The Epidermal Growth Factor Precursor Isolated from Murine Kidney Membranes

CHEMICAL CHARACTERIZATION AND BIOLOGICAL PROPERTIES*

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To understand the biology and the biochemistry of the epidermal growth factor (EGF) precursor in normal tissues we partially purified the EGF precursor from mouse kidney. The precursor was purified by affinity chromatography, using wheat germ lectin and antibodies to murine EGF. The precursor was a glycosylated integral membrane protein of apparent molecular mass of 140–150 kDa. The solubilized EGF precursor is biologically active as evidenced by its ability to compete with 125I-labeled EGF for binding to the EGF receptor in intact fibroblasts and its ability to stimulate the growth of cells dependent on EGF for growth. The EGF precursor from mouse kidney can be proteolytically processed by the EGF-associated arginine esterase into a smaller fragment (97 kDa) that retains both immunologic sensitivity to EGF antiserum and biological activity. Extensive digestion of the EGF precursor with pepsin liberates a biologically and immunologically active protein of approximately the size of mature EGF.

Epidermal growth factor (EGF), a 53-amino acid polypeptide originally isolated from mouse submaxillary glands, induces precocious eyelid opening and early incisor eruption when injected into newborn mice (1). These biological effects are a consequence of enhanced epidermal growth and keratinization. The homologous human protein (hEGF) has been purified from the urine (2, 3); it is structurally and functionally identical to urogastrone, a gastric acid antisecretory hormone (4).

EGF influences an array of biologic responses. The stimulation by EGF of the proliferation and differentiation of a variety of cultured epithelial and mesenchymal cells has been described (5). Binding of EGF to its receptor activates the intrinsic tyrosine kinase activity of the receptor and appears to initiate many other intracellular events (6). In addition, EGF has been demonstrated to have effects seemingly unrelated to its mitogenic capacity such as the stimulation of smooth muscle contraction and the inhibition of gastric acid secretion (7, 8). The normal physiologic function of EGF, however, has not yet been determined.

The relationship between EGF and the kidney first received attention when it was noted that urine contained high levels of EGF, while circulating blood contained only traces of EGF derived largely from platelets (9, 10). The high levels of EGF in urine relative to blood suggested that the kidney itself might be the source of urinary EGF. Olsen et al. (11) reported that unilateral nephrectomy reduced both rat urinary EGF levels and the EGF/creatinine ratio by about 50% supporting an important role for the kidney as the source of urinary EGF.

Examination of the cDNA of murine EGF indicated that the 53-amino acid protein is synthesized as a 1217-amino acid precursor (12, 13). The mRNA for the EGF precursor in the mouse are highest in the submandibular gland and the kidney and are orders of magnitude lower in the lung, spleen, brain, and ovary (14). In humain and other species the kidney may have the highest levels of the EGF precursor mRNA (15). In situ mRNA hybridization and immunohistochemical staining using antibodies to EGF localize the precursor to the thick ascending limb of Henle and the distal convoluted tubule (16).

From analysis of the cDNA sequence, the EGF precursor (pro-EGF) is predicted to have a transmembrane protein with an extracellular domain that contains a potential amino-terminal signal sequence, EGF, and 7–9 other EGF-like domains, a hydrophobic transmembrane domain, and a 165-amino acid cytoplasmic domain (12, 13). It is unknown whether the EGF-like sequences are processed or what potential biologic functions they could serve. It may be that, in a manner analogous to the glucagon or pro-opiomelanocortin precursors these other segments of the EGF precursor may have important biologic roles (17, 18). It is of interest that the homoeotic proteins NOTCH and tin-12 also contain multiple EGF-like sequences (19, 20).

The release of EGF from the EGF precursor would require both amino-terminal and COOH-terminal proteolytic processing. The cDNA analysis of the EGF precursor predicts an arginine immediately proximal to the amino-terminal asparagine of murine EGF and EGF, itself, has an arginine at its carboxyl terminus. Interestingly, murine EGF has been isolated as a complex containing an arginine esterase, the EGF binding protein (21). It is possible that this arginine esterase activity is involved in the processing of the precursor.

Stably transfected mouse NIH 3T3 cell lines expressing the human kidney EGF precursor cDNA have been established and the expressed precursor partially characterized (22, 23). However, little is known about the EGF precursor in normal tissues. As a first step toward understanding the biology and
biochemistry of the EGF precursor and its possible in vivo function we report the detection and partial purification of the native EGF precursor from the mouse kidney. The EGF precursor was found to be a glycosylated membrane-associated protein with an approximate molecular mass of 140–150 kDa. The intact, purified EGF precursor appears to be biologically active; it is able to bind to EGF receptors on human fibroblasts and to stimulate the growth of mouse keratinocytes (MKB cells). The EGF precursor may be proteolytically cleaved to biologically active lower molecular weight fragments.

**MATERIALS AND METHODS**

**Tissue Preparation and Membrane Solubilization—**Kidneys were harvested from 40-g mice (ICR, Harlan), frozen in liquid nitrogen, and stored at −70 °C. Thawed kidneys were homogenized in 4 ml of a buffer (20 mM Hepes, pH 7.4, 2 mM MgCl2) per g of tissue (wet weight) using a Brinkman Polytron (PTI0ST). The homogenates were centrifuged at 100,000 × g for 90 min at 4 °C. The pellets were reextracted in the same manner and the supernatants were combined.

**Lectin Chromatography—**Aliquots of the solubilized membrane proteins (4 ml) were added to 1 ml of wheat germ lectin (Sigma) in a 10-ml column (Bio-Rad). The mixture was stirred for 90 min at 4 °C. The column was then centrifuged (3 min at 100,000 × g) to collect the nonadsorbed material. The beads were washed twice with 4 ml of Buffer A and 6 times with 4 ml of Buffer A without Triton X-100. Finally, the precursor was eluted by incubating the beads in 2 ml of elution buffer (20 mM Hepes, pH 7.4, 150 mM NaCl, 1% Triton X-100) and incubated for 30 min at room temperature. The mixture was centrifuged at 100,000 × g for 30 min and the supernatant was saved. The pellets were reextracted in the same manner and the supernatants were combined.

**Immunofinity Chromatography—**Rabbit polyclonal anti-mouse EGF antiserum (Plasmon, LKB Biotechnology Inc.) or lentil lectin-Sepharose 4B (Pharmacia) and a single component

**Electrophoretic Digestion of the EGF Precursor with Pepsin—**The chitotriose eluate from the wheat germ lectin column (100 μl) was dialyzed overnight against an acid solution (0.01 M HCl, pH 1.5, 150 mM NaCl) and then incubated either in the absence or in the presence of varying concentrations (0.01, 0.1, or 1 μg) of hog pepsin (Sigma) for 1 h at 37 °C. The samples were then immediately neutralized. Aliquots were submitted to SDS gel electrophoresis with 12% gels.

**Competitive Binding Experiments—**EGF was prepared from mouse salivary gland by the method of Savage and Cohen (26). The radioactivity assay for EGF was carried out essentially as described previously (26). Human foreskin fibroblasts were grown to confluence in 60-mm dishes. The growth medium was removed and the cells were washed three times with PBS. The dishes were coated with 1× bovine serum albumin and the indicated additions, were added to each dish. The cells were incubated for 2 h at 37 °C. The dishes were then washed three times with 2 ml of PBS containing 0.1% BSA at 37 °C and the cells again were incubated with 1.5 ml of Dulbecco's medium containing 0.1% BSA and [125I]labeled EGF (32 ng, 40,000–100,000 cpm/μg). After 1 h, unbound radioactivity was removed by washing the cells 6 times with cold PBS containing 0.1% BSA. The cells were dispersed with 0.5 M NaOH (60 min at 37 °C) and cell-associated radioactivity was determined in a Gamma 4000 counter (Beckman Instruments, Inc.).

**Gel Electrophoresis and Immunoblotting—**Samples were mixed with SDS sample buffer omitting β-mercaptoethanol, heated for 2 min at 100 °C, and subjected to SDS–gel electrophoresis (29). Unless otherwise noted 7% gels were utilized. Following electrophoresis, gels were stained with Coomassie Blue or washed extensively and silver-stained (30).

**Stimulation of MKB Cell Growth—**A mouse keratinocyte cell line MKB (generously provided by S. A. Aaronson (27)) that is dependent on EGF for a growth signal was used as a bioassay for EGF. The cells were plated in 30-mm dishes at a density of 150,000 cells/dish in low calcium minimal essential medium supplemented with 10% Chelex 100 (Bio-Rad)-treated fetal calf serum and 4 ng of EGF/ml. The next day, the dishes were washed 5 times with calcium/magnesium-free PBS and incubated with 2 ml of a medium containing 160 μl of Chelex 100-treated fetal calf serum, 840 μl of low calcium medium, and the indicated amounts of EGF, precursor or EGF. The EGF precursor in the wheat germ lectin eluate (300 μl) was passed through 100 μl of Extracti-Gel (Pierce Chemical Co.) to remove traces of Triton X-100. The Extracti-Gel flow-through was then dialyzed against low calcium minimal essential medium prior to its addition to MKB cells.

**Results**

**Subcellular Distribution of the EGF Precursor in Mouse Kidney—**In preliminary experiments we were able to detect the presence, in crude kidney homogenates, of a high molecular weight immunoreactive protein (the presumed EGF precursor) by immunoblotting using a polyclonal rabbit antiserum to mature 6-kDa murine EGF (1). No immunoreactive protein band was found on immunoblots either with preimmune serum or anti-EGF antiserum preadsorbed with murine EGF (data not shown).

The kidney homogenates were centrifuged and aliquots of the soluble and particulate fractions were subjected to SDS–gel electrophoresis and immunoblotting as described under "Materials and Methods." Almost all of the EGF precursor in mouse kidney was present in the crude membrane fraction as a single component of molecular mass 140–150 kDa (Fig. 1,
Only trace amounts of a slightly lower molecular weight form could be detected in the soluble fraction (Fig. 1, lane B). No soluble mature EGF was detected. The EGF precursor appeared to be an integral membrane protein, since it remained membrane-bound after extraction with 1 M NaCl or 0.2 M Na2CO3, pH 11 (data not shown) and required detergents for solubilization.

It should be noted that immunoblotting was carried out on samples prepared in the absence of mercaptoethanol. The sensitivity for detection by the antibody was reduced 5-10-fold when either mature EGF or the EGF precursor were heated in SDS sample buffer containing mercaptoethanol. This may reflect the fact that the antibody to EGF for immunoblotting was made against native EGF and not a reduced form.

Solubilization of the EGF Precursor from Mouse Kidney Membranes—The EGF precursor present in the crude membrane fraction of kidney homogenates may be solubilized by Triton X-100. Two successive extractions with 1% Triton X-100 were sufficient to completely remove the immunoreactive material from the membrane fraction (data not shown). The two Triton X-100 extracts were pooled.

Purification of the EGF Precursor by Lectin Chromatography—The pooled Triton X-100 extract was chromatographed on a wheat germ lectin column and aliquots again were subjected to SDS-gel electrophoresis and immunoblotting. Almost all of the EGF precursor present in the Triton extract (Fig. 2A, lane A) remained adsorbed to the lectin column; only small amounts appeared in the flow-through fraction (lane B) or in the subsequent washes (lane C). The EGF precursor was specifically eluted from the column with 3 mM \(N,N',N''\)-triacetylchitotriose (lane D). The chitotriose eluate contained only 13% of the total protein applied to the wheat germ lectin column. It should be noted that the precursor was eluted from the wheat germ lectin column by \(N,N',N''\)-triacetylchitotriose in the absence of Triton X-100. The absence of detergent enabled us to assay the biologic activity of the precursor using intact cells (see later section).

The binding of the precursor to the wheat germ lectin and its specific elution suggested that the precursor was glycosylated. To further examine this, the Triton X-100 extract of mouse kidney membranes was chromatographed on lentil lectin and concanavalin A columns as described under “Materials and Methods.” The EGF precursor present in the Triton extract (Fig. 2B, lane A) was absorbed to the lentil lectin column with little appearing in the flow-through fraction (lane B). The majority of the EGF precursor was specifically eluted with \(\alpha\)-methylmannoside (lane E). Similarly, the EGF precursor was absorbed to the concanavalin A column with no immunoreactive material in the flow-through fraction. However only a small fraction could be eluted from the column with \(\alpha\)-methylmannoside (data not shown). The adsorption of the EGF precursor to all three lectin columns supports the conclusion that the EGF precursor is glycosylated.

Purification of the EGF Precursor by Immunoaffinity Chromatography—The wheat germ lectin eluate containing the EGF precursor was purified further by immunoaffinity chromatography. A rabbit antiserum to EGF was affinity purified...
on an EGF-Affi-Gel column and was then linked to cyanogen bromide-activated Sepharose 4B as described under “Materials and Methods.”

The eluate from the wheat germ lectin column, containing the EGF precursor, was applied to the anti-EGF Sepharose 4B column and the eluted fractions were subjected to SDS-gel electrophoresis and examined by immunoblotting (Fig. 3A) or silver staining (Fig. 3B). The EGF precursor present in the wheat germ lectin eluate (Fig. 3, lane A) adsorbed to the immunoaffinity column with virtually none in the flow-through fraction (Fig. 3, lane B). After several washes, the EGF precursor was eluted with an acid buffer (Fig. 3, lane E). Silver stains of each of the fractions are illustrated in Fig. 3B. The single major silver-stained band seen in lane E corresponds in mobility to the EGF precursor band detected by immunoblotting.

Biological Activity of the EGF Precursor—The biological activity of the EGF precursor eluted from the wheat germ lectin column (Fig. 2A, lane D) was examined by two independent procedures: (a) the precursor was assayed for its ability to compete with 125I-labeled murine EGF for binding to EGF receptors on intact human fibroblasts; and (b) the growth-stimulatory effects of murine EGF and the EGF precursor were compared using mouse keratinocytes (MKB cells).

Both unlabeled EGF and the EGF precursor inhibit the binding of 125I-labeled EGF to fibroblasts (Table I, Exp. 1). We estimate from these data that approximately 5 ng equivalents of EGF were present in 500 μl of the wheat germ lectin eluate. In six replicate experiments, we recovered 16–40 ng equivalents of the EGF precursor/g (wet weight) of mouse kidney. Anti-EGF immune serum but not nonimmune serum inhibited the ability of the EGF precursor in the eluate to compete for binding to the fibroblasts (Table I, Exp. 1). Similarly, anti-EGF immune serum but not nonimmune serum inhibited the ability of unlabeled 6-kDa EGF to compete for binding to the fibroblasts (data not shown).

The abilities of the EGF precursor and murine EGF to support the growth of mouse keratinocytes (MKB cells) were also examined (Fig. 4, a–d). Whereas these cells do not grow in the absence of EGF (Fig. 4, a), in the presence of either 4 ng/ml mouse EGF (Fig. 4, b) or 0.5 or 1 ml of the wheat germ lectin eluate (Fig. 4, c and d) extensive growth stimulation was observed.

Processing of the EGF Precursor by the EGF-associated Arginine Esterase—Under mild conditions EGF may be isolated from the mouse submaxillary gland as a high molecular mass complex (72 kDa) consisting of two molecules of mEGF and two molecules of an arginine esterase (21). The ability of this arginine esterase to cleave the EGF precursor was examined. The EGF precursor present in the lectin eluate was incubated with the arginine esterase and the reaction mixtures were examined by SDS-gel electrophoresis and immunoblotting as described under “Materials and Methods.” It may be seen (Fig. 5, lanes A–C) that incubation of the precursor with increasing amounts of arginine esterase produced a lower molecular mass immunoreactive material (~97 kDa). No mature EGF was detected even after prolonged (4 h) incubation (data not shown). EGF, the 53-amino acid protein, has a terminal arginine as a potential site for cleavage with the arginine esterase. Since within the EGF precursor the EGF sequence is flanked by polypeptide segments of 976 and 188 residues, the cleavage at the COOH-terminal arginine residue of the EGF sequence would generate an immunoreactive fragment of the approximate molecular weight seen in Fig. 5. Additional specific proteolytic cleavages would be required for the generation of mature EGF. Its of interest that the kidney and

![FIG. 3. Immunoaffinity chromatography of the EGF precursor.](http://www.jbc.org/) The triacetylchitotriose eluate from the wheat germ lectin column (described in Fig. 2) was chromatographed on an anti-EGF Sepharose 4B column and the adsorbed proteins eluted with an acid buffer as described under “Materials and Methods.” Aliquots (30 μl) were subjected to SDS-gel electrophoresis and analyzed by immunoblotting (A) or silver staining (B). Lanes A, wheat germ lectin eluate; lanes B, nonadsorbed material; lanes C, first wash; lanes D, final wash; lanes E, acid eluate.

![TABLE I Competitive inhibition of 125I binding to human fibroblasts by the lectin-purified and arginine esterase-truncated EGF precursor](http://www.jbc.org/)

<table>
<thead>
<tr>
<th>Preincubation conditions</th>
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<tr>
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<tr>
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<tr>
<td>500 μl of w.g. eluate and</td>
<td>11,000</td>
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<tr>
<td>500 μl of esterase-treated</td>
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<tr>
<td>w.g. eluate</td>
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<td>500 μl of esterase-treated</td>
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* The results are expressed as specific binding of 125I-EGF/dis. Nonincubation binding of 125I-EGF, determined in the presence of 10 μg of unlabeled EGF, was less than 10% of total. All experiments were done in duplicate or triplicate and the average value presented. The standard deviation for each assay was less than 6% of the average value presented. The experiments were repeated 4 to 6 times with replicate preparations and essentially identical results were obtained. ^w.g. eluate, wheat germ eluate (described in Fig. 3). The antiserum or preimmune serum was added to the wheat germ lectin eluate for 1 h at room temperature prior to assay.

* The wheat germ lectin eluate was incubated with arginine esterase in a ratio of 20 μg of arginine esterase/120 μl of wheat germ eluate for 4 h at 37 °C.
FIG. 4. Growth stimulation of MKB cells by the EGF precursor. The triacetylchitotriose eluate from the wheat germ lectin column (described in Fig. 2) was passed through an Extracti-Gel column to remove traces of Triton X-100 and was added to cultures of MKB cells as described under "Materials and Methods." a, no additions; b, 4 ng/ml murine EGF; c, 0.5 ml of EGF precursor; d, 1 ml of EGF precursor.

specifically the urine are rich in kallikreins (32).

The biological activity of the EGF precursor truncated by the arginine esterase was examined by assaying its ability to compete with ¹²⁵I-labeled EGF for binding to fibroblasts. Anti-EGF immune serum but not nonimmune serum inhibited the ability of the truncated EGF precursor to compete for binding to the fibroblasts (data not shown). In replicate experiments we estimated that pretreatment with arginine esterase nearly doubled the nanogram equivalents of EGF present in the wheat germ lectin eluates, suggesting that the truncated precursor has a greater affinity for the receptor than the intact molecule. Arginine esterase, in the absence of EGF precursor, did not affect ¹²⁵I-labeled EGF binding to human fibroblasts (data not shown).

Pepsin Digestion of the EGF Precursor—Murine EGF is stable in 0.1 M HCl and is resistant to pepsin digestion; neither the apparent molecular weight nor the immunoreactivity of murine EGF is altered by digestion with pepsin. Therefore, the effect of pepsin digestion on the structure and biological activity of the EGF precursor was examined (Fig. 6 and Table II, respectively). The EGF precursor eluted from the wheat germ lectin column (Fig. 6, lane A) was unaltered by incubation in acid alone (Fig. 6, lane B). Incubations with increasing amounts of pepsin (lanes C, D, and E) resulted in the generation of lower molecular weight immunoreactive material from the EGF precursor. Immunoreactive material, with approximately the same mobility as EGF (Fig. 6, lane G) appeared following extensive pepsin digestion of the EGF precursor (Fig. 6, lanes D and E).

The biological activity of the EGF precursor digested with pepsin was examined by assaying the ability of the digest to compete with ¹²⁵I-labeled EGF for binding to intact human fibroblasts. As can be seen in Table II, treatment of murine EGF with pepsin did not destroy its ability to compete for binding. Similarly, treatment of the EGF precursor with pepsin did not destroy and indeed appeared to enhance its ability to compete for binding to the EGF receptor. Pepsin alone did not affect binding (data not shown).

² J. A. Breyer and S. Cohen, unpublished observations.
growth factor (33, 34), colony-stimulating factor-1 (35), and tumor necrosis factor (36) appear to be derived from larger precursors with structural features characteristic of transmembrane proteins. The membrane-bound precursors may simply serve to anchor the nascent growth factors to allow correct folding prior to their proteolytic release. Alternatively, it has been postulated that these membrane glycoproteins may themselves mediate cell-cell recognition events (34). The processing and release of mature EGF from its precursor would require specific proteolytic cleavages, it is possible that in the absence of specific proteases the precursor has highly localized effects, while in the presence of these proteases the mature growth factors could be released for more distant effects. Interestingly, while the mature 53-amino acid growth factor is found in some tissues (submaxillary gland) other tissues appear to contain primarily the large EGF precursor (kidney) (14, 37). It has previously been demonstrated that in stably transfected NIH 3T3 cells the EGF precursor is expressed as a membrane-bound protein with no detectable amounts of mature EGF either in the medium or cells (22).

As a first step in understanding the biological significance of preproEGF in the kidney, we have isolated and characterized the EGF precursor from this tissue. The EGF precursor from mouse kidney is indeed membrane-associated. It appears to be an integral membrane protein, since it is not released by extraction with either high salt or alkaline buffers but may be solubilized with detergents. We have concluded that the native EGF precursor from mouse kidney is glycosylated on the basis of its specific binding to and elution from wheat germ and lentil lectin columns. It is of interest that the human EGF precursor, expressed in transfected NIH 3T3 cells, also is glycosylated. These results are in agreement with the computer-based characterization of the EGF precursor that predicted several potential glycosylation sites (38).

We have taken advantage of the binding of the precursor to wheat germ lectin and to anti-EGF antiserum to purify the EGF precursor from the membrane fraction of mouse kidney. To determine whether the intact EGF precursor is biologically active, we examined the ability of the EGF precursor to bind to receptors for EGF and to stimulate the growth of responsive cells. We have demonstrated that the EGF precursor isolated from normal kidney tissue competed with 125I-labeled EGF for binding to the EGF receptor on intact human fibroblasts. Antibody inhibition of binding suggested that the EGF domain of the precursor is interacting with the receptor. It is possible that one of the EGF-like sequences is interacting and also is recognized by the antibody. Furthermore, we have demonstrated that the EGF precursor is able to stimulate mitogenesis when added to MKB cells. It has recently been reported that membranes containing the α-transforming growth factor precursor also are able to interact with the EGF receptor (34).

Although only traces of EGF (6 kDa) have been found in kidney tissue, urine contains large amounts of mature EGF (39). Several lines of evidence suggest that the source of this urinary EGF is the kidney (11, 40). The computer-based characterization of the EGF precursor predicts that the EGF moiety is in the extracellular domain immediately adjacent to the membrane-spanning region (38). EGF immunoreactivity has been detected on the luminal plasma membrane of cells of the thick ascending limb of Henle (16). The EGF in the urine may result from proteolytic cleavage of the EGF precursor on the luminal membrane. One potential processing enzyme is the arginine esterase originally isolated as a complex with EGF. We have demonstrated that the EGF precursor can be cleaved into a lower molecular weight form by incubation with this enzyme. The cleaved fragment has a
greater affinity for the EGF receptor than the intact precursor. Although this specific arginine esterase, a kallikrein enzyme, may not be present in normal mouse kidney in detectable amounts (41), the kidney and specifically the urine to which the membrane-bound precursor may be exposed is rich in many kallikrein enzymes. We have also taken advantage of the resistance of EGF to pepsin digestion to demonstrate that the pepsin digestion of the EGF precursor generates a fragment with a similar mobility to EGF and that binds to the receptor. The cleavage of the EGF precursor by arginine esterase and pepsin is the first demonstration that the EGF precursor can be proteolytically processed to generate smaller molecular weight fragments that are active in binding to the EGF receptor. The smaller molecular weight fragments have a greater apparent affinity for the receptor, perhaps due to less steric hindrance of the binding.

The kidney, in addition to synthesizing the precursor, is also a potential target organ. EGF receptors have been demonstrated in the glomeruli, proximal tubules, cortical and inner and outer medullary collecting tubules.3 Interestingly, the EGF receptors appear to be absent in the thick ascending limb of Henle cells which have been demonstrated to contain the EGF precursor mRNA and the EGF immunoreactive protein on their luminal surface (16). This may be due to down regulation or the cells which contain the EGF precursor may not have EGF receptors, thus avoiding any autocrine effects. EGF has been demonstrated to be a mitogen for a variety of cultured renal cells (43, 44) and to have important functional effects on intact glomeruli, proximal tubules, and collecting ducts (45–47). Lastly in the whole animal it has been demonstrated that there is reduced EGF precursor mRNA and decreased urinary EGF in acute renal failure (48) and that administering EGF to animals with acute renal failure will enhance renal tubule regeneration and accelerate recovery of renal function (42). All these data suggest that the kidney may be an important target organ for either EGF or the EGF precursor.

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