Normal urinary protein composition in the female Wistar rat and its relationship to the proteinuria induced by intraperitoneal bovine albumin

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Summary

1. Normal female Wistar rats showed a wide range of baseline protein excretion and albuminuria and could be divided into two distinct groups: (a) low-to-intermediate excretors, where daily baseline albumin excretion was below 1.8 mg/24 h and (b) high excretors, where daily baseline albumin excretion was 1.8-22 mg/24 h.

2. In low-to-intermediate excretors 'sex-associated' urinary specific proteins, supposedly present only in male rat urine, were excreted in larger quantities than albumin which constituted only 10-15% of the total urinary protein.

3. In high excretors up to 50% of the urinary protein was albumin and other serum proteins were also present in relatively larger quantities than the urinary specific proteins.

4. Baseline albumin excretion appeared to be a good indicator of the inherent efficiency of the glomerular filter and this was reflected in the response to intraperitoneal injection of heterologous serum albumin since, during periods where no significant glomerular epithelial cell foot-process loss had been incurred, the levels of proteinuria induced correlated well with baseline albuminuria.

Key words: albuminuria, glomerulus, proteinuria, rat kidney, urine.

Introduction

Intraperitoneal albumin injection has been reported, in the rat, to result in the induction of high levels of proteinuria and extensive glomerular ultrastructural damage (Anderson & Recant, 1962; Kaplan & Drummond, 1972; Roy, Vernier & Michael, 1972; Brewer & Filip, 1976; Davies, Brewer & Hardwicke, 1978). Other workers (Ashworth & James, 1961; Fisher & Hellstrom, 1962; Kurtz & Feldman, 1962; Karl, Garcia, White, Recant & Kissane, 1964; Andrews, 1977) have failed to induce such marked effects after parenteral protein injection and, while this may to some extent be explained by differences in the strain and sex of rat injected and the albumin doses administered (G. M. Lawrence & D. B. Brewer, unpublished work), variable responses have been reported to occur within a single strain and sex of rat at a given albumin dose level (Kurtz & Feldman, 1962; Roy et al., 1972).

Normal baseline levels of proteinuria have been reported in a number of strains of rat of both sexes (Sellers, Goodmann, Marmorston & Smith, 1950; Deodhar, Cuppage & Gableman, 1964; Eisenbach, Van Liew & Boylan, 1975; Von Baeyer, Van Liew, Klassen & Boylan, 1976; Glasser, Velosa & Michael, 1977) and urinary protein composition investigated in some detail, particularly in the male (Dihn, Tremblay & Dufour, 1964; Roy & Neuhaus, 1966; Galaske, Van Liew & Feld, 1979). In these studies albumin was acknowledged as an important component of normal rat urine and it therefore seemed possible that inherent differences in glomerular permeability to albumin may influence the response to serum albumin overload.

The present study examines baseline total protein and albumin excretion and urinary protein composition in the female Wistar rat, and...
investigates the relationship between baseline albuminuria and the severity of the proteinuria induced by a range of intraperitoneally administered bovine albumin doses.

Methods

Animals

Female Wistar rats (Olac Ltd, 200 g body wt.), kept in individual stainless-steel metabolic cages on a modified rat/mouse breeding diet (Heygate and Sons Ltd) with water ad libitum were used throughout.

Induction of hyperalbuminaemic proteinuria

Rats (60) were injected intraperitoneally (i.p.) twice daily (at 09.30 and 17.30 hours) for 5 days with 2.5 ml of phosphate-buffered saline solution (NaCl 0.15 mol/l, sodium phosphate 0.01 mol/l; pH 7.2) containing 50–200 g of bovine albumin/l. Bovine albumin solutions were prepared by dissolving the appropriate quantity of Cohn V bovine albumin (Sigma Chemical Co.) directly into phosphate-buffered saline and the final concentrations determined from the absorbance at 280 nm ($A_{280}^\text{nm} = 6.67$ for bovine albumin (CRC Handbook of Biochemistry, 1970). Control rats (six) were injected with the same volume of phosphate-buffered saline alone. Urine samples (24 h) were collected for 3 days before and during the injection period. On day 6, rats were anaesthetized with 0.8 ml of 10% (v/v) pentobarbitone sodium/100 g body wt. (Sagatal; May and Baker Ltd) in 10% (v/v) ethanol, the kidneys removed and small sections of renal cortex processed for electron microscopy. Bovine albumin doses were calculated in terms of milligrams injected/gram body wt. on day 0 and the rats divided into dose groups 1.0–1.5, 1.5–2.0, 2.0–3.0, 3.0–3.5, 3.5–4.5, 4.5–5.5 mg day$^{-1}$ g$^{-1}$ body wt. for statistical analyses.

Electron microscopy and morphometry

Small fragments of renal cortex (0.5–1.0 mm$^3$) were immersion-fixed in ice-cold 2.5% (w/v) glutaraldehyde buffered with cacodylate (0.1 mmol/l)/HCl, pH 7.4, postfixed in Caulfield's solution (Caulfield, 1957), dehydrated in ethanol and embedded in Araldite. Sections (50–70 nm), stained with uranyl acetate and lead citrate, were examined with an AEI model EM 801 electron microscope operated at 80 kV.

Parameters determined in control rats

Baseline total protein excretion and albuminuria. Urine samples (24 h) were collected for 3 days and mean total protein and albumin content determined in 135 and 164 rats respectively.

Bovine albumin serum half-life. Rats (eight) were given a single i.p. injection containing
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FIG. 1. Serum bovine albumin (BSA) concentration after a single 1 ml i.p. injection containing 100 mg of BSA in phosphate-buffered saline.

100 mg of Cohn V bovine albumin in phosphate-buffered saline. Blood samples were obtained by tail-bleeding at 1, 2, 4, 6, 8 and 12 h, then at 12 h intervals for 3 days and at 24 h intervals for a further 2 days. Samples were collected in 1–2 mm bore capillary tubes, spun at 750 g and serum rat and bovine albumin concentrations determined.

Serum rat albumin concentrations remained constant throughout the 5 day period and maximum bovine albumin concentrations were attained 4 h after i.p. injection (Fig. 1). Bovine albumin serum half-life, obtained from a plot of log serum concentration against time, was 18.8 ± 1.6 h, a value similar to that obtained for human albumin (15.8 h) by Gaizutis, Pesce & Pollack (1975).

Method for determining the amount of bovine albumin transferred from the peritoneal cavity to the blood stream at different doses. Rats (eight) with known serum rat albumin concentrations, received single 1 ml i.p. injections containing either 50, 100, 150 or 175 mg of Cohn V bovine albumin in phosphate-buffered saline. Blood samples were taken by tail-bleeding 4 h later when maximum serum bovine albumin concentrations had been attained. No decrease in serum rat albumin concentration was apparent at this time therefore the normal serum volume for a 200 g rat (6.3 ml; Altman & Ditmer, 1961) was used to calculate the total amount of bovine albumin present in the serum at each dose level. When these values were plotted against the amount of bovine albumin injected (Fig. 2) the slope of the graph indicated that only 42% of the injected material could be accounted for when maximum serum bovine albumin levels were attained.

FIG. 2. Transfer of i.p. injected bovine albumin (BSA) from the peritoneal cavity to the blood stream at different dose levels.

Results

Urinary protein composition in the normal female Wistar rat

Total protein and baseline albumin excretion results were examined by Hoffmann's statistical method for determining normal ranges in clinical chemistry (Hoffmann, 1963); they each fell into three distinct groups containing low, intermediate or high values (Fig. 3). Ninety-three per cent of total protein excretion and 88% of baseline albumin excretion results lay in the low-to-intermediate ranges (1–12 and 0.1–1.8 mg/24 h total protein and albumin excretion respectively) and the remainder in the high range (12–56 and 1.8–22 mg/24 h total protein and albumin excretion respectively). In the low and intermediate groups albumin made up only 10–15% of the total urinary protein, whereas in the high group albumin constituted as much as 50% of the total protein excreted.

On immunoelectrophoresis (Fig. 4) samples from low-to-intermediate baseline albumin excretors (total daily excretion 0.33–0.98 mg/24 h) gave two precipitin lines against anti-whole rat serum corresponding to rat albumin and transferrin. Samples from high rat albumin excretors (total daily excretion 8–20 mg/24 h) gave seven precipitin lines against anti-whole rat serum of which five could be positively identified. The proteins present were, in decreasing order of intensity, rat albumin (1), transferrin (2), Gc-globulin (3), haptoglobin (4) and IgG (5). Fig. 4 also illustrates that urinary rat albumin had a faster electrophoretic mobility than serum rat albumin.

Cellulose acetate electrophoresis (Fig. 5a) confirmed the presence of albumin, γ-globulin and
proteins with $\alpha_1$ and $\beta$ mobilities in samples from high albumin excretors and the presence of albumin in samples from low-to-intermediate albumin excretors. In the latter, however, the major components migrated in the $\alpha_2-\beta$ region. As no components with this mobility were evident on immunoelectrophoresis with anti-(whole rat serum), those evident on cellulose acetate electrophoretic analysis were tentatively identified as urinary specific proteins.

Tube sodium dodecyl sulphate (SDS)/polyacrylamide-gel electrophoresis confirmed these findings and the results obtained are summarized in Table 1. Five urinary specific protein bands were detected in samples from low-to-intermediate albumin excretors, the major bands (mol. wt. 10 000, 18 500 and 48 000) probably corresponding to those proteins migrating in the $\alpha_2-\beta$ region on cellulose acetate electrophoresis. The three strongest urinary protein bands were also present in urine from high albumin excretors, but the relatively large amounts of serum protein also present reduced their proportions relative to total protein content. In some urine samples run on slab SDS/polyacrylamide gels a double albumin band was evident. The presence of a single or double albumin band correlated well with the nature of the band in corresponding serum samples, but not with high or low-to-intermediate albumin excretion.

**Variation of the proteinuria induced by intraperitoneal bovine albumin injection with baseline albumin excretion**

In rats with low-to-intermediate baseline albumin excretion the total protein excretion induced by 5 days i.p. injection with 1-0–1-5 or 1-5–2-0 mg of bovine albumin day$^{-1}$ g$^{-1}$ body wt. increased rapidly with rising baseline albumin excretion (Fig. 6). However, at baseline albumin excretion values above 1-0 mg/24 h, the relative increases in total protein excretion induced for an incremental rise in baseline albumin excretion fell as the latter increased until, at values above 2-0 mg/24 h, a limiting level of approximately 250 mg of total protein excretion appeared to be attained in the dose group 1-0–1-5 mg day$^{-1}$ g$^{-1}$ body wt. In the higher dose group (1-5–2-0 mg day$^{-1}$ g$^{-1}$
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Cellulose acetate strip length
Polyacrylamide running gel length

**FIG. 5.** (a) Cellulose acetate electrophoretic analysis: chromoscan traces showing the components present in urine samples from rats excreting low-to-intermediate (——) or high (———) levels of albumin. (b) SDS/polyacrylamide-gel electrophoretic analysis: characterization of urinary and serum components present in urine samples from rats excreting low-to-intermediate (——) or high (———) levels of albumin. Hpt = Haptoglobin; RSA = rat serum albumin; UP = urinary protein.

**TABLE 1.** Urinary and serum protein components in urine samples from rats excreting low-to-intermediate or high levels of albumin

<table>
<thead>
<tr>
<th>Albumin excretion</th>
<th>Serum proteins</th>
<th>Urinary proteins (UP)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low-to-intermediate</td>
<td>High</td>
<td>$R_p$ albumin</td>
</tr>
<tr>
<td>+ + + + +</td>
<td>0.05</td>
<td>175 000</td>
</tr>
<tr>
<td>+ + + +</td>
<td>0.18</td>
<td>155 000</td>
</tr>
<tr>
<td>+ + +</td>
<td>0.29</td>
<td>135 000</td>
</tr>
<tr>
<td>+ +</td>
<td>0.44</td>
<td>115 000</td>
</tr>
<tr>
<td>+ +</td>
<td>0.62</td>
<td>97 000</td>
</tr>
<tr>
<td>+ + + + + +</td>
<td>0.81</td>
<td>80 000</td>
</tr>
<tr>
<td>+ + + +</td>
<td>1.00</td>
<td>64 000</td>
</tr>
<tr>
<td>+ + +</td>
<td>1.16</td>
<td>55 000</td>
</tr>
<tr>
<td>+ + + + +</td>
<td>1.29</td>
<td>48 000</td>
</tr>
<tr>
<td>+ + + +</td>
<td>2.03</td>
<td>22 500</td>
</tr>
<tr>
<td>+ + + + +</td>
<td>2.10</td>
<td>18 500</td>
</tr>
<tr>
<td>+ + + + +</td>
<td>2.70</td>
<td>10 000</td>
</tr>
</tbody>
</table>

body wt.) the only rat with high baseline albumin excretion responded to bovine albumin injections by excreting 450 mg of total protein over the 5 day injection period.

When the total protein excretion induced during 5 days i.p. bovine albumin injection in these two low dose groups was plotted against baseline albumin excretion for rats with low-to-intermediate baseline albumin values, where no limiting levels of proteinuria were apparent (i.e. those rats with baseline albumin excretion below 1.8 mg/24 h), a good correlation was shown between the two variables ($r = 0.87; n = 18$) (Fig. 7). Albumin was the major constituent of the increased proteinuria measured in these rats and rat and bovine albumin were excreted in proportions approximately equivalent to their serum concentrations (G. M. Lawrence & D. B. Brewer, unpublished work). It therefore appeared that any increase in total serum albumin concentration above the baseline value caused a generalized increase in albumin filtration in which little if any distinction was made between rat and bovine albumin species and that the amount of albumin filtered, for a given rise in total serum albumin concentration, was controlled by the inherent glomerular permeability to albumin.

In high baseline albumin excretors the good correlation between baseline albumin excretion and induced proteinuria was no longer apparent.
and the inherent permeability characteristics of the glomerular filter no longer seemed to be the main influence on the amount of protein filtered, in response to a given low i.p. dose of bovine albumin. Since the rises in total serum albumin induced by these low bovine albumin doses would have been quite small (Fig. 2), it seemed possible that the amount of extra albumin available in the serum may have been the limiting factor which controlled the extent of the proteinuria induced in these rats with inherently inefficient glomerular filters. This hypothesis was supported by the following calculations.

Every day at 09.30 and 17.30 hours, each rat in bovine albumin dose groups 1.0–1.5 and 1.5–2.0 mg day⁻¹ g⁻¹ body wt. received a 2.5 ml i.p. injection containing 125 or 175 mg of bovine albumin respectively and 24 h urine collections were started immediately after the first of these injections. Measurements in control rats (Figs. 1, 2) indicated that the maximum serum bovine albumin levels resulting from each injection would have been attained 4 h after administration and that at this time only 42% of the bovine albumin injected would have been present in the blood stream. Results from control rats (Fig. 1) also indicated that not all of the bovine albumin reaching the serum would have been available for excretion during the remainder of the urinary collection period, as normal serum clearance mechanisms would have reduced serum bovine albumin concentrations in what appeared to be a simple exponential or first-order process. In such a process the relationship between the amount of bovine albumin present in the serum at 4 h, the start of the exponential decay period, i.e. 42% of the original amount injected (52.5 and 73.5 mg in dose groups 1.0–1.5 and 1.5–2.0 mg day⁻¹ g⁻¹ body wt. respectively) (No), and the amount present at any other time (N) is given by the equation:

\[ N = No e^{-\lambda t} \]  

where \( \lambda \) is the fraction of the injected material removed per unit time \( t \), during each urinary collection period that serum bovine albumin concentrations were reduced logarithmically by serum clearance mechanisms, i.e. the total length of the collection period less 4 h (20 and 12 h for the first and second bovine albumin injections respectively). This equation expressed in natural logarithmic form is as follows:

\[ \ln \frac{No}{N} = -\lambda t \]
Hyperalbuminaemia: factors affecting response

The serum half-life of bovine albumin \( t_1, 18.8 \pm 1.6 \text{ h} \) is defined as the period of time required to remove half of the bovine albumin present in the serum at any given time and it is related to \( x \) by the expression:

\[
\lambda = \frac{\ln 2}{t_1}
\]

Substituting this expression for \( x \) into eqn. (2) gives the following:

\[
\frac{N_0}{N} = \frac{\ln 2}{t_1}
\]

Transforming this equation from natural to common logarithms gives the following:

\[
\log \frac{N_0}{N} = \frac{\log 2}{t_1}
\]

i.e.,

\[
\log \frac{N_0}{N} = \frac{0.3010}{t_1}
\] (3)

Eqn. (3) can be used to calculate the amount of bovine albumin \( (N) \) remaining in the serum 24 h after the first i.p. injection for each injected dose at each dose level following the substitution of experimentally determined values for \( t, N_0 \) and \( t_1 \).

The amounts of extra albumin available in the serum for filtration during each 24 h urinary collection period calculated in this way were 25 + 34 = 59 mg in dose group 1.0-1.5 day\(^{-1}\) g\(^{-1}\) body wt. and 35 + 47 = 82 mg in dose group 1.5-2.0 mg day\(^{-1}\) g\(^{-1}\) body wt. The total amounts of extra albumin available over the 5 day bovine albumin injection period in each dose group were therefore 295 and 410 mg respectively. Both of these values were in reasonable agreement with the limiting protein excretion levels of 250 and 450 mg determined for the two dose groups from Fig. 6.

Obviously, such calculations must be used with care when estimating the levels of extra albumin present in the serum at various times after i.p. bovine albumin injection, as they rely on a number of basic assumptions which have not, as yet, been verified. These include the use of serum volume data determined by other workers and the extrapolation of results from control rats given a single 1 ml bovine albumin injection to albumin overloaded rats given ten 2.5 ml bovine albumin injections over a 5 day period. The close agreement between calculated and experimental values in this case does, however, indicate that the use of such calculations may be justifiable in other similar experimental situations.

At bovine albumin doses above 2.0 mg day\(^{-1}\) g\(^{-1}\) body wt. no limiting values were apparent in graphs relating the total protein excretion induced over a 5 day bovine albumin injection period to baseline albumin excretion and the correlation between baseline albumin excretion and induced proteinuria became progressively less as the amount of bovine albumin injected increased (Fig. 7) \((r = 0.67, n = 14)\) for bovine albumin doses between 2.0 and 3.5 mg day\(^{-1}\) g\(^{-1}\) body wt.; \( r = 0.17, n = 23 \) for bovine albumin doses between 3.5 and 5.5 mg day\(^{-1}\) g\(^{-1}\) body wt.). There was, however, a reasonably good correlation between baseline albumin excretion and the level of proteinuria induced on the first bovine albumin injection day in dose ranges 2.0-3.0 and 3.5-5.5 mg day\(^{-1}\) g\(^{-1}\) body wt. \((r = 0.93, n = 10 \) and \( r = 0.79, n = 23 \) respectively) and on the second bovine albumin injection day in the dose group 2.0-3.0 mg day\(^{-1}\) g\(^{-1}\) body wt. \((r = 0.89; n = 10)\).

In view of these findings, in the lowest bovine albumin dose groups (1.0-2.0, 2.0-3.0 and 3.0-3.5 mg day\(^{-1}\) g\(^{-1}\) body wt.) only the results from rats with low-to-intermediate baseline albumin excretion (30 of the 37 rats injected) were included in statistical analyses and mean baseline albumin excretion values were calculated for each dose group and tested for any significant differences before intergroup comparisons were made. In the highest bovine albumin dose groups (3.5-4.5 and 4.5-5.5 mg day\(^{-1}\) g\(^{-1}\) body wt.) all results were included in statistical analyses (23 rats).

**Total protein excretion as a function of bovine albumin dose and the relationship between proteinuria and glomerular epithelial cell footprocess loss.**

Total protein excretion over the 5 day bovine albumin injection period increased logarithmically with bovine albumin dose \((r = 0.86, n = 53)\). When results were divided into bovine albumin dose groups and analysed in histogram form (Fig. 8) the differences between mean total protein excretion values were significant in all comparisons \((P < 0.01)\) except that between dose groups 2.0-3.0 and 3.0-3.5 mg day\(^{-1}\) g\(^{-1}\) body wt. \((P > 0.05)\) where the large standard deviation in the higher dose group was probably responsible. The variable response to serum protein overload observed at this dose level was also noted by Kurtz & Feldman (1962). Maximum mean daily total protein excretion levels in the five bovine albumin dose groups studied were
FIG. 8. Histogram showing mean total protein excretion during a 5 day injection period for female Wistar rats in five different bovine albumin (BSA) dose groups: 1 = 1·0–2·0; 2 = 2·0–3·0; 3 = 3·0–3·5; 4 = 3·5–4·5; 5 = 4·5–5·5 mg day$^{-1}$ g$^{-1}$ body wt. Error bars represent standard deviations.

FIG. 10. Hyperalbuminaemic proteinuria induced by i.p. bovine albumin injection: relationship between percentage glomerular basement membrane (GBM) with normal foot processes (Fp) and total protein excretion on the fifth injection day. $n$ = Number of rats in each protein excretion range.

20 ± 16, 182 ± 65, 290 ± 217, 496 ± 189 and 667 ± 243 mg respectively (Fig. 9).

Mean percentage glomerular basement membrane with normal foot processes in glomeruli from control rats injected with phosphate-buffered saline alone was 83·19 ± 2·71%. In rats receiving i.p. bovine albumin injections, as the levels of induced proteinuria rose with increasing bovine albumin dose group, so the percentage glomerular basement membrane with normal foot processes decreased (Fig. 10) until, at the highest levels of proteinuria recorded on the fifth bovine albumin injection day (600–800 mg/24 h) only 22·53 ± 7·63% of the glomerular basement membrane was covered by normal epithelial cell foot processes. The mean values presented in this section for the percentage glomerular basement membrane with normal foot processes in each bovine albumin dose group were calculated from the average values obtained for the five glomeruli examined in each rat.

Discussion

Male rats are known to excrete higher levels of protein than female rats (Addis, Marmorston, Goodmann & Sellers, 1950; Sellers, Goodmann, Marmorston & Smith, 1950; Rümke, Breekveldt-Kielich & Van den Broeckesiddre, 1970) and such sex-related differences have been attributed, in Sprague-Dawley rats (Roy, Neuhaus &
Harrison, 1966), to the presence, in male urine alone, of sex-associated urinary proteins. The characteristics of the urinary specific proteins detected in the present study of female Wistar rats are similar to those of the 'sex-associated' proteins isolated from male rat urine by Royce (1968) and Roy et al. (1966). Since these proteins were shown, by paper electrophoresis, to be absent from female Sprague–Dawley rat urine (Roy et al., 1966), it seems that synthesis of these proteins may not be as strictly 'sex-associated' in the Wistar rat as it is in the Sprague–Dawley rat. These results indicate that further detailed studies of urinary protein composition are required before any firm conclusions concerning the basis for the different total protein content of male and female rat urine in strains other than the Sprague–Dawley can be reached.

In the present study a wide variation in baseline albumin excretion was measured in female Wistar rats which appeared to reflect inherent differences in glomerular permeability to albumin. These differences had a marked effect on the response to the serum albumin overload produced by i.p. bovine albumin injections, particularly at low parenteral protein doses. When interpreting the data concerned with the effect of baseline albumin excretion on the response to a range of bovine albumin doses, it was, however, essential to take into consideration any underlying relationships between bovine albumin dose and induced proteinuria or glomerular ultrastructural damage. Some of the results which were originally to have been included in a detailed report of the morphological and biochemical changes induced by a wide range of bovine albumin doses (G. M. Lawrence & D. B. Brewer, unpublished work) have therefore been presented in the present text (Figs. 8, 9, 10). They illustrate that the response to i.p. bovine albumin injections in the female Wistar rat was highly dose-dependent and that, as the level of induced proteinuria rose with bovine albumin dose, a corresponding increase in the degree of glomerular ultrastructural change induced also occurred. Therefore, since small inherent glomerular permeability differences could only have exerted their influence during periods when little additional glomerular ultrastructural damage had been induced, at levels of proteinuria above 100 mg/24 h [where gross glomerular ultrastructural damage must have been present in all animals regardless of their baseline albuminuria (Fig. 10)] inherent permeability differences should no longer have influenced the response to serum albumin overload.

This did, indeed, seem to be the case because, although induced proteinuria correlated well with baseline albumin excretion throughout the 5 day bovine albumin injection period in the lowest bovine albumin group studied (where percentage glomerular basement membrane with normal foot processes on day 5 was similar to the control value) a good correlation between the two variables was generally only apparent at higher bovine albumin doses during the first few injection days, when maximum daily protein excretion was well below 100 mg/24 h. The existence of this relationship between induced proteinuria and baseline albumin excretion indicated that, when the effects of two or more parenteral proteins which induce relatively low levels of proteinuria and glomerular ultrastructural damage are compared, care should be taken to ensure that baseline albumin excretion levels were similar in the groups of rats studied.

Such variations may account for the different levels of proteinuria induced by homologous and heterologous albumin by Fisher & Hellstrom (1962). The doses infused (1.0 g maximum over a 5 h period) were identical with the amounts reported by Davies et al. (1978) to induce, over a 24 h period, similar low levels of proteinuria and glomerular epithelial cell foot-process loss to those developed in response to 5 days i.p. injection with 1·0–2·0 mg of bovine albumin day–1 g–1 body wt. in the present study. It is therefore possible that differences in baseline albumin excretion may also have markedly affected the response to serum albumin overload in the first experimental situation. The results of this study cannot, therefore, be considered as definitive proof of the different quantitative effects of parenterally administered homologous and heterologous albumins as baseline albumin excretion was not determined in the two sets of rats and the differences in response measured may have been caused solely by inherent glomerular permeability differences.

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References
