high osmolarity and reduction of blood volume with low osmolarity. In the first case tremendous release of ADH was noted. This 27-fold increase of plasma ADH level (Table I) is apparently due to concomitant responses to these two potent stimuli. In dehydration with low plasma osmolarity there was also a significant elevation in plasma ADH level though less marked in degree. This 3-fold increase of plasma ADH level (Table II) is in agreement with the observation of Mirsky *et al*(14). They found that in adrenalectomized rats, a state of hypo-osmotic dehydration with salt loss, plasma ADH level increased 3 to 4 times that of control rats within 7 days after adrenalectomy. Our results imply that these two stimuli are basically independent. Elimination of osmolar stimulus did not abolish the response to reduction of blood volume.

Comparison of the hematocrit (V_c) in Tables I and II indicates that the dehydration produced by sucrose diuresis or by shifting fluid into peritoneal cavity is about the same in the two experimental procedures; sample B is 1.22 $\times$ sample A in Table I and sample B is 1.30 $\times$ sample A in Table II.

Summary. Release of antidiuretic hormone in two types of dehydration was studied in monkeys. It was found that dehydration with high plasma osmolarity led to a tremendous elevation of plasma ADH level, and dehydration with low plasma osmolarity also led to a significant increase of plasma ADH level though less marked in degree.

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Demonstration of Increased Catecholamine Excretion in the Nephrotic Syndrome.* (32306)

WILLIAM J. OLIVER, ROBERT C. KELSCH, AND JOSEPH P. CHANDLER
(Introduced by John M. Weller)
Department of Pediatrics and Communicable Diseases and Department of Biological Chemistry, University of Michigan, Ann Arbor

Hypovolemia, secondary to reduced circulating albumin, is postulated to stimulate increased secretion of aldosterone in the nephrotic syndrome(1-3); however, measurements during diuresis, when aldosterone excretion is low, have not consistently shown an increase of plasma volume(4-5). Physiologic responses to hypovolemia also include increased activity of the autonomic nervous system (adrenergic)(6). This suggested that investigation of catecholamine excretion

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might provide an approach to studying the circulatory changes postulated to occur in the nephrotic syndrome. The data presented demonstrate increased excretion of norepinephrine in edematous nephrotic children. The simultaneous, but less marked increases of epinephrine, metanephrines and vanilmandelic acid excretion support the concept that the augmented excretion of norepinephrine reflects increased release of catecholamines during the edematous phase of the nephrotic syndrome.

Subjects and methods. Seven children with the nephrotic syndrome and 7 control children, ages 8 to 18 years, were studied. The nephrotic children had no hematuria, azotemia or hypertension and steroid treatment induced a complete remission of the disease. Subjects were housed in a metabolic unit and ate a constant diet containing 2-3 mEq Na/kg/day. Activity was not regulated beyond requiring a constant time for rising and retiring. Observations were made for 3 days prior to initiating steroid therapy (prednisone 1½-2 mg/kg/day) and continued through the period of diuresis. After diuresis, patients were maintained at home on an intermittent steroid program (prednisone 1½-2 mg/kg/day on 3 successive days of each week). Each subject was re-evaluated within 4 months. Treatment was discontinued 7 days prior to readmission to the metabolic unit. After one day to permit adjustment to the diet, observations were made for 3 consecutive days.

The control subjects consisted of 3 normal siblings of nephrotic children and 4 other children with no prior history of renal disease. The control subjects were studied in the same manner as the nephrotic children when in remission.

Blood specimens were obtained in the fasting state. Freshly voided urine specimens were measured and each specimen was divided into 2 equal parts: half was placed in a dark glass bottle containing 15 ml of 1 N HCl and the remainder in an electrolyte-free bottle. The specimens were refrigerated at 0-4°C until the 24-hour collection was complete and then were held at —20°C until analyzed. Sodium, potassium, creatinine, aldosterone and protein were determined by methods previously described (5). Urinary epinephrine (E) and norepinephrine (NE) were measured by von Euler's modification of the trihydroxyindole method (7). Recoveries of norepinephrine were 83 ± 8%,† and of epinephrine 96 ± 16%.† Vanilmandelic acid (VMA) was measured by the method of Pisano *et al*(8). Recoveries averaged 99.5 ± 3.0%.† Metanephrine (M) and normetanephrine (NM) were determined by the method of Brunjes(9), modified by prior precipitation of urine protein with 10% trichloroacetic acid. Recoveries of total metanephrines were 53 ± 6% and coefficient of variation was 12%. Results for aldosterone, NE, E, NM, M, and VMA were expressed in excretion rates per mg creatinine per 24 hours. For all subjects, the excretion rates of catecholamines under the described conditions represent mean values of 3 consecutive days. Completeness of urine samples was confirmed by relative constancy of 24-hour creatinine excretion.

TABLE I. Urinary Excretion in Nephrotic Children.

Epinephrine excretion (ng/mg creatinine/24 hr)		
Edematous nephrotics	(N = 7)	13.1 ± 3.3 *
Nephrotics in remission		8.7 ± .86*
.25 > p > 0.1		
VMA excretion (μg/mg creatinine/24 hr)		
Edematous nephrotics	(N = 7)	5.35 ± .76*
Nephrotics in remission		4.56 ± .49*
p = not significant		

* SEM

Results. The 24-hour excretions of NE, E, and VMA for nephrotic children in complete remission were not significantly different from control children (Fig. 1). During active nephrotic disease with edema, values for NE excretions were higher ($p<0.05$) than in the same nephrotic children in remission (Fig. 2). Excretions of E and VMA were also higher in edematous nephrotic children but the values were not significantly different (Table I). The increased catecholamine excretion seen in 5 of the edematous nephrotic children was associated with either a simi-

† Standard Deviation.

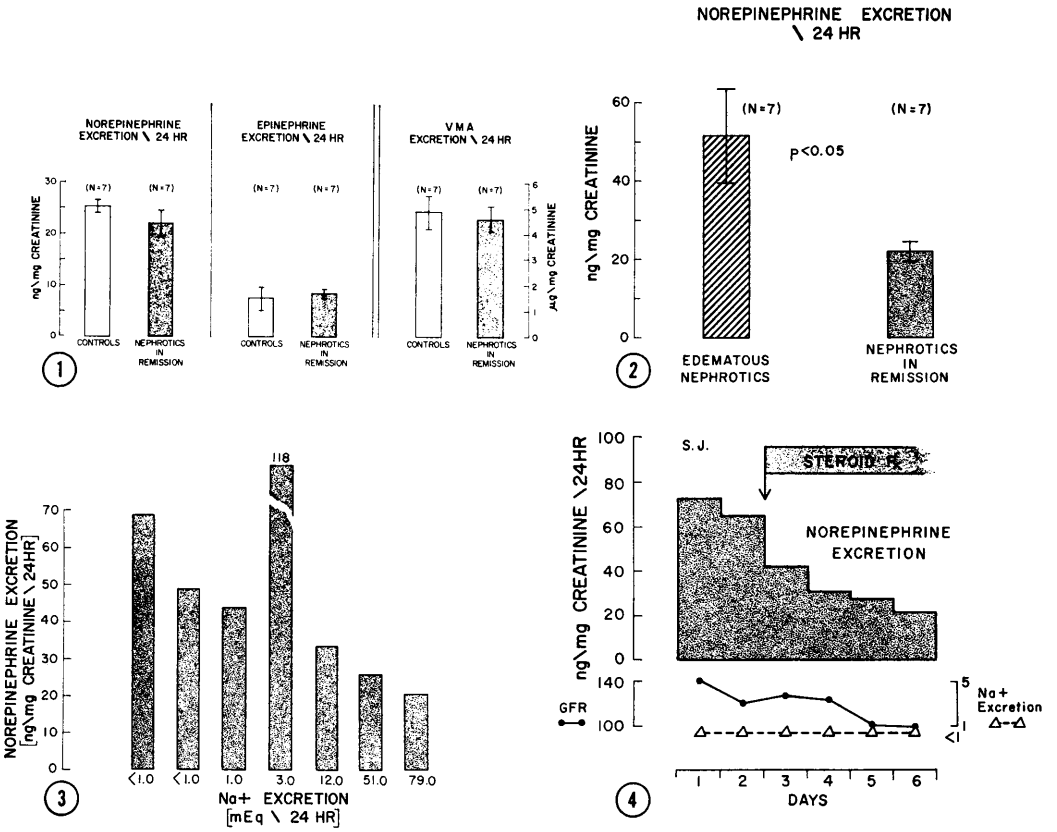

FIG. 1. Mean catecholamine excretion rates (average of 3 days) for 7 control children and 7 nephrotic children in remission. Vertical bars represent SEM.

FIG. 2. Mean NE excretion rates (average of 3 days) for 7 edematous nephrotic children compared with the same children in remission. Vertical bars indicate SEM.

FIG. 3. Relationship between mean urinary NE excretion and mean urinary sodium excretion in 7 edematous nephrotic children during constant high salt diet.

FIG. 4. Serial changes in NE and sodium excretion in an edematous nephrotic child coincident to steroid treatment. Values for urinary sodium excretion expressed in mEq/24 hours.

lar or decreased glomerular filtration rate, as estimated by endogenous creatinine clearance, when compared to rates obtained during remission. In the remaining 2 patients, glomerular filtration rates during edema were increased above values found in remission. Proteinuria decreased during steroid therapy but in the same subject similar quantities of protein excretion before and during therapy were often associated with marked differences in NE excretion (Table II). The magnitude of the NE excretion in the edematous children was inversely related to the quantities of sodium excreted despite similar sodium intake (Fig. 3). Norepinephrine excretion in the edematous children showed a positive correlation with aldosterone excretion ($r = 0.43$).

The increased excretion of NE found in the edematous nephrotic children declined toward normal within the first 48 hours of steroid therapy (Fig. 4). By the fourth day of steroid therapy, the mean excretion of catecholamines in all children had decreased (NE ↓ 41%; E ↓ 52%; M ↓ 14%;‡ VMA ↓ 14%). These changes occurred prior to cessation of proteinuria and in most subjects (5/7) were independent of creatinine clearance. As illustrated in Fig. 4, the decrease in NE was not associated with further sodium retention since renal sodium con-

‡ Measured in 2/7 subjects.

TABLE II. Comparison of Norepinephrine Excretion at Similar Levels of Protein Excretion Before and During Steroid Therapy.

	Before steroid therapy		During steroid therapy		
Patient	Protein excretion (g/24 hr)	NE excretion (μg/g protein/24 hr)	Protein excretion (g/24 hr)	NE excretion (μg/g protein/24 hr)	Ratio*
1	3.03	4.29	3.03	3.23	1.3
2	10.87	2.0	11.83	.44	4.5
3	9.4	4.9	9.20	2.7	1.8
4	20.2	1.7	22.30	1.2	1.4
5	4.9	12.6	4.90	3.1	4.1
6	4.87	4.3	4.96	2.8	1.5
7	5.26	11.9	4.14	12.8	.9

* NE excretion before steroid therapy/NE excretion during steroid therapy.

servation was maximum and sodium intake was constant. Similar data were obtained in 2 other subjects who were excreting less than 1 mEq of sodium per day.

Discussion. These data demonstrate a significantly greater excretion of NE in edematous nephrotic children compared to these same children in remission or to normal children. The increased excretion could result from 1) increased renal clearance of the hormone in presence of normal blood levels, 2) decreased rate of degradation of the hormone or 3) increased release of the hormone. Our data indicate this increased excretion of NE was not associated with increased glomerular filtration rate or quantity of urine protein. The finding of simultaneous but less marked increases in excretion of VMA militates against altered degradation of NE accounting for this increased excretion. Although plasma levels were not obtained, other observations(6) suggest that increased excretion of NE reflects increased release of the hormone.

Increased release of NE has been associated with muscular work and change of posture(6). Activity was not restricted in our patients but was similar in nephrotic and control subjects. Acute contraction of blood volume causes increased release of NE(6). Although there are no reports of catecholamine response to prolonged contraction of plasma volume, it is reasonable to suspect that chronic hypovolemia could provide a prolonged stimulus to the adrenergic nervous system. The increased excretion of catecholamines found in the edematous nephrotic children may reflect responses to regional or systemic hypovolemia. The prompt decrease in excretion of catecholamines and their metabolites, coincident to steroid therapy, makes an increase in blood volume secondary to increased circulating albumin an unlikely cause. These changes also cannot be attributed to steroid-induced salt retention with resultant expansion of plasma volume since 3 subjects on constant sodium intake were retaining sodium maximally prior to steroid therapy. The occurrence of steroid-induced shifts of sodium and water into the intravascular compartment(10) has not been excluded. It is also possible that these changes in catecholamine excretion reflect some other event unrelated to volume change.

Summary. In 7 edematous nephrotic children, the mean excretion of norepinephrine was significantly greater than in these children during remission or in 7 control subjects. The increased excretion did not correlate with glomerular filtration rate or quantity of urine protein but correlated with magnitude of sodium retention. Steroid therapy caused a prompt decrease of excretion of catecholamines and their metabolites. These findings may relate to circulatory changes or to other unrecognized abnormalities.

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Effects of Neonatal Thymectomy on Spontaneous Murine Autoimmune Disease.* (32307)

FUJIHIKO MIYASATO, JOSÉ MANALIGOD, GIUSEPPE G. PIETRA, AND VICTOR E. POLLAK (Introduced by Conrad L. Pirani)

Departments of Medicine and Pathology, Michael Reese Hospital, Research and Educational Hospital, Presbyterian-St. Luke's Hospital, and University of Illinois College of Medicine, Chicago

The possible pathogenetic role of the thymus in autoimmune disease has been the subject of much recent investigation. Tumors and other thymic abnormalities occur in patients with dysgammaglobulinemia; thymic epithelial hyperplasia and/or germinal center formation are described in systemic lupus erythematosus (SLE) and myasthenia gravis (1).

Atypical epithelial hyperplasia and structures resembling germinal centers were described by Holmes and Burnet in the thymus of NZB mice with autoimmune hemolytic anemia and nephritis(2). These authors considered the changes to be representative of the development of "forbidden clones" which initiate autoimmune reactions. As neonatal thymectomy is known to impair the full development of immunologic competence in many animal species, it was logical to attempt to determine the effect of neonatal thymectomy in murine strains with autoimmune disease. On the one hand, Helyer and Howie reported the earlier appearance of auto antibodies and more florid renal disease in neonatally thymectomized animals(3,4); whereas Holmes and Burnet noted that the appearance of Coombs antibodies was delayed 2 to 3 months in NZB mice thymectomized 2 to 6 days after birth(5,6). We report herein on the effect of neonatal thymectomy in two F_1 hybrid strains, NZB-NZW and NZB-NZC.† Nephritis is the major manifestation of the disease in the former, hemolytic anemia in the latter(7).

Material and methods. Thymectomies were done within 24 hours of birth by the technique of Helyer & Howie(3). Two groups served as controls: nonthymectomized and sham thymectomized animals. In the latter the thymus was dissected but left in place. Mice from newborn litters were assigned at random to the 3 groups.

Studies were made on mice which survived to weaning: 24 neonatally thymectomized (Tx) NZB-NZW F_1 mice and 25 controls (11 sham operated; 14 unoperated); 21 Tx NZB-NZC F_1 mice and 17 sham operated and unoperated controls. As there were no differences in the 2 control groups of each strain they are considered together. All mice were inspected and weighed weekly; blood was drawn biweekly for hematocrit, Coombs tests, antinuclear antibodies and LE cells(8). Animals were sacrificed when ill. A complete autopsy was done in 66 mice; completeness of thymectomy was confirmed by gross inspection and histologic examination in all but 2 NZB-NZC mice.

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