Renal function and morphometry in the dwarf rat following a reduction in renal mass

J. Haylor, J. Chowdry, H. Baillie, G. Cope and A. M. El Nahas
Sheffield Kidney Institute, Northern General Hospital, Department of Medicine and Pharmacology, Royal Hallamshire Hospital, Department of Biomedical Science, Sheffield University, Sheffield, UK

Abstract
Background. The compensatory increase in glomerular filtration rate (GFR) and effective renal plasma flow (ERPF) which follows a reduction in renal mass may be mediated by growth hormone, a renal vasodilator.

Methods. GFR, ERPF and glomerular morphometry were assessed in the dwarf rat, selectively deficient in GH, and compared its Lewis base strain. Studies were performed 21-days after sham-operation, unilateral nephrectomy or subtotal nephrectomy in age-matched animals. GFR and ERPF were assessed from the renal clearance of inulin and p-aminohippurate measured under barbiturate anaesthesia.

Results. The dwarf rat had a lower GFR and ERPF than the Lewis rat, in proportion to its lower body weight and lower kidney weight. Kidneys from the dwarf rat had a similar number of glomeruli to the Lewis, but smaller glomerular components in proportion to a lower kidney weight. Following unilateral nephrectomy, GFR (dwarf + 58%, Lewis + 53%) and ERPF (dwarf + 58%, Lewis + 52%) increased to a similar degree in both rat strains. Glomerular diameter, volume and capillary surface area increased in proportion to kidney growth, although compensatory renal growth (dwarf + 62%, Lewis + 78%) was somewhat lower in the dwarf. Following 5/6 subtotal nephrectomy, GFR (dwarf + 143%, Lewis + 171%) increased to a similar degree in both rat strains while ERPF (dwarf + 115%, Lewis 86%) were greater in the dwarf than the Lewis rat. Subtotal nephrectomy was also associated with an increase in the thickness of the glomerular basement membrane in both rat strains.

Conclusions. The results do not support a role for GH in the compensatory increase in renal function or hypertrophy which follows a reduction in renal mass, excluding this as a potential mechanism for GH-dependent renal scarring.

Key words: kidney; growth hormone; dwarf rat; glomerular filtration rate; renal plasma flow; compensatory renal growth.

Introduction
Recent experiments in man correlated glomerular hyperfiltration in insulin-dependent diabetes mellitus with the enhanced secretion of growth hormone [1]. The ability of growth hormone (GH) to increase the glomerular filtration rate (GFR) and effective renal plasma flow (ERPF) was first described in the dog in 1949 [2]. The effect of GH on renal function is thought to be mediated by an increase in plasma insulin-like growth factor-I (IGF-I) [3] and has been dissociated from its ability to stimulate growth [4,5]. A possible role for GH in the maintenance of renal function has been suggested from the fall in GFR and ERPF observed after the reduction of circulating GH levels following either hypophysectomy [6,7] or the administration of somatostatin [8], an inhibitor of GH release [9].

Studies in the rat indicate that the kidney plays a major role in the systemic clearance of GH [10]. Plasma GH is increased in chronic renal insufficiency [11] and may be elevated following a reduction in renal mass. The present experiments have been performed to determine the involvement of GH in the compensatory increase in GFR and ERPF which follow either unilateral nephrectomy (UNx) or subtotal nephrectomy (SNx) [12]. A novel approach was employed using the dwarf rat, a strain genetically, selectively deficient in GH [13] in comparison to the Lewis base-strain from which it was derived. Renal function and glomerular morphometry were assessed in both Lewis and dwarf rats at 10 weeks of age, 21 days after sham-operation, UNx or SNx. GFR and ERPF were assessed from the renal clearance of inulin and p-aminohippuric acid respectively. Because of the association between glomerular hyperfiltration, hypertrophy and progressive glomerular injury [14,15] any role established for GH in compensatory
hyperfiltration or growth may be an important causative factor in the development of renal scarring.

Methods

Male rats (Ola, UK) of the Lewis and dwarf strains obtained at 6 weeks of age were used in this study. The dwarf rat has only about 5% of the normal pituitary content of GH, low circulating levels with no GH surges [13]. They were housed 2–3 per cage at an ambient temperature of 17°C, humidity of 45% and light/dark cycles of 12 h and had free access to standard animal chow (casein content 18%) and tap water before and after surgery.

Nephrectomy

At 7 weeks of age sham-operation, UNx or SNx was undertaken under ether anaesthesia. For UNx, the right kidney was exposed through a flank incision, the adrenal gland was separated from the upper pole and the kidney decapsulated. The renal pedicle was ligated and the kidney removed. Sham operated rats underwent a similar flank incision under ether anaesthesia. For SNx, the left kidney was also exposed through a flank incision, the adrenal gland separated from the upper pole and the kidney decapsulated. Ligatures were then placed around the upper and lower poles of the remaining kidney and the poles removed. For SNx, sham-operated rats underwent flank incisions on both the left and right sides under ether anaesthesia without the manipulation of either kidney.

At the time of UNx/SNx and sacrifice, kidneys and their remnant fragments (Rem Kf) were removed and weighed to give the kidney wet weight (KW) and calculate the wet weight of the remnant kidney (Rem KW). Compensatory renal growth (CRG) was calculated for UNx using the formula [(left KW–right KW)/right KW] x 100 and for SNx using the formula [Rem KW/Right KW–Rem Kf] x 100. Kidney protein was measured according to the method of Lowry [16] and kidney DNA by the method of Kissane and Robins [17].

Renal clearance studies

At 10 weeks of age (21 days after nephrectomy), rats were anaesthetised with sodium thiopentone (125 mg/kg) and body temperature was maintained at 37°C. The trachea was cannulated to facilitate respiration. The left carotid artery was cannulated for the recording of blood pressure and collection of blood. The right jugular vein was cannulated for the infusion of fluids and the bladder cannulated for the collection of urine. At time 0, each rat received a bolus intravenous injection of inulin 104 mg and p-aminohippuric acid (PAH) 48 mg dissolved in 1 ml 0.9% saline followed by a continuous intravenous infusion of inulin (0.9%) and PAH (0.2%) in 0.7% saline delivered at 50 μl/min/100 g b.w. At 60, 90 and 120 min, the bladder was washed out with 3 ml of distilled water and an arterial blood sample (0.4 ml) collected. A terminal blood sample was taken at the end of each experiment and the kidneys were removed. Measurements of inulin clearance (CIn) and p-aminohippurate clearance (CPAH) obtained following UNx were compared to values obtained from sham-operated controls × 0.5. Measurements of CIn and CPAH obtained following SNx were compared to values obtained from sham-operated controls × the average fraction of total kidney mass left following SNx.

Inulin and PAH were measured in plasma and urine by modifications of the methods of Bojesen [18] and Bratton and Marshall [19] respectively following protein precipitation [20]. CIn and CPAH were calculated from the formula

\[ P = \frac{C_l}{U} \]

where \( P \) = plasma concentration (μg/ml), \( U \) = urinary concentration (μg/ml), \( V \) = urine flow rate (ml/min).

Sample preparation for morphometry

In a separate series of experiments, rats were anaesthesised (pentobarbitone 125 mg/kg) and the kidneys perfused (120–140 mmHg) with phosphate buffered saline (PBS, 150 ml) containing 0.1% sodium nitrite via an aortic cannula, followed by PBS containing 1% glutaraldehyde. Kidneys were removed, weighed, sliced (1 mm) and further fixed overnight at 4°C. For light microscopy, every third slice from a random start was rinsed with PBS, dehydrated, and embedded in JB4 resin. One complete 2 μm section was cut from the uppermost surface of each slice, stained with Toluidine blue and mounted with DPX. For electron microscopy, 1 mm3 of tissue was removed from the cortex/outer stripe of the outer medulla, using random co-ordinates [21], washed with PBS, refixed in 2% osmium tetroxide, dehydrated with graded alcoholic solutions, and embedded in araldite resin. Semi-thin sections were cut from six cubes of tissue randomly selected from each animal. From each section, the glomerulus lying closest to the centre and a glomerulus lying at least one maximal glomerular radius from the edge was identified and selected for ultramicroscopy [21]. 70 nm sections were mounted on collodion-coated G50 copper grids, stained with uranyl acetate and lead citrate and viewed using a Philips CM10 electron microscope.

Morphometric analysis

The number of glomeruli per kidney and their volume density were determined by point-count analysis. The diameter of glomerulus was determined using a digitising tablet and two orthogonal diameters per glomerular profile were measured for half the glomeruli per section (about 100 per kidney).

Mean true glomerular diameter was calculated assuming the glomeruli to be spherical. Glomerular numerical density (Nv) was calculated using the formula

\[ Nv = \frac{Na}{H} \]

where \( Nv \) = number of profiles per unit area tissue, \( H \) = mean glomerular diameter.

The volumes and surface areas of glomerular components were estimated from overlapping micrographs of three randomly sampled glomeruli per animal taken under EM (×1200) and montages constructed of the prints. These were overlaid with a lattice (10.4 μm) and volume densities (Vv) obtained by point counting. Surface density (Sv) of glomerular capillaries was determined by line cut analysis. Vv and Sv estimates were converted to absolute mean volume and area values using the mean glomerular volume determined above.

The thickness of the glomerular basement membrane (GBM) was measured on a separate systematically sampled set of photomicrographs taken from three glomeruli per animal (× 24 500). GBM thickness was calculated as the harmonic mean membrane thickness (Th) using the orthogonal intercept method where \( Th = \frac{X}{S} \times 10^{magnification \times 1 \times h} \); h being the apparent harmonic mean thickness [23,24].
Renal hyperfiltration in the dwarf rat

Statistics. Clearance data was expressed as mean ± SEM and compared using the non-paired Students t-test using Minitab software. P<0.05 was considered significant. For glomerular morphometry, an ANOVA routine was performed on SPSS. P<0.05 was also considered significant.

Results

Basal renal function

The difference in basal kidney function between the sham-operated Lewis rat (n=13) and the dwarf rat (n=17) measured at sacrifice (10 weeks of age) is shown in Table 1. The dwarf rat had a significantly lower (P<0.001) body weight, lower total kidney wet weight, lower total kidney protein and lower total kidney DNA. Renal function studies showed the dwarf rat to have a significantly lower \( C_{\text{In}} \) (1.60 ± 0.10 vs. 2.26 ± 0.12 ml/min, \( P<0.001 \)) and \( C_{\text{PAH}} \) (3.53 ± 0.24 ml/min, \( P<0.001 \)) compared to the sham-operated control rat.

Table 1. Comparison of renal function and renal tissue content between sham-operated Lewis and dwarf rats measured at 10 weeks of age

<table>
<thead>
<tr>
<th>Number</th>
<th>Lewis</th>
<th>Dwarf</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight (g)</td>
<td>289 ± 11</td>
<td>165 ± 4*</td>
</tr>
<tr>
<td>Inulin clearance (ml/min)</td>
<td>2.26 ± 0.12</td>
<td>1.60 ± 0.10*</td>
</tr>
<tr>
<td>PAH clearance (ml/min)</td>
<td>6.31 ± 0.46</td>
<td>3.53 ± 0.24*</td>
</tr>
<tr>
<td>Blood pressure (mmHg)</td>
<td>106 ± 5</td>
<td>137 ± 3*</td>
</tr>
<tr>
<td>Haematocrit</td>
<td>41.3 ± 0.8</td>
<td>43.1 ± 0.8</td>
</tr>
<tr>
<td>Filtration Fraction</td>
<td>0.37 ± 0.03</td>
<td>0.46 ± 0.02*</td>
</tr>
<tr>
<td>Total kidney wet weight (g)</td>
<td>2.27 ± 0.08</td>
<td>1.22 ± 0.03*</td>
</tr>
<tr>
<td>Total kidney protein (mg)</td>
<td>224 ± 14</td>
<td>146 ± 8.5*</td>
</tr>
<tr>
<td>Total kidney DNA (mg)</td>
<td>8.67 ± 0.39</td>
<td>6.24 ± 0.34*</td>
</tr>
</tbody>
</table>

Results are expressed as mean ± SEM.* \( P<0.01 \).

Renal function following 21 day UNx

Following UNx, \( C_{\text{In}} \) was significantly higher in both the dwarf rat (1.18 ± 0.08 versus 0.76 ± 0.06 ml/min, \( P<0.001 \)) and the Lewis rat (1.79 ± 0.09 versus 1.17 ± 0.05 ml/min, \( P<0.001 \)) compared to sham-operated controls (Figure 1). Following UNx, \( C_{\text{PAH}} \) was also significantly higher for both the dwarf rat (2.84 ± 0.10 versus 1.75 ± 0.16 ml/min, \( P<0.005 \)) and the Lewis rat (5.00 ± 0.21 versus 3.30 ± 0.34 ml/min, \( P<0.005 \)) when compared to sham-operated controls (Figure 2). However as shown in Figure 3, following UNx, no significant difference was observed between the dwarf and Lewis rat for either renal hyperfiltration (dwarf: mean increase 58%, Lewis: mean increase 53%) or renal hyperperfusion (dwarf: mean increase 62%, Lewis: mean increase 52%).

The dwarf rat (sham-operated, 139 ± 4 mmHg) had a significantly higher blood pressure than the Lewis rat (sham-operated, 106 ± 7 mmHg, \( P<0.01 \)). This difference in systemic blood pressure was maintained following UNx (dwarf, 135 ± 4 mmHg; Lewis, 97 ± 7 mmHg, \( P<0.01 \)). Therefore UNx had no effect on systemic blood pressure in either rat strain.

Renal function following 21 day SNx

Following SNx, \( C_{\text{In}} \) was significantly higher in both the dwarf rat (0.81 ± 0.10 versus 0.39 ± 0.05 ml/min, \( P<0.005 \)) and the Lewis rat (1.64 ± 0.21 versus 0.67 ± 0.02 ml/min, \( P<0.01 \)) compared to sham-operated controls (Figure 1). Following SNx, \( C_{\text{PAH}} \) in

\[ 645 \quad \text{versus} \quad 6.31 ± 0.46 \text{ml/min} \quad P<0.001 \]
the remnant kidney was also significantly higher in both the dwarf rat (2.31 ± 0.30 versus 1.09 ± 0.10 ml/min, \( P<0.05 \)) and the Lewis rat (3.97 ± 0.20 versus 2.69 ± 0.20 ml/min, \( P<0.005 \)) when compared to sham-operated controls (Figure 2). However, as shown in Figure 3, following SNx, renal hyperfiltration was not significantly lower in the dwarf compared to the Lewis rat (dwarf: mean increase 143%, Lewis: mean increase 171%) while, renal hyperperfusion in the dwarf rat was significantly higher than in the Lewis rat (dwarf: mean increase 108%, Lewis: mean increase 48%, \( P<0.05 \)).

The dwarf rat (sham-operated, 133 ± 5 mmHg) had a significantly higher mean arterial blood pressure than the Lewis rat (sham-operated 105 ± 8 mmHg, \( P<0.01 \)). This difference in systemic blood pressure was maintained following SNx (dwarf 125 ± 10 mmHg, Lewis 109 ± 7 mmHg \( P<0.05 \)). Therefore SNx, like UNx, had no effect on systemic blood pressure in either rat strain.

Renal growth following 21-day UNx and SNx

Measurements of compensatory renal growth, total kidney DNA and protein are shown in Table 2. Following UNx, compensatory renal growth was significantly lower in the dwarf (56 ± 8%) than in the Lewis (78 ± 7%) rat \( (P<0.05) \). The dwarf rat also had a lower increase in total kidney protein but no difference in the increase in kidney DNA was observed between the two rat strains. Following SNx, the mean compensatory renal growth was higher (not lower) in the dwarf

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**Fig. 2.** \( p \)-Aminohippurate (PAH) clearance in the Lewis and dwarf rat following either 21-day unilateral nephrectomy (UNx) or 21-day subtotal nephrectomy (SNx). Data is presented as paired histograms, for measurements obtained following nephrectomy (right, hatched) and sham-operation (left, unhatched) derived from an equivalent kidney wet weight at surgery. Vertical bars indicate SEM.

**Fig. 3.** Increase in insulin clearance (A) and \( p \)-aminohippurate (PAH) clearance (B) in the Lewis (unhatched) and dwarf (hatched) rat following 21-day unilateral nephrectomy (UNx) or subtotal nephrectomy (SNx). Vertical bars indicate SEM.
Table 2. Compensatory renal growth (CRG) and total kidney protein and DNA in the dwarf and Lewis rat following either UNx or SNx

<table>
<thead>
<tr>
<th>Remnant kidney</th>
<th>Unilateral nephrectomy (UNx)</th>
<th>Subtotal nephrectomy (SNx)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Wet weight (g)</td>
<td>Total protein (mg)</td>
</tr>
<tr>
<td></td>
<td>Day 0</td>
<td>Day 21</td>
</tr>
<tr>
<td>Lewis Sham UNx</td>
<td>1.00 ± 0.02</td>
<td>0.96 ± 0.02</td>
</tr>
<tr>
<td>Lewis UNx</td>
<td>0.59 ± 0.02</td>
<td>0.57 ± 0.03</td>
</tr>
<tr>
<td>Dwarf Sham UNx</td>
<td>96 ± 1</td>
<td>178 ± 7</td>
</tr>
<tr>
<td>Dwarf UNx</td>
<td>98 ± 1</td>
<td>156 ± 8</td>
</tr>
<tr>
<td></td>
<td>CRG (%)</td>
<td>Total protein (mg)</td>
</tr>
<tr>
<td></td>
<td>Day 0</td>
<td>Day 21</td>
</tr>
<tr>
<td>Lewis Sham UNx</td>
<td>135 ± 18</td>
<td>124 ± 24</td>
</tr>
<tr>
<td>Lewis UNx</td>
<td>113 ± 9</td>
<td>77 ± 5</td>
</tr>
<tr>
<td>Dwarf Sham UNx</td>
<td>191 ± 18*</td>
<td>73 ± 6</td>
</tr>
<tr>
<td>Dwarf UNx</td>
<td>199 ± 11</td>
<td>82 ± 7*</td>
</tr>
<tr>
<td></td>
<td>Total DNA (mg)</td>
<td>Day 0</td>
</tr>
<tr>
<td>Lewis Sham UNx</td>
<td>3.60 ± 0.4</td>
<td>2.48 ± 0.1</td>
</tr>
<tr>
<td>Lewis UNx</td>
<td>4.8 ± 0.6</td>
<td>2.89 ± 0.3</td>
</tr>
<tr>
<td>Dwarf Sham UNx</td>
<td>3.81 ± 0.5</td>
<td>2.72 ± 0.4</td>
</tr>
<tr>
<td>Dwarf UNx</td>
<td>5.33 ± 0.6*</td>
<td>3.50 ± 0.2*</td>
</tr>
</tbody>
</table>

Values at day 0 were for (a) UNx, from the left kidney removed at day 0 and for (b) SNx, from renal tissue removed at day 0 subtracted from the total kidney mass. Results are expressed as mean ± SEM. *P<0.01.

(115 ± 29%) than in the Lewis (87 ± 6%) rat although, the difference did not reach significance. The change in renal growth following UNx and SNx for the dwarf and Lewis rat is compared in Figure 4.

Renal morphometry following 21-day UNx and SNx

Despite its lower body weight and kidney weight, the kidney from the dwarf rat contained a similar number of glomeruli (45,000 to 48,000) compared to kidneys from the Lewis rat, with no difference in the glomerular basement membrane thickness. Glomeruli from the dwarf rat had a significantly smaller glomerular volume, diameter and capillary surface area, but not tissue tuft volume in comparison to the Lewis (P<0.05). A comparison of the morphometric analysis for aged-matched dwarf and Lewis rats following 21-day sham-operation, UNx and SNx is shown in Tables 3 and 4.

UNx was not associated with an increase in GBM thickness in either rat strain. Following UNx, a significant increase in glomerular capillary surface area, glomerular diameter and volume, urinary space and tissue tuft volumes was detected for kidneys obtained from both the dwarf and Lewis rat. However there were no significant differences in the extent of such changes between the two rat strains. Two differences in response to UNx which could be detected between kidneys from dwarf and Lewis rat were in compensatory renal growth and capillary volume. The dwarf rat had a smaller increase in kidney wet weight (confirming data from the clearance study) and showed no significant increase in capillary volume following UNx, unlike the Lewis rat.
SNx, unlike UNx, was associated with an increase in GBM thickness in both rat strains. Following SNx, the Lewis rat showed similar glomerular changes to UNx including a significant increase in glomerular diameter, volume, volumes of glomerular components and capillary surface area. Following SNx, the dwarf also showed an increase in glomerular diameter and volume, though the increase in urinary space and capillary volume was not accompanied by a parallel increase in tuft tissue volume or capillary surface area. However, within the sample size employed, following SNx, no significant differences between the dwarf and Lewis rat following SNx could be established.

### Discussion

The ability of growth hormone (GH) to elevate GFR and RBF in both man [2,3] and the rat [25] is well documented and probably mediated by an increase in plasma IGF-I [3]. In the present experiments a novel approach, using the dwarf rat, was employed to examine the role of endogenous GH in the maintenance of renal function and its increase following a reduction in renal mass. The dwarf rat has a very low pituitary approach, using the dwarf rat, was employed to examine the decrease in GFR and RBF following either hypophysectomy [5,6], or the administration of somatostatin [7]. However, the decrease in GFR and RBF previously described to occur following somatostatin could also be produced by either a direct effect on the kidney [26] or an indirect effect mediated by systemic hormones from other endocrine glands such as glucagon [27]. Experiments in the rat using a receptor antagonist of GH releasing hormone indicate that GH may contribute to the tonic activation of GFR [28].

#### Table 3. Glomerular morphometry in age-matched Lewis and dwarf rats following 21-day sham-operation (Sham), unilateral nephrectomy (UNx) or subtotal nephrectomy (SNx)

<table>
<thead>
<tr>
<th></th>
<th>Lewis</th>
<th>UNx</th>
<th>SNx</th>
<th>Lewis</th>
<th>UNx</th>
<th>SNx</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight (g)</td>
<td>300±24</td>
<td>318±14</td>
<td>320±29</td>
<td>176±13*</td>
<td>174±15</td>
<td>173±21</td>
</tr>
<tr>
<td>Kidney wet weight (g)</td>
<td>1.55±0.11</td>
<td>2.18±0.14*</td>
<td>1.04±0.5*</td>
<td>0.86±0.13*</td>
<td>1.30±0.19§</td>
<td>0.60±0.11§</td>
</tr>
<tr>
<td>Glomeruli/kidney</td>
<td>45±150±7434</td>
<td>nd</td>
<td>nd</td>
<td>47710±5191*</td>
<td>nd</td>
<td>nd</td>
</tr>
<tr>
<td>Glomerular basement membrane thickness (nm)</td>
<td>110±8</td>
<td>110±12*</td>
<td>133±1†</td>
<td>109±10***</td>
<td>109±4***</td>
<td>119±1†</td>
</tr>
</tbody>
</table>

n=4 per group, *P<0.05 (ANOVA) versus Lewis Sham; †P<0.05 (ANOVA) versus Lewis sham; §P<0.05 (ANOVA) versus dwarf sham. nd = no data available.

#### Table 4. Morphometric analysis of individual glomeruli from age-matched Lewis and dwarf rats following 21-day sham-operation (Sham), unilateral nephrectomy (UNx) or subtotal nephrectomy (SNx)

<table>
<thead>
<tr>
<th></th>
<th>Lewis</th>
<th>UNx</th>
<th>SNx</th>
<th>Lewis</th>
<th>UNx</th>
<th>SNx</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diameter (μm)</td>
<td>120±9</td>
<td>144±4†</td>
<td>140±8*</td>
<td>104±12*</td>
<td>118±4§</td>
<td>110±3§</td>
</tr>
<tr>
<td>Volume (μm³ 10⁻⁵)</td>
<td>9.0±2.0</td>
<td>15.6±1.4†</td>
<td>14.7±2.8†</td>
<td>6.0±2.0*</td>
<td>8.6±0.9§</td>
<td>6.9±0.5§</td>
</tr>
<tr>
<td>Urinary space (μm³ 10⁻⁵)</td>
<td>3.4±1.0</td>
<td>7.1±1.5†</td>
<td>4.5±1.5†</td>
<td>2.3±1.8**</td>
<td>3.0±0.9§</td>
<td>3.0±0.3**</td>
</tr>
<tr>
<td>Capillary volume (μm³ 10⁻⁵)</td>
<td>2.0±0.1</td>
<td>3.6±0.7†</td>
<td>3.1±0.4†</td>
<td>1.3±0.1*</td>
<td>2.0±0.5**</td>
<td>2.1±0.3§</td>
</tr>
<tr>
<td>Tuft tissue volume (μm³ 10⁻⁵)</td>
<td>3.6±1.2</td>
<td>4.9±0.8†</td>
<td>7.1±1.2†</td>
<td>2.4±0.3**</td>
<td>3.6±0.7§</td>
<td>2.1±0.3**</td>
</tr>
<tr>
<td>Capillary surface area (μm²)</td>
<td>121±5</td>
<td>197±19†</td>
<td>189±12†</td>
<td>84±7*</td>
<td>113±8§</td>
<td>88±8**</td>
</tr>
</tbody>
</table>

n=4 per group. *P<0.05 (ANOVA) versus Lewis Sham; †P<0.05 (ANOVA) versus Lewis sham; §P<0.05 versus dwarf sham.
Renal hyperfiltration in the dwarf rat

Although, evidence from patients with GH deficiency suggests any additional role such as activating renal functional reserve to be unlikely [29]. However, the expression of GFR and ERPF per unit kidney weight or unit body weight may be misleading under conditions where GH could also exert a tonic influence on renal function. In addition, the possibility that the dwarf rat might develop an exaggerated response to GH through enhanced receptor binding or up-regulation cannot be entirely ignored.

The results of the present experiments do not provide any evidence of a role for GH in the compensatory increase in renal function which follows unilateral nephrectomy where, a similar increase in GFR and ERPF was obtained in both the dwarf and Lewis rat. Glomerular morphometry following UNx, showed a similar increase in glomerular volume/diameter and capillary surface area in both Lewis and dwarf rats, in proportion to their increased kidney size. UNx had no effect on glomerular basement membrane thickness in either rat strain. Evidence of a role for GH in compensatory hyperfiltration had previously been obtained in experiments involving hypophysectomy [30]. Our own pilot study using the dwarf rat also indicated a possible contribution of GH [31] while, a small contribution of GH to compensatory renal growth still remains a possibility. The hypothesis that GH contributes to compensatory hyperfiltration includes evidence that the kidney may be an important route for its systemic clearance. Isotopic studies demonstrate the renal clearance of rat GH [10,32,33], but not human GH [34], to be markedly reduced in the anephric but non-uroaemic rat. Plasma GH levels were not measured in the present experiments, but the higher level of GH which might be anticipated to follow SNx, rather than UNx, could then contribute to the compensatory renal function. However following SNx, hyperfiltration, hyperperfusion and CRG were, if anything, even greater in the dwarf than the Lewis rat. Glomeruli showed either similar morphometric changes following SNx, compared to UNx, in the Lewis rat with parallel changes being less well defined in the dwarf. An obvious difference seen in both rat strains caused by the more severe extent of renal ablation was an increase in glomerular basement membrane thickness. The marked increase in GFR which follows SNx (+150%), compared to the 50% following UNx was not therefore associated with any further increase in glomerular size. This data is consistent with the concept that the adaptive glomerular hyperfiltration of remnant nephrons may serve as a mechanism for renal growth, mediated perhaps by the local renal production of IGF-I [35].

The anaesthetised rat has been commonly employed in studies of hyperfiltration following a reduction in renal mass [36,37]. We were concerned however, that the smaller size of the dwarf rat could confer a greater susceptibility to changes in blood volume produced by inappropriate protocol's for blood sampling and fluid infusion. Such problems may explain the results of our own pilot study which suggested inhibition of hyperfiltration in the dwarf rat following UNx [31]. The dwarf rat did however have a higher mean blood pressure than the lewis rat during renal clearance experiments which remained unaffected by either the presence or the severity of the renal ablation. Similar high blood pressures have been recorded in the dwarf rat prepared for micropuncture [38] but were not observed in a conscious rat study [39]. Hypertension in the dwarf rat could have masked any attenuation of hyperfiltration (caused by a lack of GH) since renal autoregulation is also inhibited by the reduction in renal mass [40,41]. However, evidence for its inhibition in the dwarf rat has not as yet been obtained while in more recent studies, the inhibition of renal autoregulation seen following a reduction in renal mass was much less prominent [42,43].

The dwarf rat has a markedly reduced capacity to develop glomerulosclerosis following SNx [39]. Hypertrophy, hypertension and autoregulation are all involved in the concept that the adaptive glomerular hyperfiltration of remnant nephrons may serve as a common mediator for progressive glomerular injury, postulated from micropuncture studies in the rat remnant kidney model [14,15]. The results of the present experiments do not support a role for differences in either hypertrophy or hyperfiltration in explaining the lack of sensitivity of the dwarf rat to develop glomerulosclerosis following SNx. GH-dependent renal scarring, as seen in the transgenic mouse [44], might therefore result either from sodium retention increasing systemic blood pressure [45] or more probably from a direct effect of GH on renal matrix protein accumulation [46].

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References


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