Effect of ambient cold on urine concentration by the intact and denervated dog kidney[1]

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Summary. In dogs transferred from room temperature to a low temperature of 1 ± (SD)3.5 °C, the plasma norepinephrine (NE) level increased significantly. Urine concentration decreased in the cold, in the intact and similarly in the chronically denervated kidney. After return to a warm environment urine concentration improved in both kidneys while plasma NE remained elevated. The data speak against an essential role of the renal nerves in the adaptation of renal function to ambient cold.

The mechanism causing the reduction in urine concentration at low ambient temperatures (cold diuresis) appears to be very complex and may involve changes in the activity of antidiuretic hormone (ADH), glucocorticoids and the adrenergic system[2-4]. The latter is unequivocally stimulated by cold and might affect urine concentration either by opposing ADH release or its action on the tubule[5-7].

The present study attempts to define the role of sympathetic input to the kidney via renal nerves in renal function changes occurring in dogs exposed to cold. Our experimental model; conscious, unilaterally renal-denervated animals, prepared for separate urine collection from the intact and denervated kidney, seemed to be particularly well-suited for the purpose.

Material and methods. Mongrel female dogs weighing 11-24 kg were prepared for experiments by left renal denervation and surgical division of the urinary bladder performed under sodium pentobarbital anesthesia. The denervation consisted of cutting all visible nerve fibres in the renal hilum and exposing the renal artery to 10% phenol solution in absolute ethanol. Polyethylene cannulas were implanted into the surgically formed hemibladders to permit urine collection separately from each kidney. The details of anesthesia and surgical procedures have been described in a recent publication, which also documents the effectiveness of denervation by demonstrating the disappearance of norepinephrine from denervated kidney tissue[8].

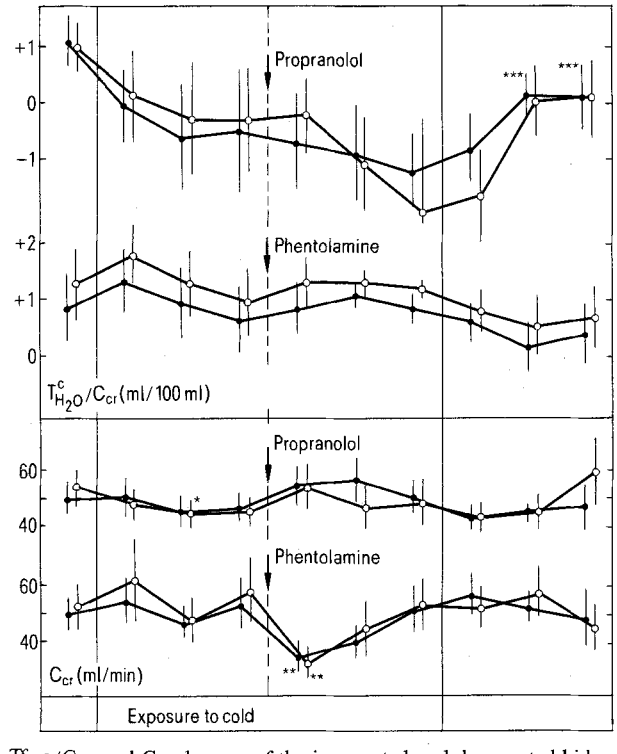
In 4 dogs the aorta was chronically catheterized via the right omocervical artery[9] for measurement of arterial pressure. Experiments were performed in dogs that had fully recovered from surgery, 2-5 weeks after renal denervation, and had been trained to stand quietly in the Pavlov sling. Renal clearances of exogenous creatinine (C_{cr}) and p-aminohippurate (C_{PAH}) were measured by a standard constant rate infusion technique as described previously[8]. Osmolar clearance (C_{osm}) and sodium excretion ($U_{Na}V$) were also determined. The free water reabsorption per 100 ml glomerular filtrate ($T^c_{H_2O}/C_{cr}$) was used as an index of urine concentration.

After due equilibration time 3 10-15 min control clearance periods were obtained at an ambient temperature of 19 ± (SD)2 °C. Subsequently, the dogs were transferred together with the Pavlov sling to the outdoor winter temperature of +1 ± 3.5 °C where they remained for 6 clearance periods, and then were transferred back to a room temperature of 18 ± 2 °C to obtain 3 final bilateral urine collections (recovery periods). In addition to this basic protocol, after 3 periods in cold the dogs were given an i.v. injection of propranolol, 0.25-0.3 mg/kg b.wt (7 experiments), or phentolamine, 0.35 mg/kg b.wt (7 experiments). In 2 experiments no drugs were administered.

Analytical methods for plasma and urine osmolality, sodium, exogenous creatinine and PAH have been described previously[8]. In control periods, after 30 min in the cold, and at the end of recovery periods, plasma norepinephrine was determined in 5 dogs by the fluorometric method[10]. The statistical significance of differences between means was tested using Student's t-test.

Results and discussion. The table shows, jointly for all 16 experiments, urine flow (V), glomerular filtration (C_{cr}) and urine concentration ($T^c_{H_2O}/C_{cr}$) changes during the 1st 3 clearance periods of exposure to cold. $T^c_{H_2O}/C_{cr}$ tended to increase in control periods but started to decrease in the cold and after 30 min the values for the intact and denervated kidney were significantly lower than for the control. These data are in agreement with the previously reported decrease in free water reabsorption during exposure of dogs to moderate cold[3].

The activation of the adrenergic system at low ambient temperature is well established and has been confirmed for our experimental conditions by norepinephrine (NE) studies. Plasma NE increased from a control value of 1.05 ± (SE) 0.1 µg/l to 1.44 ± 0.3 µg/l after about 30 min of exposure ($n = 5$, $p < 0.01$). Increased sympathetic activity could impair urine concentration by antagonizing ADH



$T^c_{H_2O}/C_{cr}$ and C_{cr} changes of the innervated and denervated kidney after propranolol and phentolamine injections during exposure to cold. Mean values ± SE. Open or full circles denote denervated and innervated kidney, respectively. *Significantly different from the mean control value before exposure. **Significantly different from the last period before phentolamine injection. ***Significantly different from the last period in cold.

Urine flow (V), glomerular filtration (C_{cr}) and free water reabsorption ($T^c_{H_2O}/C_{cr}$) in the innervated (I) and denervated (D) kidney as affected by exposure to cold (mean values ± SE, n = 16)

		Control periods			Exposure to cold		
		1	2	3	4	5	6
V	I	1.24±0.18	1.34±0.20	1.16±0.15	1.44±0.15	1.40±0.19	1.70±0.29
ml/min	D	1.31±0.22	1.41±0.24	1.30±0.16	1.48±0.16	1.43±0.14	1.60±0.22
C_{cr}	I	49.2±4.2	51.7±4.3	45.4±3.5	51.6±4.6[a]	45.2±3.6	50.1±4.6
ml/min	D	55.8±6.1	53.3±4.4	50.7±4.5	53.7±6.7	46.3±3.8	51.3±5.1
$T^c_{H_2O}/C_{cr}$	I	1.27±0.20	1.14±0.24	1.40±0.27	1.09±0.34	0.82±0.43[b]	0.92±0.45[c]
ml/100 ml	D	1.27±0.26	1.31±0.26	1.50±0.28	1.28±0.41	0.89±0.49	0.74±0.45[a]

[a] Significantly different from the last control period at $p<0.05$; [b] at $p<0.02$; [c] at $p<0.01$.

action on the renal collecting duct[5] or by inhibiting ADH release[6,7]. Since in the present experiments the decrease in $T^c_{H_2O}/C_{cr}$ occurred both in the innervated and denervated kidney, it is unlikely that this effect is mediated by renal nerves; however, the action of circulating NE cannot be excluded.

GFR and, not shown in the table, C_{PAH}, increased transiently at the beginning of exposure to cold; the increase was more pronounced and significant in the innervated kidney. The mechanism of this change is unclear. Stimulation of β-adrenoreceptors was shown to cause renal vasodilatation in conscious dogs[11], but it is unlikely that this effect should prevail over vasoconstriction dependent on the stimulation of α-receptors. If plasma dopamine level rises in the cold, in concert with the 2 other catecholamines, renal hemodynamics could increase depending on the well-known dilatatory action of this mediator on kidney vessels[13]. Urine flow, U_{osm}, C_{osm}, $U_{Na}V$ and $U_{Na}V/100$ ml filtration rate did not change significantly after exposure to cold.

Effects of β-adrenoreceptor blockade with propranolol, and α-adrenoreceptor blockade with phentolamine, on $T^c_{H_2O}/C_{cr}$ and C_{cr} during exposure to cold are presented in figure. Within the 2 subgroups renal function parameters were highly variable and the changes related to exposure to cold or drug injection were often not significant. Thus, for instance, decreases in free water reabsorption during the 1st 30 min of exposure to cold were not significant for either of 2 subgroups whereas they were highly significant for the whole experimental series (table). Fortuitously, in animals that were later given propanolol $T^c_{H_2O}/C_{cr}$ decreased rather steeply in the cold while in those later given phentolamine the decreasing tendency was preceded by an initial increase.

Propranolol appeared to enhance the fall in $T^c_{H_2O}/C_{cr}$ observed after exposure to cold, particularly in the denervated kidney. Let it be noted that NE, acting on β-receptors, was reported to stimulate adenyl cyclase activity in the rabbit collecting tubule[13]. If accumulation of cyclic AMP leads to increased permeability of the tubule to water, β-receptor blockade would be expected to impair urine concentration, as was indeed observed in our experiments. Since the effect was more pronounced in the denervated kidney, it would be due to circulating NE to which this kidney might be hypersensitive. However, Rayson et al.[6] were unable to demonstrate any effect of β-receptor stimulation on the water permeability of the collecting duct.

Propranolol injection during exposure to cold did not affect mean blood pressure. It increased slightly GFR of the innervated kidney while ipsilateral V, C_{osm} and $U_{Na}V$ increased significantly ($p<0.02$, $p=0.05$ and $p<0.05$, respectively).

Phentolamine injection during exposure to cold decreased mean arterial pressure from 109±(SE)7 mm Hg to 85±7 mm Hg. Simultaneously, GFR (fig.) and C_{PAH} decreased on the average 41% for the denervated and 33% for the intact kidney ($p<0.01$ and $p<0.05$, respectively). Unlike propanolol, phentolamine appeared to stop the decreasing trend in $T^c_{H_2O}/C_{cr}$ observed in the cold. However, the concurrent fall in GFR per se could have been responsible for a relative improvement of urine concentration. Urine flow, C_{osm} and $U_{Na}V$ for both kidneys decreased significantly after phentolamine, in parallel with the fall in renal hemodynamics.

At the end of the experiments, 30–45 min after return to room temperature, plasma NE level was still 1.37±0.21 µg/l i.e. only 0.07 µg/l below the concentration measured in the cold. Therefore, the impressive recovery of $T^c_{H_2O}/C_{cr}$ after cessation of exposure to cold was not related to decreasing adrenergic activity as reflected by plasma catecholamine level. More specifically, since the recovery occurred both in the intact and in the denervated kidney, it was unrelated to possibly decreasing impulse traffic in the renal sympathetic nerves.

No recovery of free water reabsorption was observed in dogs treated with phentolamine. However, it should be noticed that in this subgroup urine concentration decreased only slightly and not significantly in the cold. In neither of the 2 subgroups was the transition from a cold to a warm environment associated with significant changes in renal hemodynamics or parameters describing renal excretion.

In summary, the data failed to expose any major difference in the response to cold between the innervated and denervated kidney. Accordingly, the renal nerves seem to have no essential role in adaptation of renal function to low ambient temperature.

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