect is abolished by β-receptor blockade (9) (Table I). Neither heparin nor adequate dosages of atropine influence potassium-loaded dogs with nephrectomy or intact renal function (personal observation).

Commercial heparin, a mixture of anionic, polyelectrolyte-sulfated polymers of alternating glucoseamide and uronic acid rings, is used almost exclusively as an anticoagulant (11); however, among its other properties is the ability to inhibit adrenal aldosterone production (12). It is well known that renal activity of aldosterone influences electrolyte metabolism and that the mineralocorticoid has extrarenal activity (13). However, that activity does not include potassium transfer and it is generally agreed that the hormone acts over a period of days to weeks (14); none of the experiments reported lasted over 3 hr.

A human with traumatic shock and anuria at the time of the trauma is potassium loaded by the release of potassium-rich ICF from crushed soft tissue. Heparin has been reported to protect against experimental trauma (11).

This work was Supported by NIH General Research Support Grant and the Blum-Kovler Foundation.

We gratefully acknowledge the help of Ms. Joye L. Nunn-Hill, the staff at Ms. Ellen Green's Health Science Information Center, and Dr. M. B. Davidson and Robert Karajala.

1. Surawicz B. Role of electrolytes in etiology and management of cardiac arrhythmias. Prog Cardiovasc Dis **8**:364–387, 1966.
2. Soeldner J, Slone D. Critical variables in the radioimmune assay by the double antibody technique. Diabetes **14**:771–779, 1965.
3. Hiatt N, Chapman LW, Davidson MB. Influence of epinephrine and propranolol on transmembrane K transfer in anuric dogs with hyperkalemia. J Pharmacol Exp Ther **209**:282–286, 1979.
4. Walser M, Seldin DW, Grollman A. The evaluation radiosulfate for the determination of extracellular fluid space in man and dogs. J Clin Invest **32**:299–311, 1953.
5. Hoff HE, Deavers J, Huggins, RA. Effect of hypertonic glucose and mannitol on plasma volume. Proc Soc Exp Biol Med **122**:630–634, 1966.
6. Coldman MF, Good W, Quinn PL, White IG. Blood and other body fluids. FASEB J **3**:1778, 1974.
7. Hiatt N, Chapman LW, Davidson MB, Sheinkopf JA, Miller A. Inhibition of transmembrane K transfer in ureter-ligated dogs infused with KCl. Am J Physiol **231**:1660–1664, 1976.
8. Hiatt N, Chapman LW, Mack H, Davidson MB, Sheinkopf J. Transmembrane K transfer in hyperkalemic dogs. Horm Metab Res **13**:386–389, 1981.
9. Hiatt N, Chapman LW, Mack H, Hiatt JR. K transfer in ureter ligated dogs loaded with KCl. Biomed Biochim Acta **4–5**:395–400, 1988.
10. Nagy Z, Peters H, Huttner I. Charge-related alterations of the cerebral endothelium. Lab Invest **49**:662–671, 1983.
11. Jacques LB. Heparins-anionic polyelectrolyte drugs. Pharmacol Rev **31**:99–166, 1977.
12. O'Kelly R, Magee F, McKenna TJ. Routine heparin therapy inhibits adrenal aldosterone production. J Clin Endocrinol Metab **56**:108–112, 1983.
13. Spital A, Sterns RH. Extrarenal potassium adaption: The role of aldosterone. Clin Sci **76**:213–219, 1989.
14. Young DB. Quantitative analysis of aldosterone's role in potassium regulation. Am J Physiol **255**:F811–F822, 1988.