Virchows Archiv A Pathol Anat Histopathol (1988) 412:459–470

Virchows Archiv A
Pathological Anatomy and Histopathology

The juxtaglomerular apparatus in Bartter's syndrome and related tubulopathies*

An immunocytochemical and electron microscopic study

R. Taugner[1], R. Waldherr[2], H.W. Seyberth[3], E.G. Erdös[4], J. Menard[5], and D. Schneider[1]

[1] Institute of Physiology, [2] Institute of Pathology and [3] Children's Hospital, University of Heidelberg, Im Neuenheimer Feld, D-6900 Heidelberg, Federal Republic of Germany
[4] Department of Internal Medicine, University of Texas Health Science Center, Dallas, TX 75235, USA
[5] Department of Clinical Research, Ciba-Geigy, CH-4002 Basel, Switzerland

Summary. A comparative immunocytochemical and electron microscopic study was performed on renal biopsies from two children with classical Bartter's syndrome (BS) and three children with a recently described variant, the so-called hyperprostaglandin E-syndrome (HES). Compared to age-matched controls, kidney specimens from patients with BS and HES disclosed a marked hypertrophy and hyperplasia of the juxtaglomerular apparatus (JGA). In addition, in HES focal tubular and interstitial calcifications accompanied by interstitial fibrosis and tubular atrophy were noted. On immunocytochemistry, chronic stimulation of the JGA in BS and HES was characterized by an increase in the number of renin-positive cells, particularly in the media of afferent arterioles, but also in efferent arterioles and in the glomerular stalk. The length of the renin-positive portion of the preglomerular arterioles was significantly increased when compared to controls (100 ± 32 vs. 49 ± 17 μm; $p < 0.001$). In addition, the immunoreactivity of individual renin-positive cells was markedly enhanced. On electron microscopy, "hypertrophy" of the RER and of Golgi complexes with paracrystalline deposits in dilated RER cisterns and protogranules indicated an increased renin synthesis. Renin could be identified in mature secretory granules as well as protogranules by immune electron microscopy. Angiotensinogen was present in hypertrophied epithelial cells of Bowman's capsule. Converting-enzyme reactivity was observed in controls as well as in BS and HES in the brush border of the proximal tubule. In contrast to previous reports, Angiotensin II was completely negative in control as well as in diseased kidneys. We conclude from our results that both BS and HES are characterized by a marked activation of the JGA and severe stimulation of the renin-angiotensin system. Since activation of this system, however, leads – independently of the primary stimulus – to qualitatively very similar morphological reactions, these results do not implicate a common pathogenetic mechanism to both conditions.

Key words: Bartter's syndrome – Hyperprostaglandin E-syndrome – Juxtaglomerular apparatus – Renin-angiotensin system – Immunocytochemistry

Introduction

In 1962, Bartter described a syndrome characterized by hypokalaemia, metabolic alkalosis, hyperreninaemia, secondary hyperaldosteronism, and normal blood pressure. In subsequent years it became apparent that Bartter's syndrome (BS) is not a uniform disease, but comprises a spectrum of conditions. Even if these disorders share the above mentioned basic features, there are some fundamental clinical and pathophysiological differences which justify the separation of variants (Stein 1985), or even distinct entities (Seyberth et al. 1985).

From a morphological point of view, all these conditions are characterized by a marked enlargement of the juxtaglomerular apparatus (JGA) on renal biopsy, and it was postulated (Bartter and Rodriguez 1982) that this finding constitutes an

* Dedicated to Prof. Dr. V. Becker, D-8520 Erlangen, on the occasion of his 65th birthday

Offprint requests to: R. Waldherr

Table 1. Laboratory values in patients with classical Bartter's syndrome (L.A., A.Z.) and the hyperprostaglandin E-variant (T.M., S.S., D.C.) prior to renal biopsy

Patients (Age)	K^+ serum (mmol/l)	Ca^{2+} urine (ng/kg/d)	C-CR (ml/min/1.73m^2)	PRA (mg/ml/h)	Aldo (ng/dl)	PGE_2	PGE-M (ng/h/1.73m^2)
L.A., ♂ (8 y)	2.6	0.7	106	35	12	20.5	499
A.Z., ♀ (4 y)	3.2	1.2	104	23	43	10.3	367
T.M., ♀ (1 y)	3.0	14.9	47	280	86	43.0	3772
S.S., ♀ (1 y)	2.6	14.5	43	400	172	57.1	820
D.C., ♀ (1 y)	2.5	13.3	49	260	375	99.5	2302
Normal	3.8–5.2	<5	90–130	1–6	<20	2–16	104–664

Abbreviations. C-CR – creatinine clearance; PRA – plasma renin activity; ALDO – aldosterone; PGE_2 – prostaglandin E_2; PGE-M – 7α-hydroxy-5,11-diketotetranorprostane-1,16-dioic acid (major urinary metabolite of E-prostaglandins)

essential requisite of the syndrome. Since comparative analysis of the JGA in BS and its subsets are lacking, we performed a detailed immunohistochemical and electron microscopic study on renal biopsies from children with classical BS and a recently described variant (Seyberth et al. 1985), the so-called hyperprostaglandin E syndrome (HES).

Materials and methods

Surgical renal biopsies were performed in two children with classical BS and three patients with HES. For comparison, renal biopsies from three age-matched normotensive children with isolated intermittent microscopic haematuria, completely normal on light microscopy and negative on routine immunohistology, were used as controls.

For light microscopy, the tissue was fixed in Dubosq-Brazil fluid and embedded in paraplast. Sections were stained routinely with haematoxylin and eosin, trichrome light green, periodic acid-Schiff with haematoxylin and chromotrope 2R silver methenamine.

For routine immunohistology, biopsy specimens were snap-frozen in isopentane chilled with liquid nitrogen. 3–5 µm cyostate sections were incubated with FITC-conjugated antisera to human IgA, IgG, IgM, C1q, C4, C3c, C3d, properdin, and fibrinogen-related antigen.

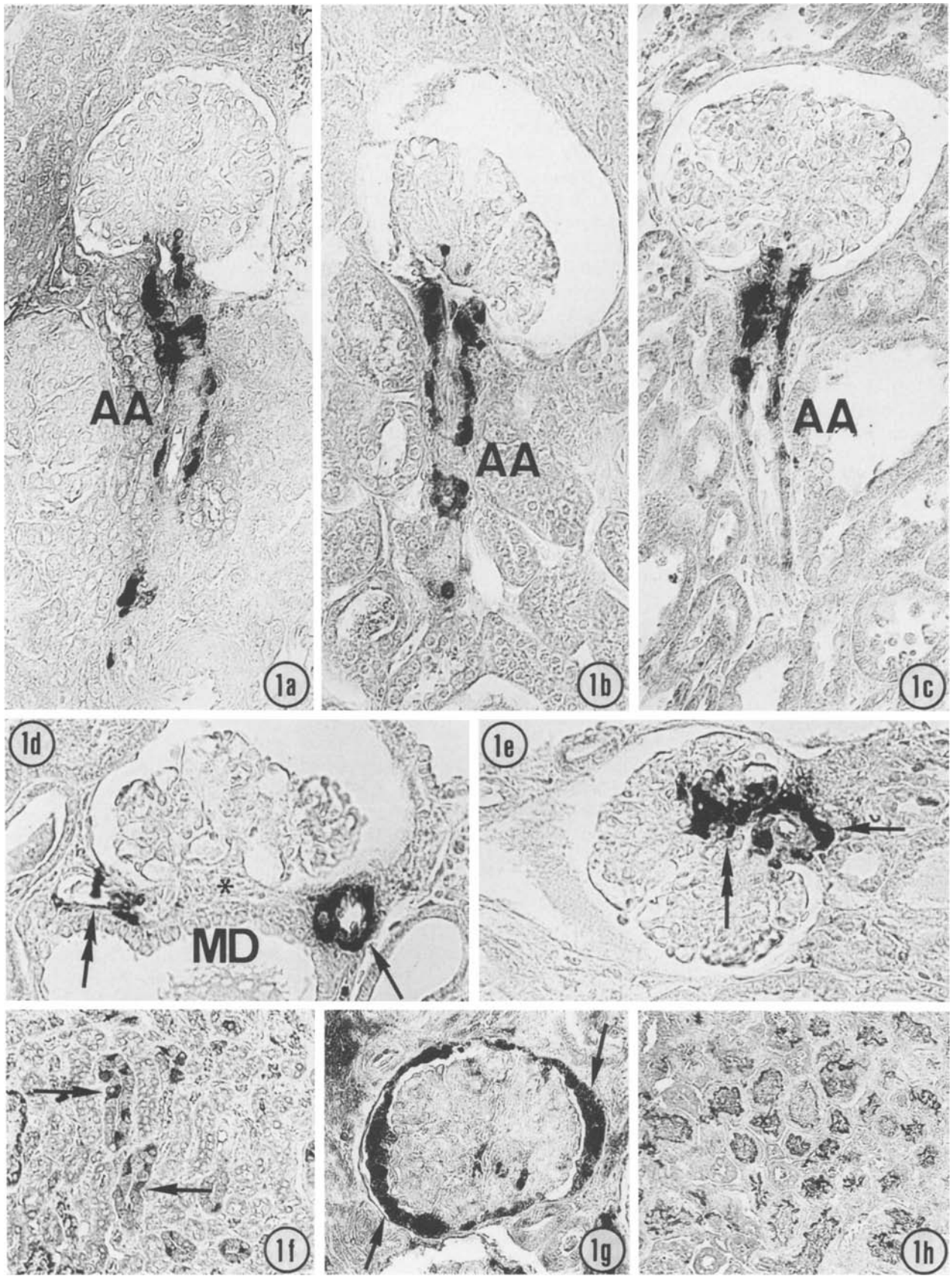
Immunohistochemistry was done on Paraplast sections according to the PAP-method (Taugner et al. 1979). The following antisera were used: anti-renin, anti-angiotensinogen, anti-converting enzyme and anti-angiotensin II.

Conventional electron microscopy was carried out on tissue fixed in 2.5% buffered glutaraldehyd and embedded in Epon. For immune electron microscopy, specimens were fixed in phosphate buffered 1% glutaraldehyde and embedded in London White resin. Postembedding staining was performed with the Protein A-gold method as described previously (Taugner et al. 1984a).

Results

All five patients showed typical clinical symptoms of BS such as hypokalaemia, metabolic alkalosis, hyperreninaemia, secondary hyperaldosteronism, and normal blood pressure. In addition, three patients (T.M., S.S., D.C.) presented with some remarkable features such as prenatal onset with polyhydramnios and prematurity at birth, episodic diarrhoea, hypercalciuria and nephrocalcinosis, associated with a marked increase of not only renal, but also systemic prostaglandin E_2 activity (see Seyberth et al. 1985). All other causes of hypokalaemia such as repeated vomiting, chronic diarrhoea, diuretic abuse, anorexia nervosa etc. were excluded. The pertinent laboratory data prior to renal biopsy are summarized on Table 1.

Fig. 1a–h. Immunocytochemical staining of the components of the renin-angiotensin-system. **a, b** Renin-positive reactions in the media of an afferent arteriole (*AA*) from patient S.S. (with HES). Antiserum dilution 1:5000 and 1:10000, respectively. Note the alternation between renin-positive and renin-negative medial cells in the proximal part of the vessel. ×320. **c** Renin-positive reaction in the media of an afferent arteriole (*AA*) from control kidney. Anti-renin dilution 1:2500. ×320. **d** Renin-positive reactions (patient D.C.) in afferent (↖) and efferent glomerular arterioles (↖ ↖). ×380. **e** Renin-positive cells (patient D.C.) in the glomerular (↖) and extraglomerular mesangium (↖). ×380. **f** Renin-positive reaction (patient A.Z.) of intercalated cells in collecting ducts of the outer medulla (↖). Antibody dilution 1:500. ×150. **g** Angiotensinogen-positive reaction (patient S.S.) of the parietal layer of Bowman's capsule (↖). ×290. **h** Converting enzyme-positive reaction (patient A.Z.) of the brush border of the proximal tubule. ×150


AA
1a
AA
1b
AA
1c
1d
MD
1e
1f
1g
1h

Renal biopsies disclosed marked hypertrophy and hyperplasia of the JGA in all patients. In addition, the three children with HES showed focal tubular and interstitial calcification associated with tubular atrophy, interstitial fibrosis and scattered mononuclear infiltrates. Vacuolization of proximal tubular cells was noted in all three biopsy specimens. Hyperplasia of renal interstitial medullary cells was absent in two biopsies containing medullary tissue. Features indicating a recently described familial proximal tubulopathy (Güllner et al. 1983) with an increased staining of proximal tubular cells and thickening of tubular basement membranes which ressembles BS on clinical grounds were not detected on light or electron microscopy. Routine immunohistology revealed minor depositis of C3c and C3d in arteriolar walls and along the basement membranes of atrophic tubules.

Immunohistochemical investigations showed no differences between the JGAs from kidneys of patients with BS and HES. The bulk of renin-positive cells was constantly found to be localized in the media of the juxtaglomerular portion of the afferent arterioles in control as well as in BS and HES kidneys (Fig. 1a–c). However, the length of the renin-positive segment of the preglomerular arteriole was significantly increased ($p < 0.001$) in the diseased kidneys (100 ± 32 µm; $n = 15$; Fig. 1a, b) when compared with controls (49 ± 17 µm; $n = 57$; Fig. 1c). Characteristically, not only the number of renin-positive cells but also their immunoreactivity were increased. In control kidneys, the antiserum could not be diluted more than 1:2500 to produce reliable immunostaining (Fig. 1c). In the diseased kidneys, a dilution of 1:10000 was still effective (Fig. 1b); smaller dilutions (e.g. 1:5000) already led to a diffuse spreading of the reaction product.

In contrast with controls, in patients with BS and HES single or small groups of renin-positive cells were also found at the origin of the afferent arteriole, in the juxtaglomerular portion of the efferent arteriole (Fig. 1d) and in the mesangium of the glomerular stalk (Fig. 1e). Furthermore, immunoreactive renin was observed in the apical portions of proximal tubular cells, in the intercalated cells of the connecting tubules and cortical as well as outer medullary collecting ducts (Fig. 1f).

In contrast with control kidneys, hypertrophied epithelial cells of the parietal layer of Bowman's capsule reacted intensely with the antibody against angiotensiogen (Fig. 1g). Converting enzyme-reactivity was encountered both in unchanged and in kidneys with BS and HS, particularly in the area of the brush border of proximal tubules (Fig. 1h).

Angiotensin I and angiotensin II were completely negative in controls as well as in the kidneys of patients with BS and HES.

Ultrastructure of biopsies from patients with BS and HES, showed proliferative reactions not only in the media of the hilar arterioles, but also in the area of the Goormaghtigh cell field and in the glomerular mesangium. Several profiles of typical Goormaghtigh cells and those of centrolobular mesangial cells presented round homogeneous electron dense organelles highly suggestive of secretory granules (Fig. 2a, b). However, because of their scarcity, these organelles could not be identified in the immunocytochemical experiments, and it remains unclear whether or not they contained renin.

In BS and HES, the media of the juxtaglomerular afferent arteriole was composed of a broadened collar of epithelioid cells (Fig. 3a). These cells generally had distinct and rarely extremely widened RER cisterns (Fig. 3e). Their Golgi complex was hypertrophied and filled with some paracrystalline protogranules (Fig. 3c) or conglomerate granules (Fig. 3d). In addition, paracrystalline deposits were found in dilated RER cisterns of some epithelioid cells (Fig. 3a, b).

Unlike controls, in which the media of the postglomerular arteriole consisted of a uniform type of pericyte-like cells, two types of granulated cells were observed in the efferent vessels of diseased kidneys (Fig. 4a): oval cells with an electronlucent cytoplasm, largely corresponding to the epithelioid cells of the preglomerular arteriole, and osmiophilic cells with numerous ramifications, which ressembled Goormaghtigh cells (compare Fig. 2a). At higher magnifications, all characteristics of an increased synthetic activity was observed in both types of postglomerular epithelioid cells: dilated RER cisterns containing material of moderate electron density, hypertrophied Golgi complexes, protogranules and mature secretory granules (Fig. 4b). With the protein A gold method, renin could be identified in the mature secretory granules as well as in the protogranules of these cells (Fig. 5).

Discussion

The initial observation by Bartter et al. (1962) that the juxtaglomerular apparatus (JGA) is enlarged in BS has been confirmed repeatedly (Bohle et al. 1982; McLaren and MacDonald 1982; Camilleri et al. 1983; for references to earlier publications see Christensen et al. 1976). In renal biopsies of

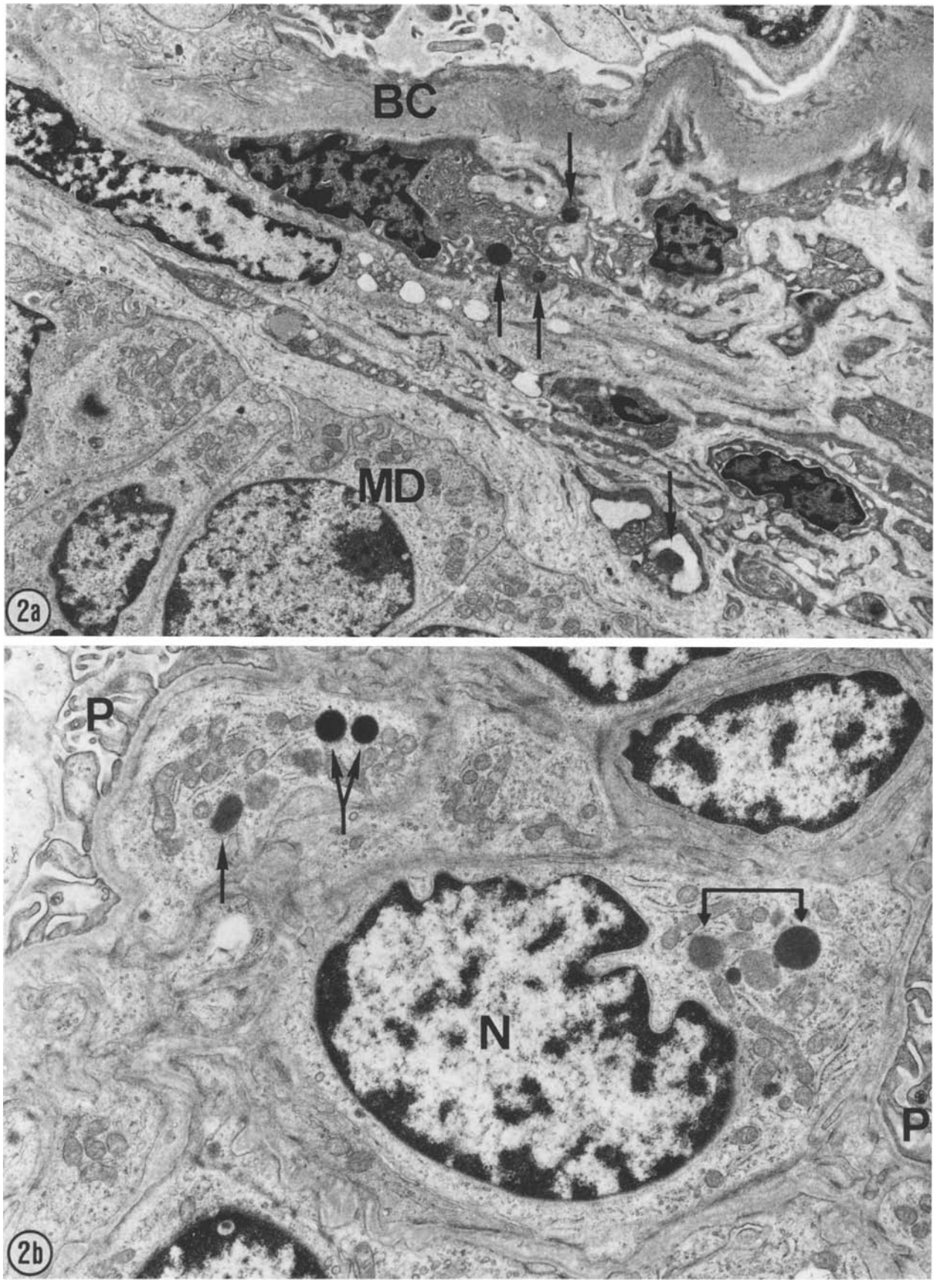
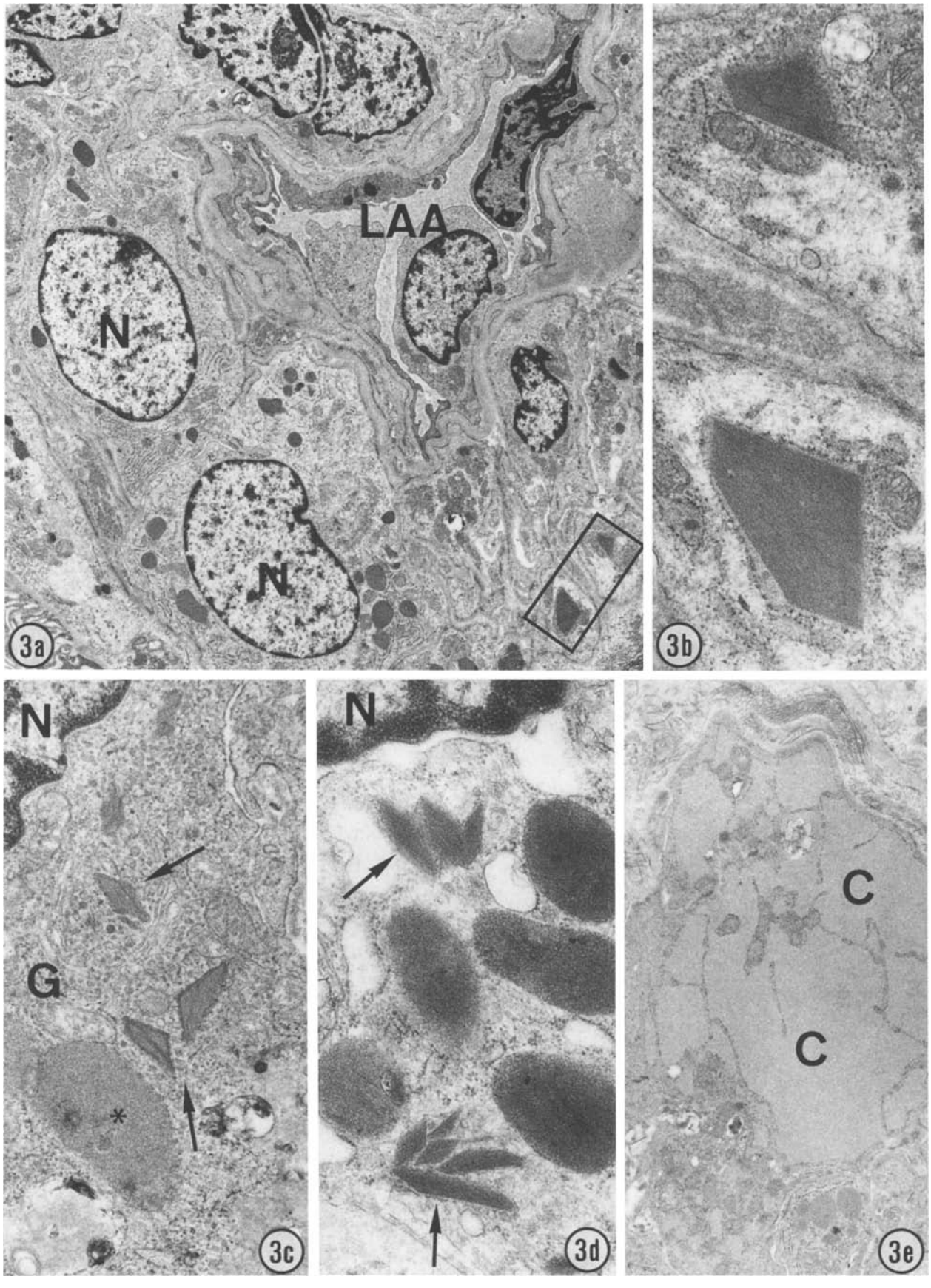


Fig. 2a, b. Secretory granules (↖) in Goormaghtigh (**a**) and mesangial cells (**b**) of patient S.S. (with HES). *BC* Bowman's capsule; *MD* Macula densa; *P* podocytes; *N* nucleus. ×7000 and 13600, respectively


LAA
N
N
3a
3b
N
G
*
3c
N
3d
C
C
3e

our patients with BS all signs of a marked chronic stimulation of renin synthesis were evident. The renin-positive portion of the preglomerular arteriole was considerably elongated when compared with controls. This finding implies that stimulation of renin synthesis had been followed by the recruitment of further granulated cells from the pool of plain smooth muscle cells by way of metaplastic transformation (Cantin et al. 1977; Taugner et al. 1984a). In addition, the number of renin-positive cells was augmented in the juxtaglomerular portion of the efferent arteriole and in the glomerular stalk. Finally, from the increased immunoreactivity and the hypergranulation of the individual epithelioid cells it can be concluded that the renin depot of the individual granulated cells is significantly increased in BS. In contrast to the report of Christensen et al. (1976), no hyperplasia of the macula densa or enlargement of the Goormaghtigh cell field were observed.

Likewise, renal specimens from three patients with HES, a variant of hypokalaemic tubular disorders recently described by Seyberth et al. (1985), disclosed all the above mentioned features of a marked chronic stimulation of renin synthesis. No difference was noted between biopsies of patients with BS and HES, neither with respect to the ultrastructure of the JGA, nor to the number and immunoreactivity of the renin-containing cells.

Therefore the question arises whether both syndromes share a common primary renal defect. In BS, several mechanisms have been suggested as the primary cause of hyperreninaemia including various functional defects of the renal tubule (Stein 1985) in particular a deficient chloride reabsorption in the ascending limb of Henle's loop (Bartter et al. 1962; Gill and Bartter 1978; Baehler et al. 1980; Bartter and Rodriguez 1982) with increased NaCl concentration of the distal tubular fluid at the level of the macula densa (Christensen et al. 1976); another suggestion is hyperplasia of the renomedullary interstitial cells with an inappropriate release of renal prostaglandins (Verbeckmoes et al. 1976) and also effects of the sympathetic nervous system (McLaren and MacDonald 1982). In HES, variations of the prostaglandin synthesis could play a role (cf. Seyberth et al. 1985; Table 1). However, on the basis of the present knowledge, it is impossible to define the effective stimulus of increased renin synthesis from the morphological changes encountered in the region of the JGA. Experimental studies suggest that any chronic stimulation of renin synthesis may lead to qualitatively similar effects in the JGA and the afferent arteriole, largely independent of the nature of the stimulus involved (Taugner et al. 1987). In view of recent results (Wagner et al. 1979; Bohle et al. 1982; McLaren and MacDonald 1982; Camilleri et al. 1983; Lindop and Lever 1986; Lindop 1987), similar conclusions may be drawn for the JGA in humans. Therefore, it would not be reasonable to conclude more than a comparable extent of stimulation of the renin-producing cells from the similar appearance of the JGA in BS and HES.

For quantification of the kidney renin status, specific antibodies and reproducible immunohistochemical methods are available. Apart from counting the immunoreactive JGAs per number of glomeruli encountered (Taugner et al. 1981; Camilleri et al. 1983; Nochy et al. 1983), the measurement of the renin-positive portion of the afferent arteriole as reported here represents a reliable method (Taugner et al. 1981). The results of both methods correlate well with the volume density of renin-positive cells as well as with the renin content of the renal cortex (Wurfer et al., in preparation).

The amount of extractable renin in the human kidney has been reported to be small when compared with that in other species (Schaffenburg et al. 1960). Celio and Inagami (1981), using the PAP technique, found only a very discrete immunostaining in the kidneys of normotensive healthy human subjects. Similarily, Nochy et al. (1983) detected only a few renin-containing cells in human kidneys by indirect immunofluorescence.

These observations are at variance with our results. The length of the renin-positive portion of the afferent arteriole, reflecting the number of renin-producing cells, amounted to 43 ± 19 µm ($n = 54$) in healthy adults (Hackenthal et al. 1987) and is shown here to be 49.0 ± 17 µm ($n = 57$) in normal children. These values are similar to that found in mice (45.4 ± 10.5 µm; $n = 60$) and surpass the length of the renin-positive portion in rats (36.8 ± 35 µm; $n = 38$) as well as in golden hamsters (24.0 ± 10.7 µm; $n = 60$; Hackenthal et al. 1987).

Fig. 3a–e. Epithelioid cells in the media of the afferent glomerular arteriole; **a–c** from patient A.Z. with BS, **d** from patient S.S. with HES. **a** Overview. *LAA* Lumen of the afferent glomerular arteriole; *N* nucleus. ×6000. **b** Area marked in **a** at higher magnification with paracrystalline inclusions in cisterns of the RER. ×32000. **c** Golgi complex (*G*) with adjacent mature granule (*) and several juvenile granules (↖) showing the lines of previous protogranule fusion. *N*: nucleus. ×31000. **d** Detail of an epithelioid cell with mature granules and conglomerates of protogranules (↖). ×37700. **e** Epithelioid cell process with bizarre dilatation of RER-cisterns (*C*). ×27000

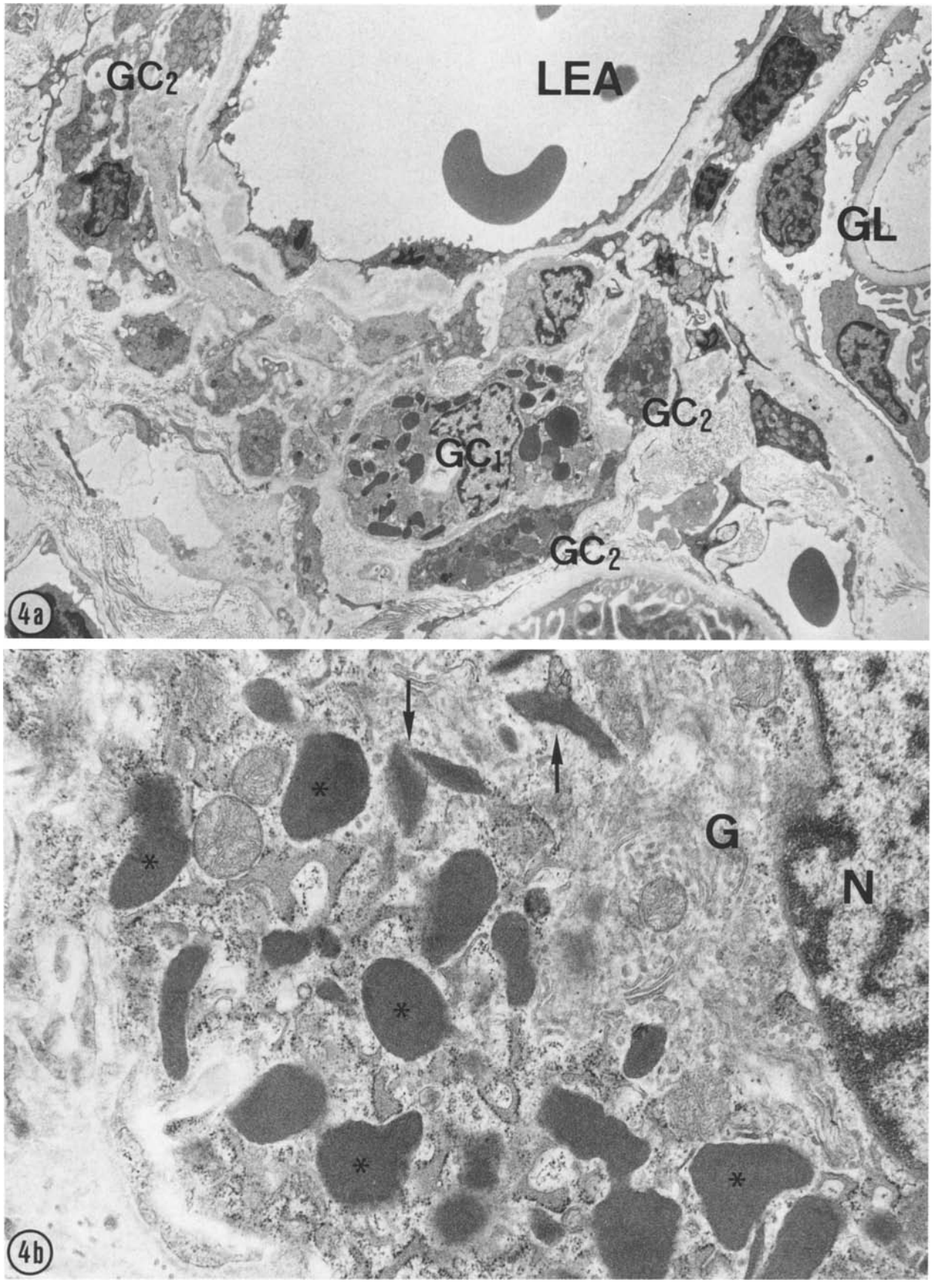


Fig. 4a, b. Granulated cells in the media of an efferent glomerula arteriole (patient L.A. with BS). **a** Overview: note the presence of two types of granulated media cell with different electron density (G_1 and GC_2, respectively). *LEA* Lumen of the efferent arteriole; *GL* glomerulus. ×4800. **b** Granulated cell (GC_1 in **a**) at higher magnification with mature secretory granules (*) and protogranules (↖). *G* Golgi complex; *N* nucleus. ×33000

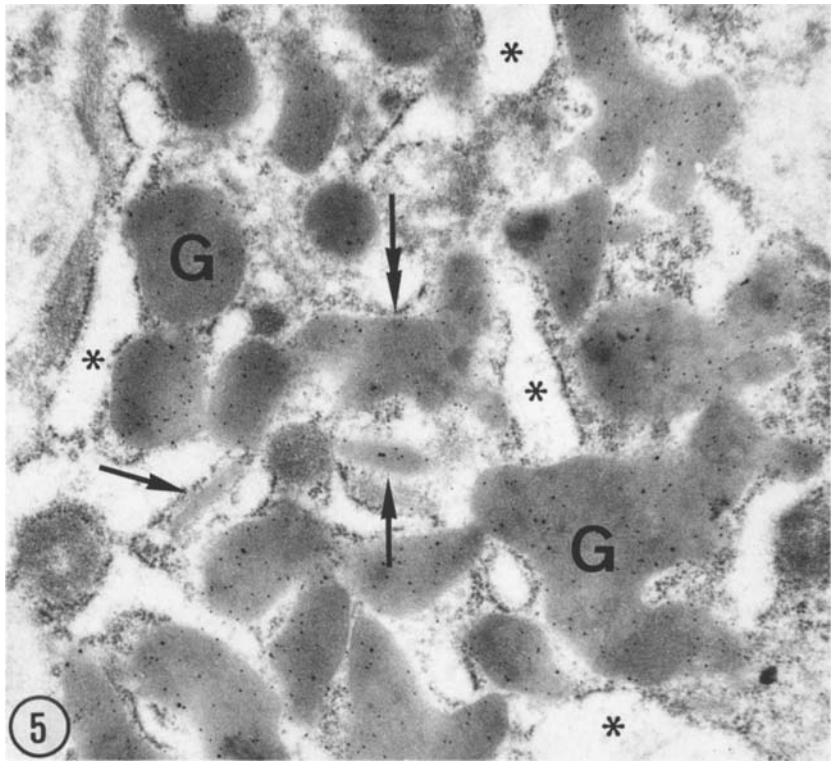


Fig. 5. Detail of an epithelioid cell from the afferent arteriole (patient T.M. with HES) showing renin-positive mature granules (*G*), protogranules (↖), protogranule conglomerates (↖ ↖) and dilated RER cisterns (*). Protein A-gold method, antiserum dilution 1:100. ×27200

The large deviations in the lengths of the renin-positive portion of the afferent arterioles encountered in all species are due to the extraordinarily large internephron heterogeneities in the equipment of individual nephrons with renin-positive cells.

In contrast to control kidneys, renin-positive cells were also found in the area of the glomerular mesangium in the kidneys of patients with BS and HES. This finding is in agreement with experimental results (Dunihue and Boldosser 1963; Barajas et al. 1976) and indicates that mesangial cells – like smooth muscle cells – posses the capacity to transform into epithelioid cells upon severe chronic stimulation. However, the renin-positive cells in BS and HES were still in connection with the glomerular stalk and not located between glomerular loops (cf. Barajas et al. 1976). In order to answer the question whether, apart from the mesangial cells of the glomerular stalk, peripheral (centrolobular) mesangial cells are also capable of transforming into epithelioid cells, it should be verified that the granules present in these cells are renin-positive.

Renin-positive Goormaghtigh cells could neither be identified in controls nor in BS or HES. However, because the lacis merges into the media of the glomerular arterioles without clear boundaries, we cannot exclude the possibility that some of the immunoreactive cells observed in the region of the JGA were transformed Goormaghtigh cells located at the periphery of the lacis. Hence, the need for specific markers for the different vascular component of the JGA and especially for Goormaghtigh and mesangial cells becomes apparent.

Angiotensinogen has been shown by to be localized in the apical region of proximal tubular cells (Richoux et al. 1984). It is very likely that it represents filtered angiotensinogen reabsorbed by pinocytosis. In agreement with the findings of Wagner et al. (1979) on pseudo-BS, we frequently observed hypertrophy of the parietal layer of Bowman's capsule in our cases of BS and HES. The outstandingly high angiotensinogen reactivity found in this area may be attributed to particularly high pinocytotic reabsorption of filtered angiotensinogen by these cells. Likewise, the immunoreactivity of the intercalated cells in the connecting and collecting tubules is likely to be due to pinocytosed angiotensinogen. However, it should be considered that the presence of mRNA coding for angiotensinogen (Campbell and Habener 1986) suggests that this peptide could be synthesized in the renal cortex.

Converting-enzyme has been found in high concentrations in the brush border of the proximal tubule (Erdös et al. 1967; Ward et al. 1975, 1976).

This is in agreement with our observations in BS and HES.

In the rat kidney, angiotensin II (ANG II) has been shown to coexist with renin in epithelioid cells (Celio and Inagami 1981; Taugner et al. 1981; Taugner et al. 1984a). The interpretation of this finding has been controversial (for reviews see Inagami et al. 1986; Hackenthal et al. 1987; Taugner and Hackenthal 1987). A strong argument against the suggestion that incorporate the presence of ANG II in epithelioid cell granules of the rat in far-reaching hypotheses on the function of the intrarenal renin-angiotensin system is the observation that the immunostaining for ANG II in the JGA of several other species is completely negative. We show here that even with very sensitive antibodies (Taugner et al. 1983) and in kidneys with excessive stimulation of renin synthesis ANG II is not detectable in the human JGA. These findings are at variance with positive results reported by Celio (1981). However, our negative findings correspond to the fact that neither angiotensinogen nor ANG I and converting-enzyme could be detected in epithelioid cells of humans and several other species (Hackenthal et al. 1987).

The ultrastructure of epithelioid cells in the media of the afferent arteriole in control kidneys corresponded largely to that of the original description by Biava and West (1966). In the epithelioid cells of kidneys with BS and HES, all signs of an increased renin synthesis were observed, some of them suggestive of an overcharge in the packaging of the secretory product. According to Barajas (1966), granulopoiesis in epithelioid cells proceeds via protogranules, shown to pinch off from the dilated rims of the innermost Golgi cisterns. Upon stimulation of the JGA, e.g. by reduction of the pressure in the renal arterial tree, the number of rhomboid protogranules with paracrystalline contents increases. Subsequently, several protogranules may merge into conglomerate granules, in which the individual structure of the original protogranules may still be seen, until finally the large mature granules with amorphous contents emerge (Barajas 1966; Lindop and Downie 1984; Taugner and Metz 1986).

Juvenile granules with paracrystalline contents were regularly seen in the epithelioid cells of patients with BS and HES. They have been described as "renin bodies" with sharply angulate shapes in a case of Bartter's syndrome by Zavagli et al. (1983) and have been shown to contain prorenin (Taugner et al. 1986; 1987). However, paracrystalline deposits were also found in the dilated cisterns of the RER in BS as well as in HES kidneys. These RER deposits differed from the rhomboid protogranules, which, as post-Golgi deposits, are enclosed by a smooth membranous sack, particularly by their membranous envelope studded with ribosomes. In addition, the RER deposits were usually bigger than protogranules and had a sharp-edged, asymmetrical profile instead of being rhomboid. Although paracrystalline deposits in RER cisterns of other cell types have been known for many years, there have been no reports on the existence of polyhedral paracrystalline structures in the RER of human epithelioid cells. In mice, the corresponding RER inclusions are globular (Kaneta et al. 1981; Hackenthal et al. 1987). Since both protogranules and paracrystalline RER inclusions prevail upon severe stimulation of renin synthesis, they may be interpreted as a sign of overcharge in the processing and/or packaging of the secretory product. Paracrystalline inclusions have repeatedly been observed in the cells of renin-producing tumours (Mimran et al. 1978; Baldet et al. 1983; Camilleri et al. 1984; Galen et al. 1984; Squires et al. 1984; Tetu et al. 1984; Lindop et al. 1986) indicating a similar disturbance of activation and packaging of renin. It is of particular note that such tumour cells secrete predominantly inactive renin, i.e. prorenin, and high levels of inactive plasma renin have also been reported in BS (Nagai et al. 1984).

References

Baehler RW, Work J, Kotchen TA, McMorrow G, Guthrie G (1980) Studies on the pathogenesis of Bartter's syndrome. Am J Med 69:933–938

Baldet P, Mimran A, Granier M, Dupont M (1983) Formes histologiques et ultrastructurales des tumeurs bénignes du rein avec sécrétion de rénine. Ann Pathol 3:225–234

Barajas L (1966) The development and ultrastructure of the juxtaglomerular cell granule. J Ultrastruct Res 15:400–413

Barajas L, Wang P, Bennett CM, Wilburn R (1976) The renal sympathetic system and juxtaglomerular cells in experimental renovascular hypertension. Lab Invest 35:574–587

Bartter FC, Rodriguez A (1982) Bartter's syndrome. Adv Intern Med Pediatr 50:79–103

Bartter FC, Pronove P, Gill JR Jr, MacCardle RC (1962) Hyperplasia of the juxtaglomerular complex with hyperaldosteronism and hypokalemic alkalosis. A new syndrome. Am J Med 33:811–828

Biava CG, West M (1966) Fine structure of normal human juxtaglomerular cells. Am J Pathol 49:679–721

Bohle A, Christensen J, Meyer DS, Laberke HG, Strauch M (1982) Juxtaglomerular apparatus of the human kidney: correlation between structure and funtion. Kidney Int 22 (Suppl 12):S18–S23

Camillieri J-P, Hinglais N, Nochy D, Phat VN, Bariety J (1983) Immunohistochemistry of renin in human diseased kidney. Clin Exp Hyper-Theor Pract A5:1179–1190

Camillieri J-P, Hinglais N, Bruneval P, Bariety J, Tricottet V,

Rouchon M, Maniclla-Jimenez R, Corvol P, Menard J (1984) Renin storage and cell differentiation in juxtaglomerular cell tumors: an immunohistochemical and ultrastructural study of three cases. Hum Pathol 15:1069–1079

Campbell DJ, Habener JF (1986) Angiotensinogen gene is expressed and differentially regulated in multiple tissues of the rat. J Clin Invest 78:31–39

Cantin M, Araujo-Nascimento M-F, Benchimol S, Desormeaux Y (1977) Metaplasia of smooth muscle cells into juxtaglomerular cells in the juxtaglomerular apparatus, arteries and arterioles of the ischemic (endocrine) kidney. Am J Pathol 87:581–602

Cantin M, Gutkowska J, Lacasse J, Ballak M, Ledoux S, Inagami T, Beuzeron J, Genest J (1984) Ultrastructural immunocytochemical localization of renin and angiotensin II in the juxtaglomerular cells of the ischemic kidney in experimental renal hypertension. Am J Pathol 115:212–224

Celio M, Inagami T (1981) Angiotensin II reactivity coexists with renin in the juxtaglomerular granular cells of the kidney (immunoperoxidase techniques/intracellular peptide synthesis/paracrine secretion/tubulo-glomerular feedback). Proc Natl Acad Sci USA 78:3897–3900

Christensen JA, Bader H, Bohle A, Meyer DS (1976) The structure of the juxtaglomerular apparatus in Addison's disease, Bartter's syndrome, and Conn's syndrome. Virchows Arch [Pathol Anat] 370:103–112

Dunihue FW, Boldosser WG (1963) Observations on the similarity of mesangial to juxtaglomerular cells. Lab Invest 12:1228–1240

Erdös EG, Yang HYT (1967) An enzyme in microsomal fraction of kidney that inactivates bradykinin. Life Sci 6:569–574

Galen FX, Devaux C, Houot AM, Menard J, Corvol P, Corvol MT, Gubler MC, Mounier F, Camillieri JP (1984) Renin biosynthesis by human tumoral juxtaglomerular cells. J Clin Invest 73:1144–1155

Gill JR Jr, Bartter FC (1978) Evidence for a prostaglandin-independent defect in chloride reabsorption in the loop of Henle as a proximal cause of Bartter's syndrome. Am J Med 65:766–772

Güllner HG, Bartter FC, Gill JR Jr, Dickman PS, Wilson CB, Tiwari JL (1983) A sibship with hypokalemic alkalosis and renal proximal tubulopathy. Arch Intern Med 143:1534–1540

Hackenthal E, Metz R, Bührle CP, Taugner R (1987) Intrarenal and intracellular distribution of renin and angiotensin. Kidney Int 31 (Suppl 20):S4–S17

Inagami T, Kawamura M, Naruse K, Okamura T (1986) Localization of components of the renin-angiotensin system within the kidney. Fed Proc 45:1414–1419

Kaneta M, Abe K, Ito T (1981) Appearance of dense granules in the rough endoplasmic reticulum of the juxtaglomerular cells in mice administered with captopril. Cell Tissue Res 220:219–222

Lindop GBM (1987) Morphological aspects of renin synthesis, processing storage and secretion. Kidney Int 31 (Suppl 20):S18–S34

Lindop GBM, Downie TT (1984) New morphological evidence for the synthesis and storage of renin in the human kidney: an ultrastructural immunocytochemical study. J Hypertension 2:7–10

Lindop GBM, Lever AF (1986) Anatomy of the renin-angiotensin system in the normal and pathological kidney. Histopathology 10:335–362

Lindop GMB, Leckie B, Winearls CG (1986) Malignant hypertension due to a renin-secreting renal cell carcinoma-an ultrastructural and immunocytochemical study. Histopathology 10:1077–1088

McLaren KM, MacDonald MK (1982) Histological and ultrastructural studies of the human juxtaglomerular apparatus in Bartter's syndrome and renal artery stenosis. J Pathol 136:181–197

McLaren KM, MacDonald MK (1983) Histological and ultrastructural studies of the human juxtaglomerular apparatus in benign and malignant hypertension. J Pathol 139:41–55

Mimran A, Leckie BJ, Fourcade JC, Baldet P, Navratil H, Barjon P (1978) Blood pressure, renin-angiotensin system and urinary kallikrein in a case of juxtaglomerular cell tumor. Am J Med 65:527–536

Nagai H, Matsunaga M, Ogawa K, Kuwahara T, Kanatsu K, Pak CH, Hara A, Tamura T, Kono T, Kawai C (1984) High level of plasma inactive renin in Bartter's syndrome. Jpn Circ J 48:633–637

Nochy D, Barres D, Camillieri J-P, Bariety J, Corvol P, Menard J (1983) Abnormalities of renin-containing cells in human glomerular and vascular renal diseases. Kidney Int 23:375–379

Richoux JP, Cordonnier JL, Bouhnik J, Clauser E, Corvol P, Menard J, Grignon G (1983) Immunocytochemical localization of angiotensinogen in rat liver and kidney. Cell Tissue Res 233:439–451

Schaffenburg CA, Haas E, Goldblatt H (1960) Concentration of renin in kidneys and angiotensin in serum of various species. Am J Physiol 199:788–792

Seyberth HW, Rascher W, Schweer H, Kühl PG, Mehls O, Schärer K (1985) Congenital hypokalemia with hypercalciuria in preterm infants: a hyperprostaglandinuric tubular syndrome different from Bartter's syndrome. J Pediatr 107:694–701

Squires JP, Ulbright TM, Deschryver-Kecskemeti K, Engleman W (1984) Juxtaglomerular cell tumor of the kidney. Cancer 53:516–523

Stein JH (1985) The pathogenetic spectrum of Bartter's syndrome. Kidney Int 28:85–93

Taugner C, Poulson K, Hackenthal E, Taugner R (1979) Immunocytochemical localization of renin in mouse kidney. Histochemistry 62:19–27

Taugner R, Hackenthal E (1988) On the character of the secretory granules in juxtaglomerular epitheloid cells. Int Rev Cytol in press

Taugner R, Metz R (1986) Development and fate of the secretory granules of juxtaglomerular epitheloid cells. Cell Tissue Res 246:595–606

Taugner R, Hackenthal E (1981) Angiotensin II in epitheloid (renin containing) cells of rat kidney. Histochemistry 72:499–509

Taugner R, Hackenthal E, Nobiling R, Harlacher M, Reb G (1981) The distribution of renin in different segments of the renal arterial tree. Histochemistry 73:75–88

Taugner R, Bührle CP, Ganten D, Hackenthal E, Hardegg C, Hardegg G, Nobiling R, Unger T (1983) Angiotensin-like activity in resistance vessels. Histochemistry 78:61–70

Taugner R, Bührle CP, Hackenthal E, Mannek E, Nobiling R (1984a) Morphology of the juxtaglomerular apparatus and secretory mechanisms. Contrib Nephrol 43:76–101

Taugner R, Mannek E, Nobiling R, Bührle CP, Hackenthal E, Ganten D, Inagami T, Schröder H (1984b) Coexistence of renin and angiotensin II in epitheloid cell secretory granules of rat kidney. Histochemistry 81:39–45

Taugner R, Murakami K, Kim S-J (1986) Renin activation in juvenile secretory granules? Histochemistry 85:107–109

Taugner R, Kim S-J, Murakami K, Waldherr R (1987) The

fate of prorenin during granulopoiesis in epithelioid cells. Histochemistry 86:249–253

Tetu B, Totovic V, Bechtelsheimer H, Smend J (1984) Tumeur rénale à sécrétion de rénine. Ann Pathol 4:55–59

Verbeckmoes R, van Damme B, Clement J, Amery A, Michielsen P (1976) Bartter's syndrome with hyperplasia of renomedullary cells: successful treatment with indomethacin. Kidney Int 9:302–307

Wagner PK, Pippig L, Thoenes W (1979) Zur Histopathologie der Niere beim Pseudo-Bartter-Syndrom durch chronischen Diuretikaabusus. Klin Wochenschr 57:135–142

Ward PE, Gedney CD, Dowben RM, Erdös EG (1975) Isolation of membrane-bound renal kallikrein and kininase. Biochem J 151:755–758

Ward PE, Erdös EG, Gedney CD, Dowben RM (1976) Isolation of membrane-bound enzymes that metabolize kinins and angiotensins. Biochem J 157:643–650

Zavagli G, Aleotti A, Farinelli A (1983) Human renin granules: ultrastructural aspects. Nephron 33:29–33

Accepted October 27, 1987