Neonatal uninephrectomy causes hypertension in adult rats

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Woods, Lori L. Neonatal uninephrectomy causes hypertension in adult rats. Am. J. Physiol. 276 (Regulatory Integrative Comp. Physiol. 45): R974–R978, 1999.—This study was designed to test the hypothesis that a reduced number of nephrons from birth leads to increased arterial pressure in adulthood. Newborn Sprague-Dawley rat pups were uninephrectomized during the first 24 h after birth. In chronically instrumented adult animals (22 wk), mean arterial pressure on a normal (0.20%)-Na⁺ diet was higher in uninephrectomized rats (133 ± 2 mmHg vs. 121 ± 2 mmHg in controls, P < 0.0001). Body weights were not significantly different, but the total kidney-to-body weight ratio was significantly reduced by 14% in adult uninephrectomized animals (P < 0.05). Glomerular filtration rate was reduced by ~30% in uninephrectomized rats (1.84 ± 0.09 vs. 2.63 ± 0.14 ml/min, P < 0.0002), and effective renal plasma flow was reduced to a lesser degree (6.37 ± 0.38 vs. 7.87 ± 0.51 ml/min, P < 0.03), such that the filtration fraction was also reduced (0.291 ± 0.007 vs. 0.338 ± 0.014, P < 0.01). After 7–10 days on a high (3.15%)-Na⁺ diet, arterial pressure increased more in uninephrectomized animals than in controls (20 ± 3 vs. 1 ± 1 mmHg, P < 0.003). Thus surgical removal of 50% of the nephrons, when done during development, caused reduced renal function and a salt-sensitive hypertension in adulthood. These data suggest that a reduced nephron endowment from birth, caused by genetic and/or perinatal environmental factors, could contribute to essential hypertension in adulthood.

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least three occasions to acclimatize them to the study conditions.

Experimental protocol. On the days on which physiological measurements were made, the rat was placed in a wire restrainer and urine was allowed to drain continuously through the bladder catheter into a tube. Mean arterial pressure was measured through the arterial catheter using a pressure transducer (Statham, Oxnard, CA) connected to a polygraph (Grass Instruments, Quincy, MA), and a reading was taken after at least 30 min, once the pressure had stabilized. All blood pressure measurements were made between 6:00 and 9:00 AM. An arterial blood sample was taken for measurement of microhematocrit, plasma protein, and plasma renin activity (PRA). The sample for PRA was placed on ice in a tube containing sodium EDTA and centrifuged at 4°C, and the plasma was frozen at −20°C. Inulin (Sigma, St. Louis, MO) and p-aminohippurate (PAH) (Sigma) in 5% dextrose were given intravenously as a bolus (0.45 ml containing 56 mg inulin and 5.6 mg PAH) followed by a continuous infusion (0.024 ml/min of 74 mg/ml inulin and 7.4 mg/ml PAH) throughout the rest of the experiment. At least 60 min after the beginning of the inulin/PAH infusion, three or four successive 20-min urine collections (clearance periods) were done, with a blood sample taken at the midpoint of each. Blood was collected in sterile heparinized syringes. Urine was allowed to drain continuously during the first 24 h of postnatal life and the arterial pressure measurements were repeated at 7 and 10–11 days. Mean arterial pressure was significantly increased after neonatal uninephrectomy (P < 0.0001). Absolute GFR (P < 0.0002) and ERPF (P < 0.03) were reduced in UNX animals, as were GFR and ERPF normalized to body weight. GFR normalized to kidney weight was significantly reduced; however, the reduction in ERPF normalized to kidney weight did not reach statistical significance. Filtration fraction was also significantly reduced (Table 1).

There was no significant difference in body weights of control or UNX rats on the normal diet compared with weights after 7–10 days on the high-Na+ diet (change of 0 ± 3 g). In control animals, arterial pressure did not increase significantly (a change of 1 ± 1 mmHg) on the high-Na+ diet (Fig. 2). By comparison, in UNX animals, arterial pressure increased by 20 ± 3 mmHg during the high-Na+ diet (P < 0.005), a response that was significantly greater than that in the control animals (P < 0.003). Thus the hypertension in UNX animals was salt sensitive.

PRA in control animals was 4.1 ± 0.8 ng ANG I·ml
-1·h
-1 on the normal-Na+ diet and was suppressed to 1.6 ± 0.5 ng ANG I·ml
-1·h
-1 on the high-Na+ diet (P < 0.02). Similarly, in UNX animals, PRA was 4.2 ± 0.9 ng ANG I·ml
-1·h
-1 on the normal-Na+ diet and was suppressed to 0.3 ± 0.2 ng ANG I·ml
-1·h
-1 on the high-Na+ diet (P < 0.02). The baseline values and the

Table 1. Body weights and renal hemodynamic variables in control rats and rats uninephrectomized during the first 24 h of postnatal life

<table>
<thead>
<tr>
<th>Variable</th>
<th>Control (n = 9)</th>
<th>Uninephrectomized (n = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body wt at weaning, g</td>
<td>63 ± 4</td>
<td>60 ± 3</td>
</tr>
<tr>
<td>Body wt at study, g</td>
<td>269 ± 7</td>
<td>282 ± 8</td>
</tr>
<tr>
<td>Total kidney wt, g</td>
<td>1.750 ± 0.077</td>
<td>1.646 ± 0.073</td>
</tr>
<tr>
<td>Filtration fraction</td>
<td>0.338 ± 0.014</td>
<td>0.291 ± 0.007*</td>
</tr>
<tr>
<td>Kidney-to-body wt ratio, %</td>
<td>0.694 ± 0.026</td>
<td>0.596 ± 0.028*</td>
</tr>
</tbody>
</table>
| GFR/kidney wt, ml·min
-1·g
-1               | 1.51 ± 0.08    | 1.13 ± 0.06*               |
| GFR/body wt, ml·min
-1·100 g
-1           | 4.52 ± 0.28    | 3.93 ± 0.25                |
| GFR/kidney wt, ml·min
-1·100 g
-1          | 0.973 ± 0.034  | 0.652 ± 0.029*             |
| ERPF/kidney wt, ml·min
-1·100 g
-1         | 2.910 ± 0.128  | 2.261 ± 0.126*             |

Values are means ± SE. Total kidney weight represents combined weight of both kidneys in control animals and weight of single remaining kidney in uninephrectomized animals. GFR, glomerular filtration rate; ERPF, effective renal plasma flow. *P < 0.05 or better compared with controls.
response to the high-Na\textsuperscript{+} diet were not significantly different in the two groups of animals.

Histopathological analysis showed no significant glomerular lesions in either group, although the average glomerular size appeared larger in the UNX animals. UNX rats also showed focal areas of cortical tubular collapse, associated chronic inflammation, and local tubular dilatation. This was only a minor finding, affecting only small, widely spaced zones in an otherwise normal tubulointerstitial compartment.

DISCUSSION

The most important finding of this study is that a surgical reduction in the number of nephrons by 50% in early postnatal development results in an increased arterial blood pressure in adulthood. These data provide the first direct evidence that a reduced endowment of nephrons from birth can lead to adult hypertension.

The effect on blood pressure of a 50% reduction in nephron number (uninephrectomy) in adult animals or humans remains controversial. In humans, some investigators have reported an increased prevalence of hypertension (13), whereas others found no significant change in blood pressures 10 or more years after uninephrectomy (25, 30, 32). Unfortunately, most human studies looking at this issue are retrospective, some lack appropriate controls, and often subjects were taking antihypertensive medication, making interpretation difficult. Thus it remains possible that the blood pressures of adult humans increase after nephrectomy without necessarily reaching the "hypertensive" range. Uninephrectomy in adult rats of several strains has been reported to have no significant effect on either mean arterial pressure or systolic pressure (6, 8, 22). Indeed, some (10) but not all (29) investigators have found that even a more extreme reduction in renal mass (5⁄6 nephrectomy) fails to cause hypertension if it does not involve infarction.

Somewhat in contrast to uninephrectomy in adulthood, there are at least suggestions in the literature that uninephrectomy relatively early in life may increase blood pressure (9, 21), although this is also controversial (16, 24). In the guinea pig, uninephrectomy within the first 36 h after birth resulted in an increased arterial pressure at 10 days of age compared with sham-operated animals (5).

In the present study, it was found that adult mean arterial pressures were significantly increased in rats UNX within the first 24 h of postnatal life. An important distinction between the present study and previous work is the stage of development at which renal mass was reduced. In the present work, uninephrectomy was performed midway through the period of nephrogenesis, which occurs from midgestation until 7–10 days after birth in the rat (18). In contrast, in humans and guinea pigs, nephrogenesis is completed before birth (7). Thus it seems likely that reducing the number of nephrons during the developmental period, when physiological control mechanisms are still plastic, may reprogram the future blood pressure set point in a way that does not occur if the reduction takes place later in life.

There are several possible mechanisms that could be responsible for the increased arterial pressure in the UNX animals in this study. In general, hypertension occurs because of the inability of the kidney to maintain sodium and water balance at a normal arterial pressure. This inability can be due to elevated levels of hormonal factors such as angiotensin or vasopressin as well as to a reduced glomerular filtration coefficient.
rons have increased their filtration rate by
remaining glomeruli were not sufficient to completely
the compensatory changes in the capillaries of the
in UNX animals compared with controls suggests that
were not obtained, the fact that total GFR was reduced
sufficiently to make up for the number of nephrons lost.
A reduced glomerular capillary filtration surface area
could also have contributed to the need for an
increased arterial pressure to maintain fluid and elec-
trolyte balance in the UNX animals in this study. Initially,
the total filtration surface area must have been reduced by half, because 50% of the glomeruli
were removed by uninephrectomy. However, by the
time of study in adulthood, the size of the individual
remaining glomeruli appeared to be increased in the
UNX animals in this study. The phenomenon of compen-
satory hypertrophy of remaining nephrons after a
reduction in nephron number is well established in
animals in which nephrons are lost in adulthood (10). A
reduced nephron (glomerular) number would be ex-
pected to lead to hypertension unless the capillary
surface area and/or the hydraulic permeability of the
capillary walls in the remaining glomeruli increased
sufficiently to make up for the number of nephrons lost.
Although direct measurements of these parameters
were not obtained, the fact that total GFR was reduced
in UNX animals compared with controls suggests that
the compensatory changes in the capillaries of the
remaining glomeruli were not sufficient to completely
make up for the lost nephrons.
Finally, it was documented that the remaining neph-
rons have increased their filtration rate by ~35%,
assuming that their number remains constant postne-
phrectomy. This hyperfiltration and a possible accompa-
nying increase in glomerular hydrostatic pressure in
the remaining nephrons could have contributed, over
the long term, to glomerular injury and progressive
nephron loss, thus further increasing the arterial pres-
sure needed to maintain sodium and water balance.
Indeed, uninephrectomy in adult rats causes the re-
maind nephrons to increase their individual filtration
rates by ~50% on average (6, 8, 22). If the same was
true initially in the animals in this study, this could
then have been followed by a gradual decrease associ-
ated with progressive glomerular damage due to life-
long hypertension. However, the histopathological
evidence does not seem to support this idea, because UNX
animals did not appear to have more glomerular dam-
age than controls. The fact that urine protein excre-
tions were not different in UNX animals compared with
controls also argues against the presence of major
glomerular damage in UNX animals. On the other
hand, the possibility cannot be ruled out that a more
comprehensive histopathological analysis or study of
the animals at an older age could reveal more subtle
glomerular damage that was not seen. Of note, male
rats UNX at 10 days of age have been reported to show
significant glomerular damage and proteinuria by 12
wk after surgery (23). However, the animals in the
present study were female, and female rats in general
tend to be more resistant to developing renal disease
and hypertension than males.
Classically, renal function curves have been used to
analyze arterial pressure regulation in altered func-
tional or pathological states of the kidneys (12). The
salt sensitivity of the hypertension in the UNX animals
in this study, illustrated by the slope of the renal
function curve in Fig. 2, would also be consistent with
at least two different underlying hypertensive mecha-
isms: reduced renal mass per se and inappropriately
increased levels of ANG II or aldosterone (12). In the
adult, surgical reduction of renal mass is known to lead
to a salt-sensitive type of hypertension, and because it
is known that renal mass was reduced in the UNX
animals in this study (at least initially), it is likely that
this factor itself contributes to the salt sensitivity of
their hypertension. On the other hand, a high Na+
intake reduced PRA in UNX animals to a similar extent
as in controls. This suggests that the salt sensitivity of
their hypertension is not due to an inadequate suppres-
sion of the activity of the renin-angiotensin system and
thus physiologically inappropriate high ANG II levels
under these conditions.

Perspectives
The results of the present study may have important
implications for the etiology of human hypertension.
Accumulating evidence in the epidemiological litera-
ture indicates that babies that are born smaller or grow
more slowly during the first year of life have an
increased incidence of hypertension and death from
cardiovascular disease when they reach adulthood (2, 19).
This suggests that some factor(s) in the perinatal
environment, probably related to maternal nutrition,
can "program" the individual for increased cardiovascu-
lar risk later in life. Recent evidence in the rat indicates
that maternal dietary protein restriction during preg-
nancy leads to hypertension in adult offspring (17),
suggesting that the protein content of the maternal diet
may provide one important link between the epidemi-
ological findings. Moreover, perinatal exposure to matern-
al protein restriction is known to result in a reduced
number of nephrons in the offspring (33). The results of
the present studies, in which the number of nephrons
was surgically reduced during the perinatal period,
provide strong evidence that a reduced number of
nephrons from birth per se could account for the
hypertension seen in the offspring of protein-restricted
mothers. It is speculated that the protein content of the
mothers' diets and consequent differences in the num-er of nephrons with which their babies are endowed
may provide an important link in the association
between early growth rates and later cardiovascular
risk reported in human studies.
REFERENCES


