Acute Renal Failure: Cellular Features of Injury and Repair

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Although ischemic acute renal failure (ARF) is likely the result of many different factors, much tubule injury can be traced back to a number of specific lesions that occur at the cellular level in ischemic polarized epithelial cells. At the onset of an ischemic insult, rapid and dramatic biochemical changes in the cellular environment occur, most notably perturbation of the intracellular levels of ATP and free calcium and increases in the levels of free radicals, which lead to alterations in structural and functional cellular components characteristic of renal epithelial cells [1–7]. These alterations include a loss of tight junction integrity, disruption of actin-based microfilaments, and loss of the apical basolateral polarity of epithelial cells. The result is loss of normal renal cell function [7–12].

After acute renal ischemia, the recovery of renal tubule function is critically dependent on reestablishment of the permeability barrier, which is crucial to proper functioning of epithelial tissues such as renal tubules. After ischemic injury the formation of a functional permeability barrier, and thus of functional renal tubules, is critically dependent on the establishment of functional tight junctions. The tight junction is an apically oriented structure that functions as both the "fence" that separates apical and basolateral plasma membrane domains and the major paracellular permeability barrier (gate). It is not yet clear how the kidney restores tight junction structure and function after ischemic injury. In fact, tight junction assembly under normal physiological conditions remains ill-understood; however, utilization of the

CHAPTER 16

"calcium switch" model with cultured renal epithelial cells has helped to elucidate some of the critical features of tight junction bioassembly. In this model for tight junction reassembly, signaling events involving G proteins, protein kinase C, and calcium appear necessary for the reestablishment of tight junctions [13–19]. Tight junction injury and recovery, like that which occurs after ischemia and reperfusion, has similarly been modeled by subjecting cultured renal epithelial cells to ATP depletion ("chemical anoxia") followed by repletion. While there are many similarities to the calcium switch, biochemical studies have recently revealed major differences, for example, in the way tight junction proteins interact with the cytoskeleton [12]. Thus, important insights into the basic and applied biology of tight junctions are likely to be forthcoming from further analysis of the ATP depletion-repletion model. Nevertheless, it is likely that, as in the calcium switch model, tight junction reassembly is regulated by classical signaling pathways that might potentially be pharmacologically modulated to enhance recovery after ischemic insults.

More prolonged insults can lead to greater, but still sublethal, injury. Key cellular proteins begin to break down. Many of these (*eg*, the tight junction protein, occludin, and the adherens junction protein, E-cadherin) are membrane proteins. Matrix proteins and their integrin receptors may need to be resynthesized, along with growth factors and cytokines, all of which pass through the endoplasmic reticulum (ER). The rate-limiting events in the biosynthesis and assembly of these proteins occur in the ER and are catalyzed by a set of ER-specific molecular chaperones, some of which are homologs of the cytosolic heat-shock proteins [20]. The levels of mRNAs for these proteins may increase 10-fold or more in the ischemic kidney, to keep up with the cellular need to synthesize and transport these new membrane proteins, as well as secreted ones.

If the ischemic insult is sufficiently severe, cell death and/or detachment leads to loss of cells from the epithelium lining the kidney tubules. To recover from such a severe insult, cell regeneration, differentiation, and possibly morphogenesis, are necessary. To a limited extent, the recovery of kidney tubule function after such a severe ischemic insult can be viewed as a recapitulation of various steps in renal development. Cells must proliferate and differentiate, and, in fact, activation of growth factor–mediated signaling pathways (some of the same ones involved in kidney development) appears necessary to ameliorate renal recovery after acute ischemic injury [21–30].

The Ischemic Epithelial Cell

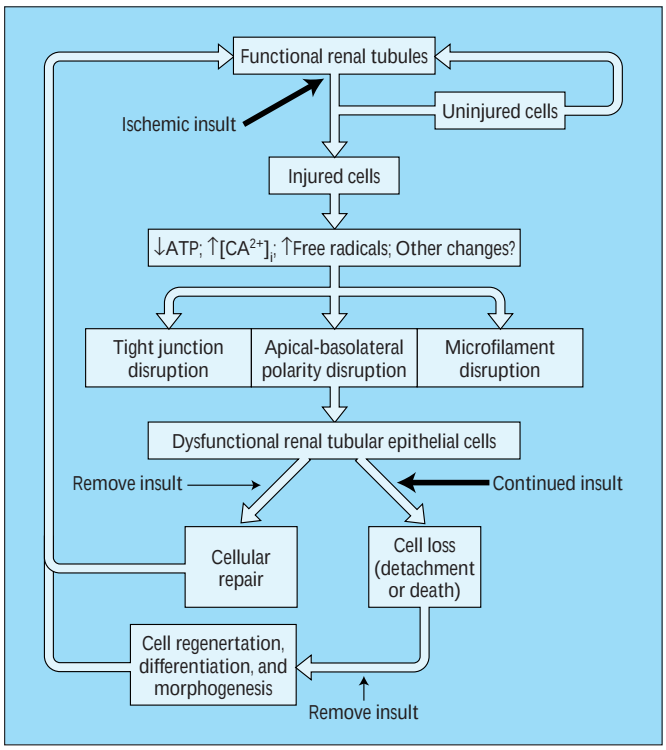

FIGURE 16-1

Ischemic acute renal failure (ARF). Flow chart illustrates the cellular basis of ischemic ARF. As described above, renal tubule epithelial cells undergo a variety of biochemical and structural changes in response to ischemic insult. If the duration of the insult is sufficiently short, these alterations are readily reversible, but if the insult continues it ultimately leads to cell detachment and/or cell death. Interestingly, unlike other organs in which ischemic injury often leads to permanent cell loss, a kidney severely damaged by ischemia can regenerate and replace lost epithelial cells to restore renal tubular function virtually completely, although it remains unclear how this happens.

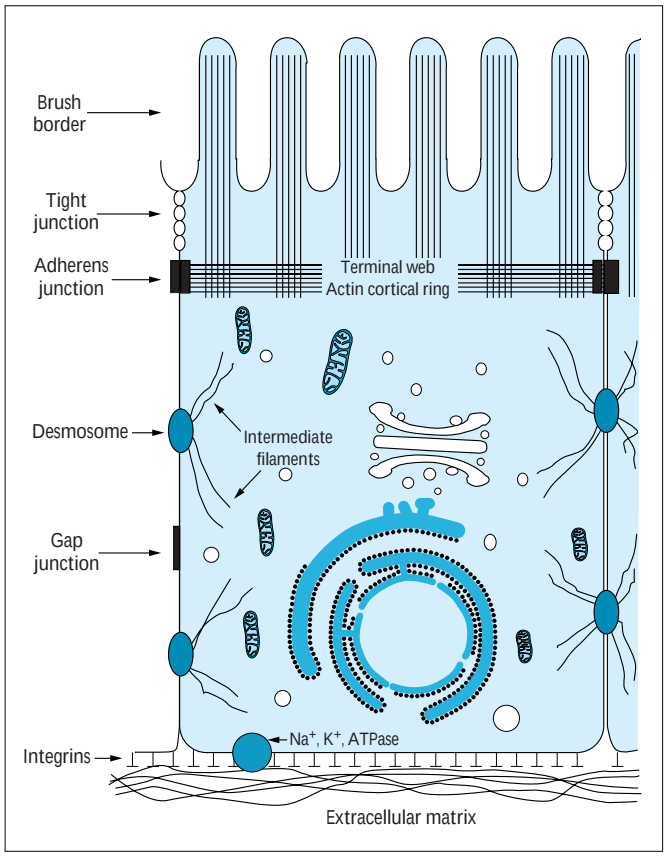

FIGURE 16-2

Typical renal epithelial cell. Diagram of a typical renal epithelial cell. Sublethal injury to polarized epithelial cells leads to multiple lesions, including loss of the permeability barrier and apical-basolateral polarity [7–12]. To recover, cells must reestablish intercellular junctions and repolarize to form distinct apical and basolateral domains characteristic of functional renal epithelial cells. These junctions include those necessary for maintaining the permeability barrier (*ie*, tight junctions), maintaining cell-cell contact (*ie*, adherens junctions and desmosomes), and those involved in cell-cell communication (*ie*, gap junctions). In addition, the cell must establish and maintain contact with the basement membrane through its integrin receptors. Thus, to understand how kidney cells recover from sublethal ischemic injury it is necessary to understand how renal epithelial cells form these junctions. Furthermore, after lethal injury to tubule cells new cells may have to replace those lost during the ischemic insult, and these new cells must differentiate into epithelial cells to restore proper function to the tubules.

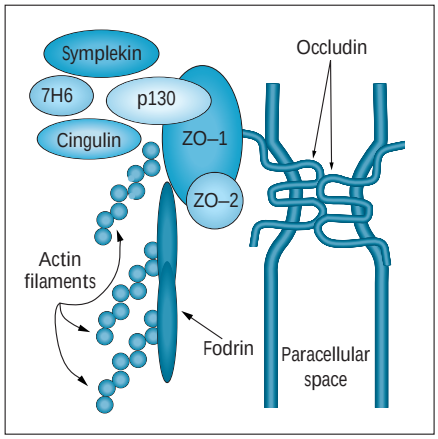

FIGURE 16-3

The tight junction. The tight junction, the most apical component of the junctional complex of epithelial cells, serves two main functions in epithelial cells: 1) It separates the apical and basolateral plasma membrane domains of the cells, allowing for vectorial transport of ions and molecules; 2) it provides the major framework for the paracellular permeability barrier, allowing for generation of chemical and electrical gradients [31]. These functions are critically important to the proper functioning of renal tubules. The tight junction is comprised of a number of proteins (cytoplasmic and transmembrane) that interact with a similar group of proteins between adjacent cells to form the permeability barrier [16, 32–37]. These proteins include the transmembrane protein occludin [35, 38] and the cytosolic proteins zonula occludens 1 (ZO-1), ZO-2 [36], p130, [39], cingulin [33, 40], 7H6 antigen [34] and symplekin [41], although other as yet unidentified components likely exist. The tight junction also appears to interact with the actin-based cytoskeleton, probably in part through ZO-1–fodrin interactions.

Reassembly of the Permeability Barrier

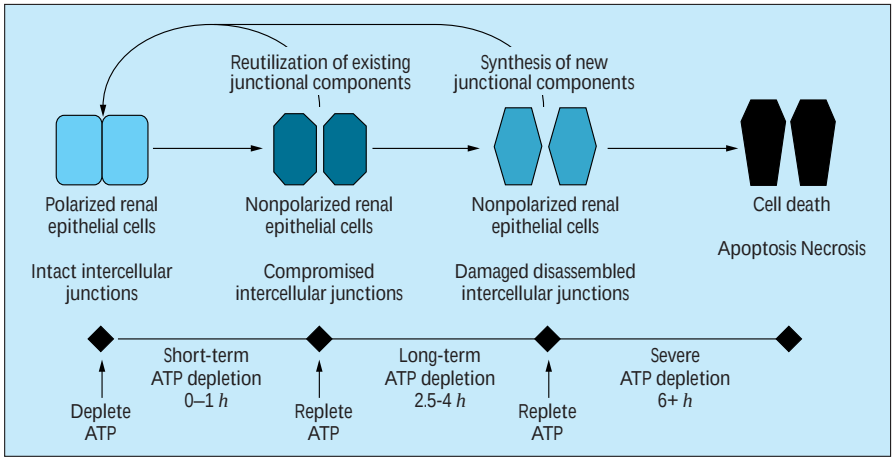

FIGURE 16-4

Cell culture models of tight junction disruption and reassembly. The disruption of the permeability barrier, mediated by the tight junction, is a key lesion in the pathogenesis of tubular dysfunction after ischemia and reperfusion. Cell culture models employing ATP depletion and repletion protocols are a commonly used approach for understanding the molecular mechanisms underlying tight junction dysfunction in ischemia and how tight junction integrity recovers after the insult [6, 12, 42]. After short-term ATP depletion (1 hour or less) in Madin-Darby canine kidney cells, although some new synthesis probably occurs, by and large it appears that reassembly of the tight junction can proceed with existing (disassembled) components after ATP repletion. This model of short-term ATP depletion-repletion is probably most relevant to transient sublethal ischemic injury of renal tubule cells. However, in a model of longterm ATP depletion (2.5 to 4 hours), that probably is most relevant to prolonged ischemic (though still sublethal) insult to the renal tubule, it is likely that reestablishment of the permeability barrier (and thus of tubule function) depends on the production (message and protein) and bioassembly of new tight junction components. Many of these components (membrane proteins) are assembled in the endoplasmic reticulum.

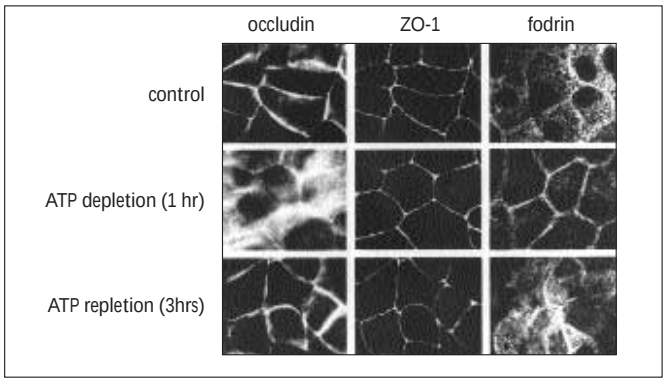

FIGURE 16-5

Immunofluorescent localization of proteins of the tight junction after ATP depletion and repletion. The cytosolic protein zonula occludens 1 (ZO-1), and the transmembrane protein occludin are integral components of the tight junction that are intimately associated at the apical border of epithelial cells. This is demonstrated here by indirect immunofluorescent localization of these two proteins in normal kidney epithelial cells. After 1 hour of ATP depletion this association appears to change, occludin can be found in the cell interior, whereas ZO-1 remains at the apical border of the plasma membrane. Interestingly, the intracellular distribution of the actin-cytoskeletal–associated protein fodrin also changes after ATP depletion. Fodrin moves from a random, intracellular distribution and appears to become co-localized with ZO-1 at the apical border of the plasma membrane. These changes are completely reversible after ATP repletion. These findings suggest that disruption of the permeability barrier could be due, at least in part, to altered association of ZO-1 with occludin. In addition, the apparent co-localization of ZO-1 and fodrin at the level of the tight junction suggests that ZO-1 is becoming intimately associated with the cytoskeleton.

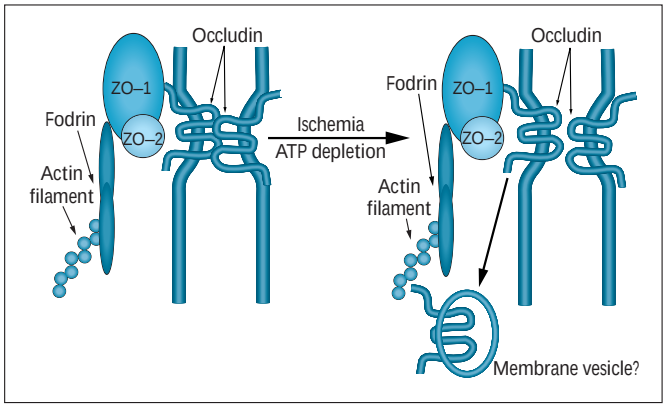

FIGURE 16-6

ATP depletion causes disruption of tight junctions. Diagram of the changes induced in tight junction structure by ATP depletion. ATP depletion causes the cytoplasmic tight junction proteins zonula occludens 1 (ZO-1) and ZO-2 to form large insoluble complexes, probably in association with the cytoskeletal protein fodrin [12], though aggregation may also be significant. Furthermore, occludin, the transmembrane protein of the tight junction, becomes localized to the cell interior, probably in membrane vesicles. These kinds of studies have begun to provide insight into the biochemical basis of tight junction disruption after ATP depletion, although how the tight junction reassembles during recovery of epithelial cells from ischemic injury remains unclear.

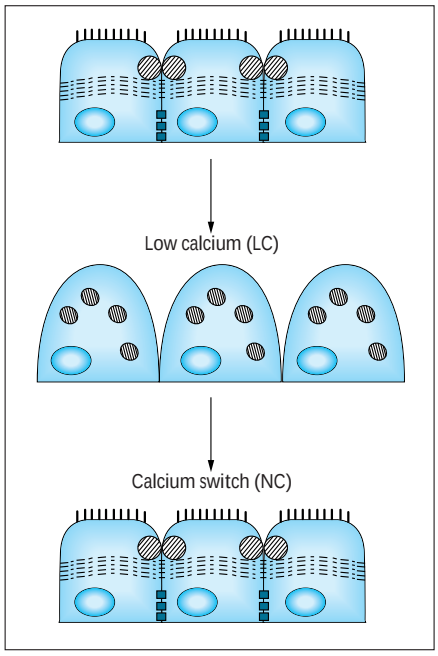

FIGURE 16-7

Madin-Darby canine kidney (MDCK) cell calcium switch. Insight into the molecular mechanisms involved in the assembly of tight junctions (that may be at least partly applicable to the ischemia-reperfusion setting) has been gained from the MDCK cell calcium switch model [43]. MDCK cells plated on a permeable support form a monolayer with all the characteristics of a tight, polarized transporting epithelium. Exposing such cell monolayers to conditions of low extracellular calcium (less than 5μM) causes the cells to lose cell-cell contact and to "round up." Upon switching back to normal calcium media (1.8 mM), the cells reestablish cell-cell contact, intercellular junctions, and apical-basolateral polarity. These events are accompanied by profound changes in cell shape and reorganization of the actin cytoskeleton. (*From* Denker and Nigam [19]; with permission)

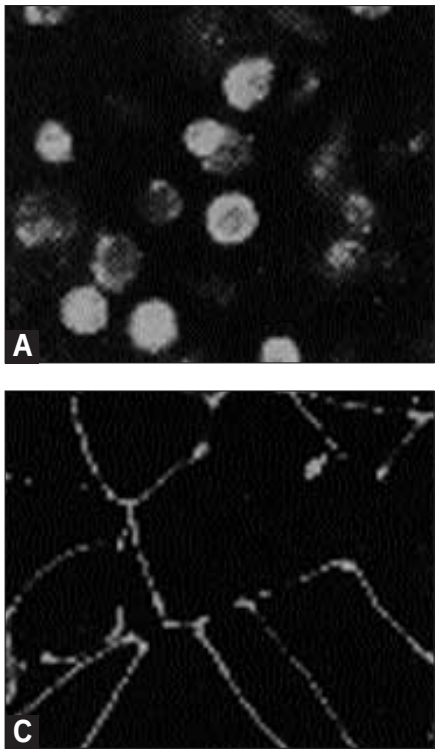

FIGURE 16-8

Protein kinase C (PKC) is important for tight junction assembly. Immunofluorescent localization of the tight junction protein zonula occludens 1 (ZO-1) during the Madin-Darby canine kidney (MDCK) cell calcium switch. In low-calcium media MDCK cells are round and have little cell-cell contact. Under these conditions, ZO-1 is found in the cell interior and has little, if any, membrane staining, **A**. After 2 hours incubation in normal calcium media, MDCK cells undergo significant changes in cell shape and make extensive cell-cell contact along the lateral portions of the plasma membrane. **B**, Here, ZO-1 has redistributed to areas of cell-cell contact with little apparent intracellular staining. This process is blocked by treatment with either 500 nM calphostin C, **C**, or 25μM H7, **D**, inhibitors of PKC. These results suggest that PKC plays a role in regulating tight junction assembly. Similar studies have demonstrated roles for a number of other signaling molecules, including calcium and G proteins, in the assembly of tight junctions [12, 13, 16–19, 37, 44–46]. An analogous set of signaling events is likely responsible for tight junction reassembly after ischemia. (*From* Stuart and Nigam [16]; with permission.)

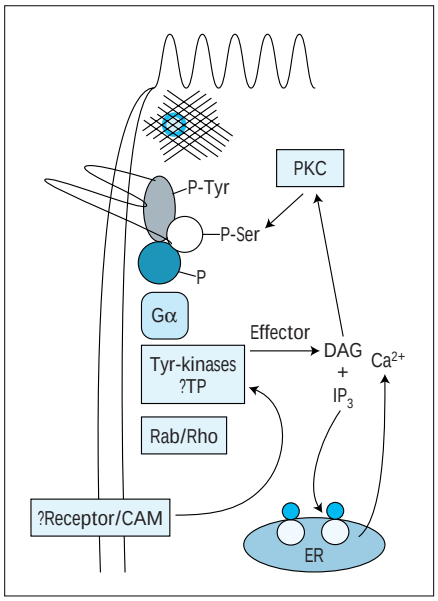

FIGURE 16-9

Signalling molecules that may be involved in tight junction assembly. Model of the potential signaling events involved in tight junction assembly. Tight junction assembly probably depends on a complex interplay of several signaling molecules, including protein kinase C (PKC), calcium (Ca^{2+}), heterotrimeric G proteins, small guanodine triphosphatases (Rab/Rho), and tyrosine kinases [13–16, 18, 37, 44–53]. Although it is not clear how this process is initiated, it depends on cell-cell contact and involves wide-scale changes in levels of intracellular free calcium. Receptor/CAM—cell adhesion molecule; DAG—diacylglycerol; ER—endoplasmic reticulum; Gα—alpha subunit of GTP-binding protein; IP3—inositol trisphosphate. (*From* Denker and Nigam [19]; with permission.)

The Endoplasmic Reticulum Stress Response in Ischemia

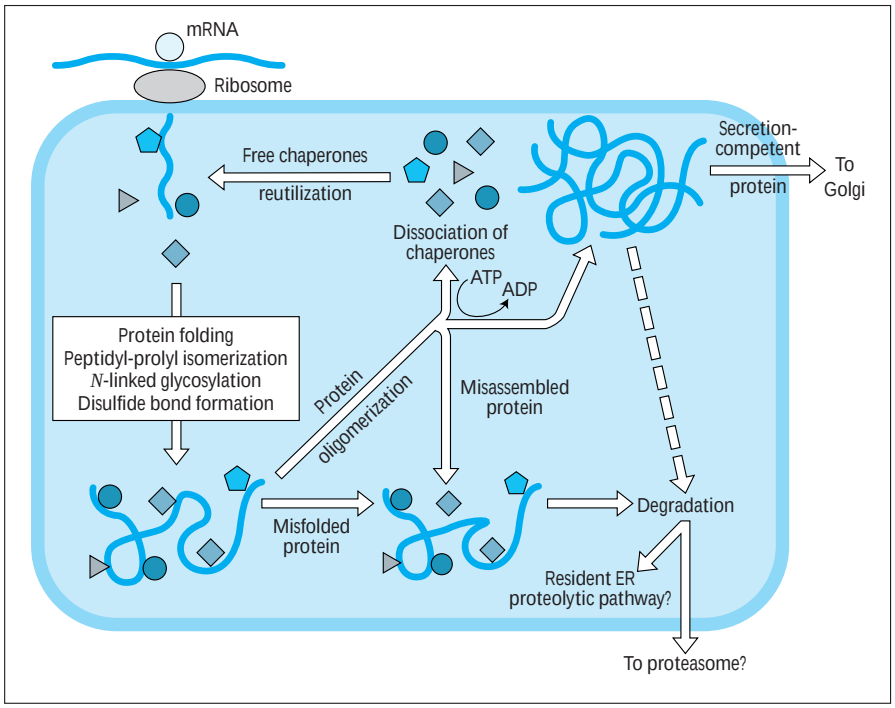

FIGURE 16-10

Protein processing in the endoplasmic reticulum (ER). To recover from serious injury, cells must synthesize and assemble new membrane (tight junction proteins) and secreted (growth factors) proteins. The ER is the initial site of synthesis of all membrane and secreted proteins. As a protein is translocated into the lumen of the ER it begins to interact with a group of resident ER proteins called molecular chaperones [20, 54–57]. Molecular chaperones bind transiently to and interact with these nascent polypeptides as they fold, assemble, and oligomerize [20, 54, 58]. Upon successful completion of folding or assembly, the molecular chaperones and the secretion-competent protein part company via a reaction that requires ATP hydrolysis, and the chaperones are ready for another round of protein folding [20, 59–61]. If a protein is recognized as being misfolded or misassembled it is retained within the ER via stable association with the molecular chaperones and is ultimately targeted for degradation [62]. Interestingly, some of the more characteristic features of epithelial ischemia include loss of cellular functions mediated by proteins that are folded and assembled in the ER (*ie*, cell adhesion molecules, integrins, tight junctional proteins, transporters). This suggests that proper functioning of the protein-folding machinery of the ER could be critically important to the ability of epithelial cells to withstand and recover from ischemic insult. ADP—adenosine diphosphate.

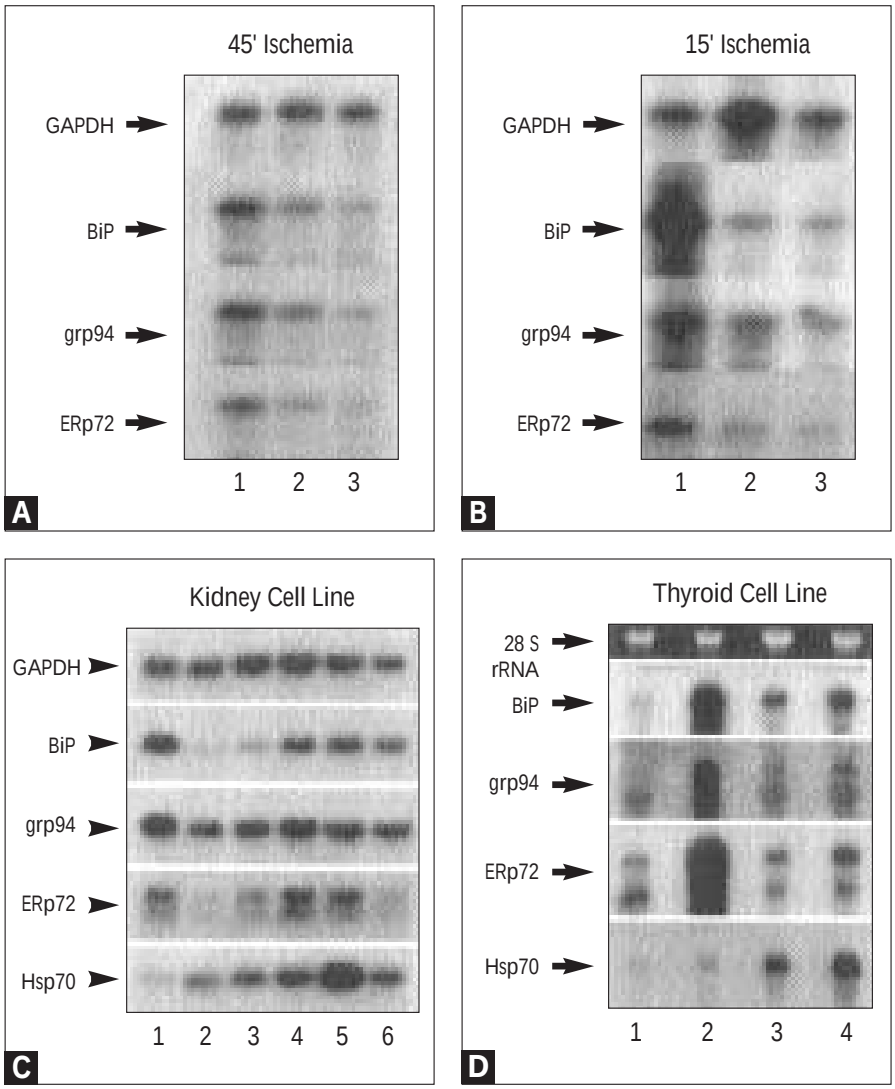

FIGURE 16-11

Ischemia upregulates endoplasmic reticulum (ER) molecular chaperones. Molecular chaperones of the ER are believed to function normally to prevent inappropriate intra- or intermolecular interactions during the folding and assembly of proteins [20, 54]. However, ER molecular chaperones are also part of the "quality control" apparatus involved in the recognition, retention, and degradation of proteins that fail to fold or assemble properly as they transit the ER [20, 54]. In fact, the messages encoding the ER molecular chaperones are known to increase in response to intraorganelle accumulation of such malfolded proteins [11, 20, 54, 55]. Here, Northern blot analysis of total RNA from either whole kidney or cultured epithelial cells demonstrates that ischemia or ATP depletion induces the mRNAs that encode the ER molecular chaperones, including immunoglobulin binding protein (BiP), 94 kDa glucose regulated protein (grp94), and 72 kDa endoplasmic reticulum protein (Erp72) [11]. This suggests not only that ischemia or ATP depletion causes the accumulation of malfolded proteins in the ER but that a major effect of ischemia and ATP depletion could be perturbation of the "folding environment" of the ER and disruption of protein processing. GAPDH—glyceraldehyde-3-phosphate dehydrogenase; Hsp70—70 kDa heat-shock protein. (*From* Kuznetsov *et al.* [11]; with permission.)

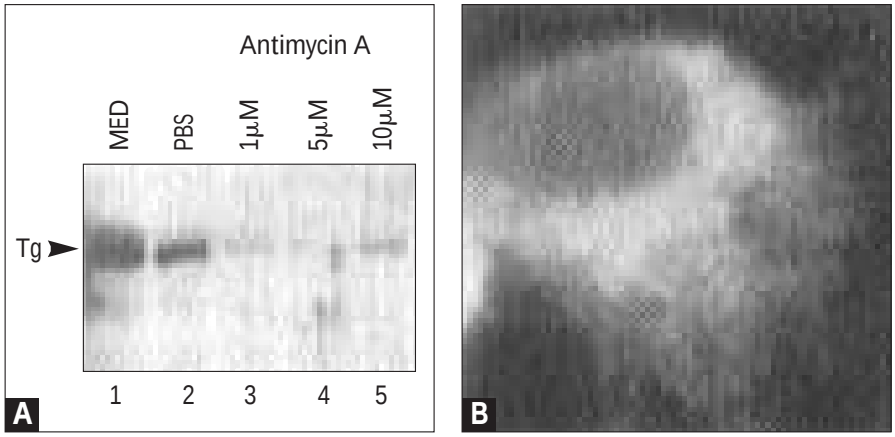

FIGURE 16-12

ATP depletion perturbs normal endoplasmic reticulum (ER) function. Because ATP and a proper redox environment are necessary for folding and assembly [20, 54, 63, 64] and ATP depletion alters ATP levels and the redox environment, the secretion of proteins is perturbed under these conditions. Here, Western blot analysis of the culture media from thyroid epithelial cells subjected to ATP depletion (*ie*, treatment with antimycin A, an inhibitor of oxidative phosphorylation) illustrates this point. **A**, Treatment with as little as 1μM antimycin A for 1 hour completely blocks the secretion of thyroglobulin (Tg) from these cells.

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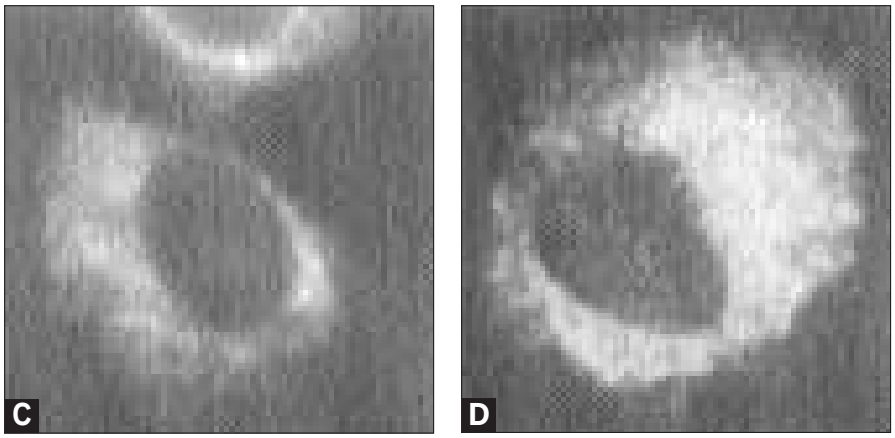


FIGURE 16-12 *(Continued)*

B–D, Moreover, indirect immunofluorescence with antithyroglobulin antibody demonstrates that the nonsecreted protein is trapped almost entirely in the ER. Together with data from Northern blot analysis, this suggests that perturbation of ER function and disruption of the secretory pathway is likely to be a key cellular lesion in ischemia [11]. MED—control media; PBS—phosphate-buffered saline. (*From* Kuznetsov *et al*. [11]; with permission.)

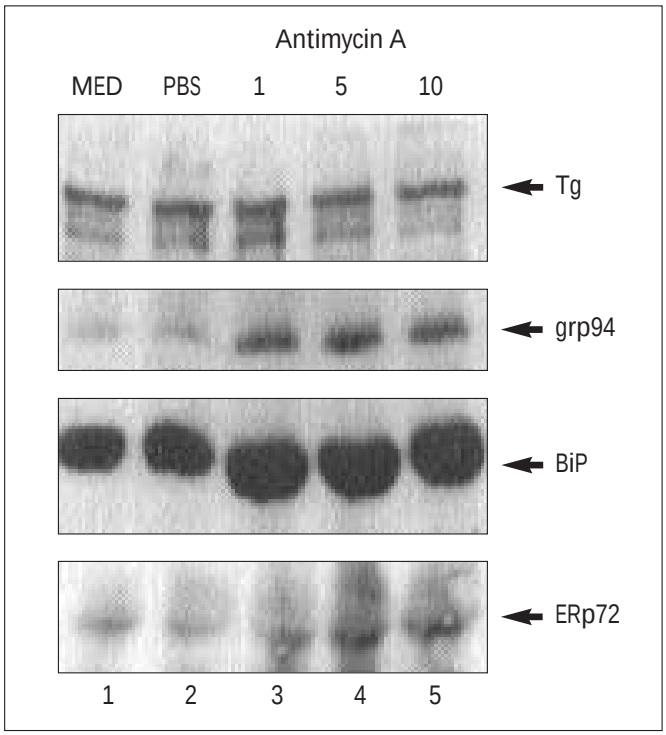
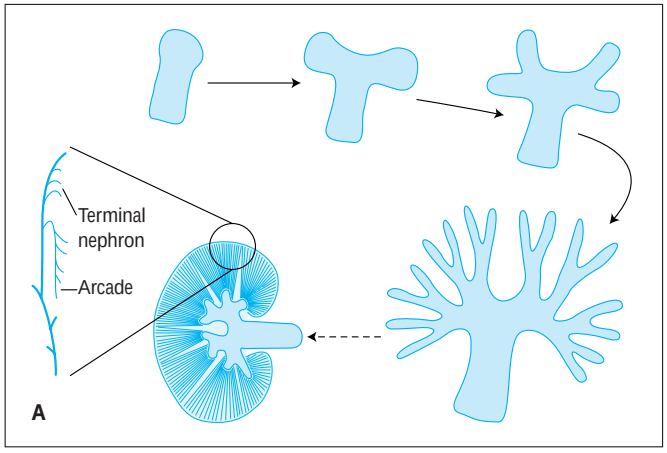
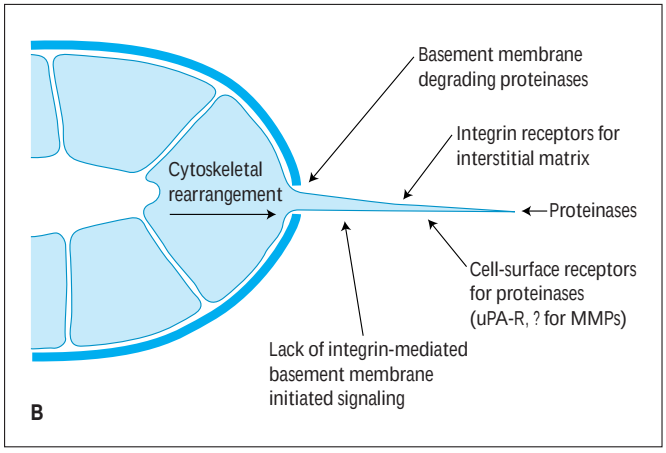


FIGURE 16-13

ATP depletion increases the stability of chaperone-folding polypeptide interactions in the endoplasmic reticulum (ER). Immunoglobulin binding protein (BiP), and perhaps other ER molecular chaperones, associate with nascent polypeptides as they are folded and assembled in ER [20, 54, 56, 57, 65–73]. The dissociation of these proteins requires hydrolysis of ATP [69]. Thus, when levels of ATP drop, BiP should not dissociate from the secretory proteins and the normally transient interaction should become more stable. Here, the associations of ER molecular chaperones with a model ER secretory protein is examined by Western blot analysis of thyroglobulin (Tg) immunoprecipitates from thyroid cells subjected to ATP depletion. After treatment with antimycin A, there is an increase in the amounts of ER molecular chaperones (BiP, grp94 and ERP72) which co-immunoprecipitate with antithyroglobulin antibody [11], suggesting that ATP depletion causes stabilization of the interactions between molecular chaperones and secretory proteins folded and assembled in the ER. Moreover, because a number of proteins critical to the proper functioning of polarized epithelial cells (*ie*, occludin, E-cadherin, Na-K-ATPase) are folded and assembled in the ER, this suggests that recovery from ischemic injury is likely to depend, at least in part, on the ability of the cell to rescue the protein-folding and -assembly apparatus of the ER. Control media (MED) and phosphate buffered saline (PBS)—no ATP depletion; 1, 5, 10μM antimycin A—ATP-depleting conditions. (*From* Kuznetsov *et al*. [11]; with permission.)

Growth Factors and Morphogenesis





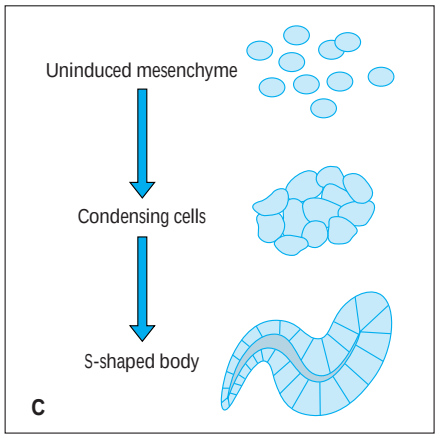


FIGURE 16-14

Kidney morphogenesis. Schematics demonstrate the development of the ureteric bud and metanephric mesenchyme during kidney organogenesis. During embryogenesis, mutual inductive events between the metanephric mesenchyme and the ureteric bud give rise to primordial structures that differentiate and fuse to form functional nephrons [74-76]. Although the process has been described morphologically, the nature and identity of molecules involved in the signaling and regulation of these events remain unclear. **A**, Diagram of branching tubulogenesis of the ureteric bud during kidney organogenesis. The ureteric bud is induced by the metanephric mesenchyme to branch and elongate to form the urinary collecting system [74-76]. **B**, Model of cellular events involved in ureteric bud branching. To branch and elongate, the ureteric bud must digest its way through its own basement membrane, a highly complicated complex of extracellular matrix proteins. It is believed that this is accomplished by cellular projections, "invadopodia," which allow for localized sites of proteolytic activity at their tips [77-81]. **C**, Mesenchymal cell compaction. The metanephric mesenchyme not only induces ureteric bud branching but is also induced by the ureteric bud to epithelialize and differentiate into the proximal through distal tubule [74–76]. (*From* Stuart and Nigam [80] and Stuart *et al*. [81]; with permission.)

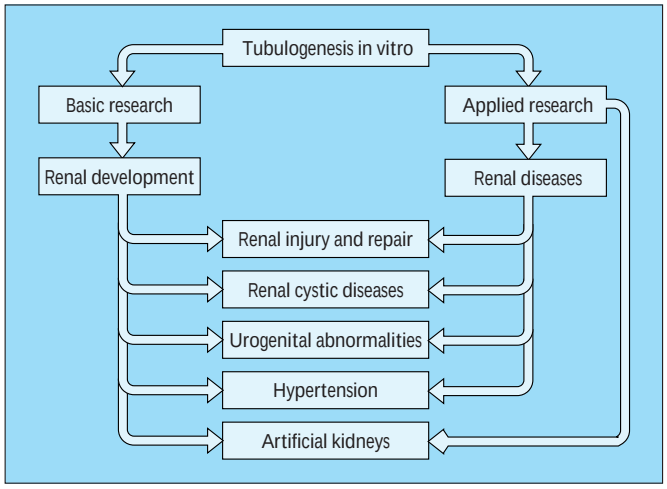


FIGURE 16-15

Potential of in vitro tubulogenesis research. Flow chart indicates relevance of in vitro models of kidney epithelial cell branching tubulogenesis to basic and applied areas of kidney research. While results from such studies provide critical insight into kidney development, this model system might also contribute to the elucidation of mechanisms involved in kidney injury and repair for a number of diseases, including tubular epithelial cell regeneration secondary to acute renal failure. Moreover, these models of branching tubulogenesis could lead to therapies that utilize tubular engineering as artificial renal replacement therapy [82].

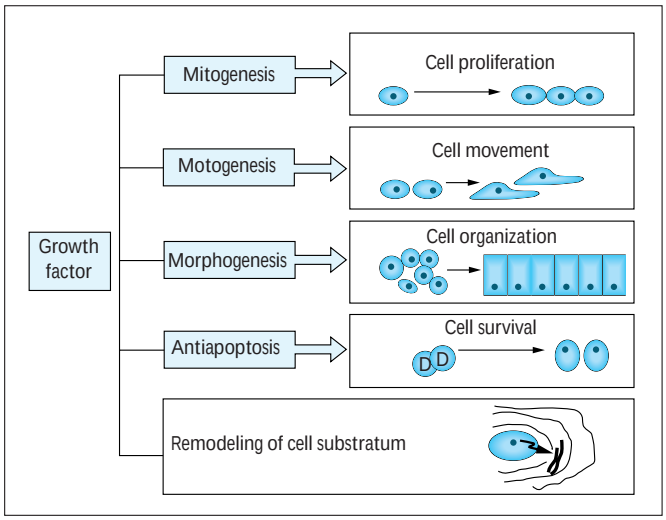


FIGURE 16-16

Cellular response to growth factors. Schematic representation of the pleiotrophic effects of growth factors, which share several properties and are believed to be important in the development and morphogenesis of organs and tissues, such as those of the kidney. Among these properties are the ability to regulate or activate numerous cellular signaling responses, including proliferation (mitogenesis), motility (motogenesis), and differentiation (morphogenesis). These characteristics allow growth factors to play critical roles in a number of complex biological functions, including embryogenesis, angiogenesis, tissue regeneration, and malignant transformation [83].

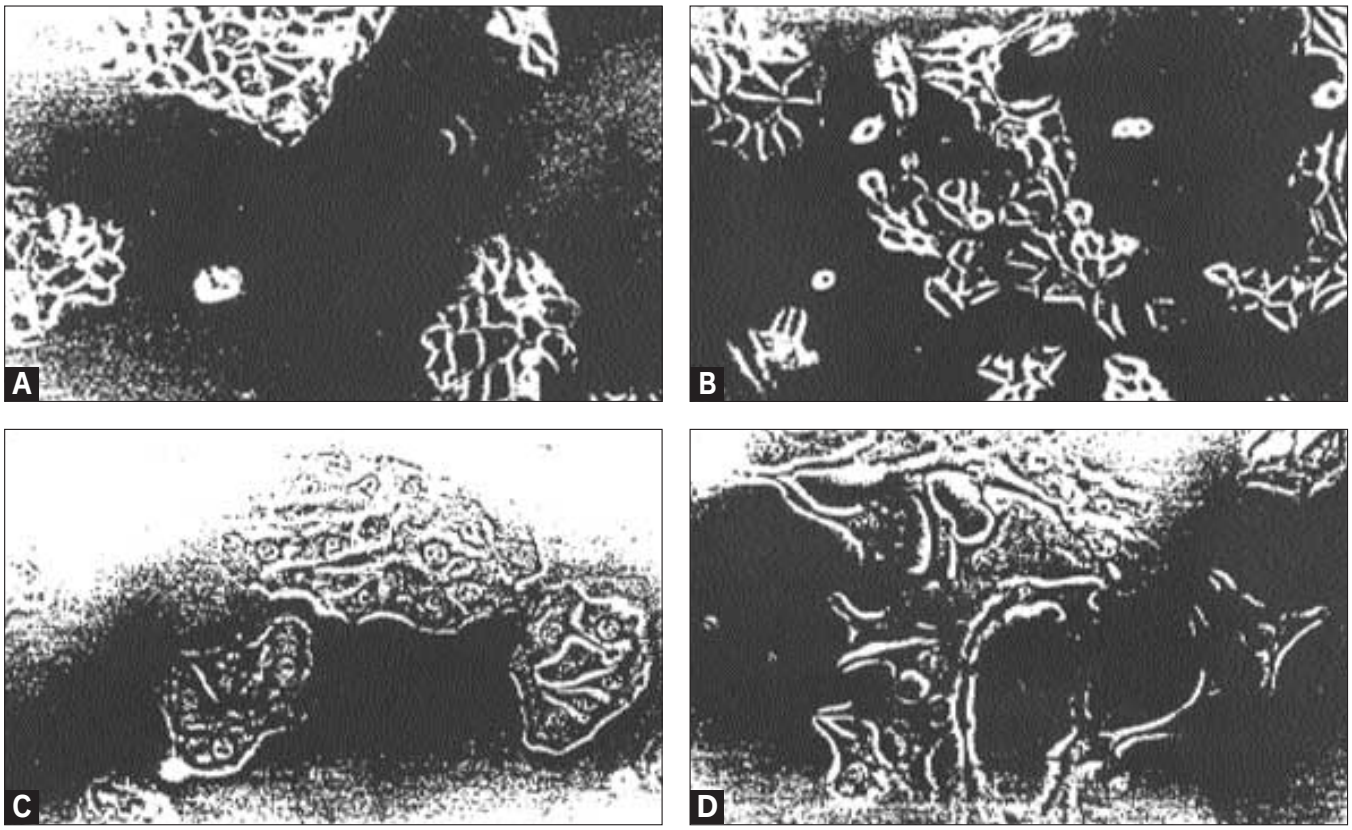


FIGURE 16-17

Motogenic effect of growth factors—hepatocyte growth factor (HGF) induces cell "scattering." During development or regeneration the recruitment of cells to areas of new growth is vital. Growth factors have the ability to induce cell movement. Here, subconfluent monolayers of either Madin-Darby canine kidney (MDCK) **C, D**, or murine inner medullary collecting duct (mIMCD) **A, B**, cells were grown for 24 hours in the absence, **A, C**, or presence **B, D**, of 20 ng/mL HGF. Treatment of either type of cultured renal epithelial cell with HGF induced the dissociation of islands of cells into individual cells. This phenomenon is referred to as scattering. HGF was originally identified as *scatter factor*, based on its ability to induce the scattering of MDCK cells [83]. Now, it is known that HGF and its receptor, the transmembrane tyrosine kinase *c-met*, play important roles in development, regeneration, and carcinogenesis [83]. (*From* Cantley *et al.* [84]; with permission.)

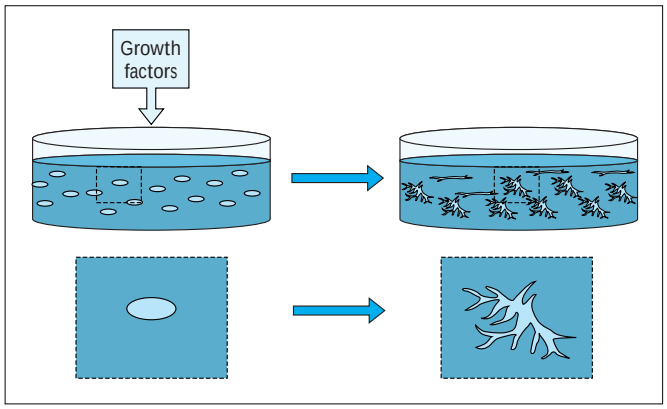


FIGURE 16-18

Three-dimensional extracellular matrix gel tubulogenesis model. Model of the three-dimensional gel culture system used to study the branching and tubulogenesis of renal epithelial cells. Analyzing the role of single factors (*ie*, extracellular matrix, growth factors, cell-signaling processes) involved in ureteric bud branching tubulogenesis in the context of the developing embryonic kidney is an extremely daunting task, but a number of model systems have been devised that allow for such investigation [77, 79, 85]. The simplest model exploits the ability of isolated kidney epithelial cells suspended in gels composed of extracellular matrix proteins to form branching tubular structures in response to growth factors. For example, Madin-Darby canine kidney (MDCK) cells suspended in gels of type I collagen undergo branching tubulogenesis reminiscent of ureteric bud branching morphogenesis in vivo [77, 79]. Although the results obtained from such studies in vitro might not correlate directly with events in vivo, this simple, straightforward system allows one to easily manipulate individual components (*eg*, growth factors, extracellular matrix components) involved in the generation of branching epithelial tubules and has provided crucial insights into the potential roles that these various factors play in epithelial cell branching morphogenesis [77, 79, 84–87].

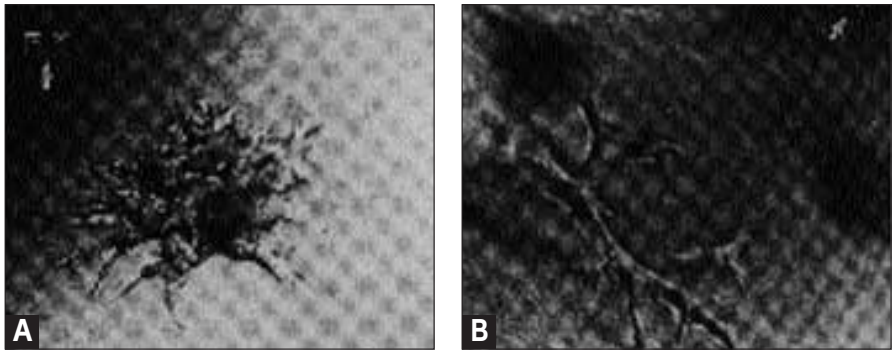


FIGURE 16-19

An example of the branching tubulogenesis of renal epithelial cells cultured in three-dimensional extracellular matrix gels. Microdissected mouse embryonic kidneys (11.5 to 12.5 days) were cocultured with **A**, murine inner medullary collecting duct (mIMCD) or, **B**, Madin-Darby canine kidney (MDCK) cells suspended in gels of rat-tail collagen (type I). Embryonic kidneys (EK) induced the formation of branching tubular structures in both mIMCD and MDCK cells after 48 hours of incubation at 37°C. EKs produce a number of growth factors, including hepatocyte growth factor, transforming growth factor-alpha, insulin-like growth factor, and transforming growth factor–β, which have been shown to effect tubulogenic activity [86–93]. Interestingly, many of these same growth factors have been shown to be effective in the recovery of renal function after acute ischemic insult [21–30]. (*From* Barros *et al.* [87]; with permission.)

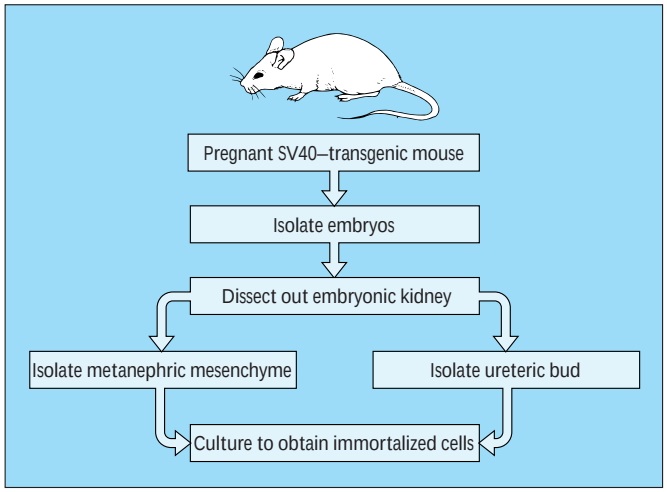


FIGURE 16-20

Development of cell lines derived from embryonic kidney. Flow chart of the establishment of ureteric bud and metanephric mesenchymal cell lines from day 11.5 mouse embryo. Although the results obtained from the analysis of kidney epithelial cells—Madin-Darby canine kidney (MDCK) or murine inner medullary collecting duct (mIMCD) seeded in three-dimensional extracellular matrix gels has been invaluable in furthering our understanding of the mechanisms of epithelial cell branching tubulogenesis, questions can be raised about the applicability to embryonic development of results using cells derived from terminally differentiated adult kidney epithelial cells [94]. Therefore, kidney epithelial cell lines have been established that appear to be derived from the ureteric bud and metanephric mesenchyme of the developing embryonic kidney of SV-40 transgenic mice [94, 95]. These mice have been used to establish a variety of "immortal" cell lines.

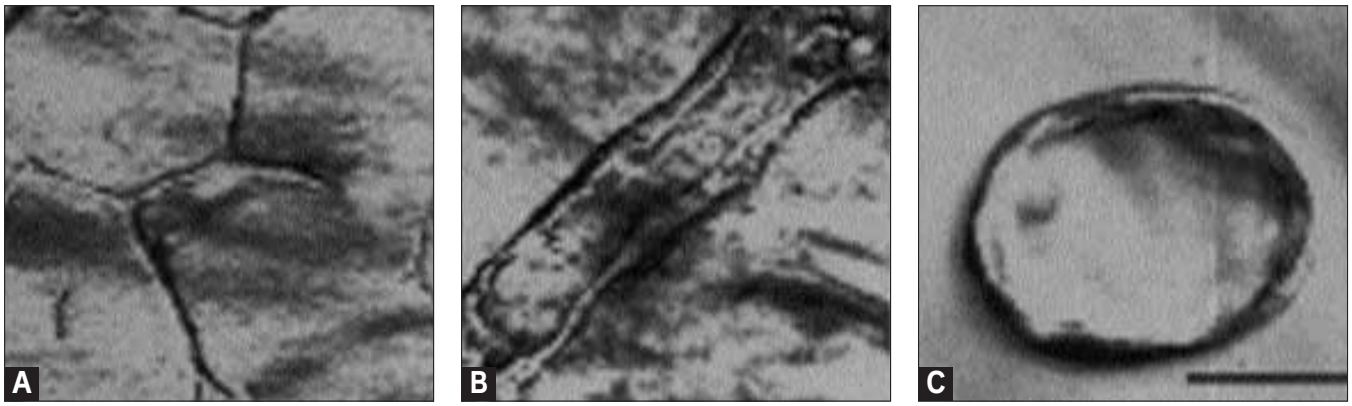


FIGURE 16-21

Ureteric bud cells undergo branching tubulogenesis in three-dimensional extracellular matrix gels. Cell line derived from ureteric bud (UB) and metanephric mesenchyme from day 11.5 mouse embryonic kidney undergo branching tubulogenesis in three-dimensional extracellular matrix gels. Here, UB cells have been induced to form branching tubular structures in response to "conditioned" media collected from the culture of metanephric mesenchymal cells. During normal kidney morphogenesis, these two embryonic cell types undergo a mutually inductive process that ultimately leads to the formation of functional nephrons [74–76]. This model system illustrates this process, ureteric bud cells being induced by factors secreted from metanephric mesenchymal cells. Thus, this system could represent the simplest in vitro model with the greatest relevance to early kidney development [94]. **A**, UB cells grown for 1 week in the presence of conditioned media collected from cells cultured from the metanephric mesenchyme. Note the formation of multicellular cords. **B**, After 2 weeks' growth under the same conditions, UB cells have formed more substantial tubules, now with clear lumens. **C**, Interestingly, after 2 weeks of culture in a three-dimensional gel composed entirely of growth factor–reduced Matrigel, ureteric bud cells have not formed cords or tubules, only multicellular cysts. Thus, changing the matrix composition can alter the morphology from tubules to cysts, indicating that this model might also be relevant to renal cystic disease, much of which is of developmental origin. (*From* Sakurai *et al.* [94]; with permission.)

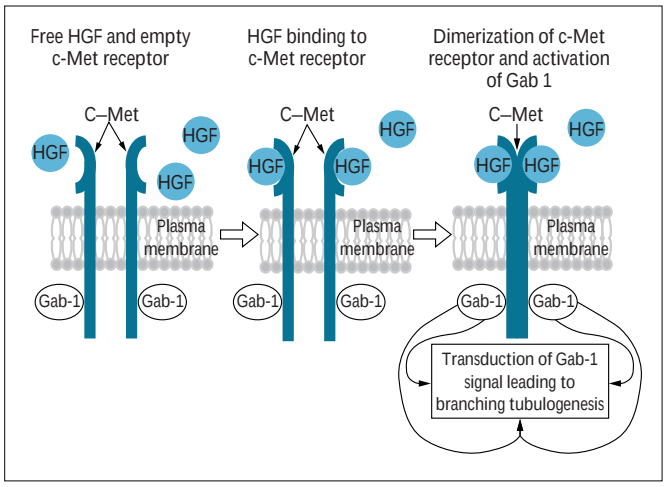


FIGURE 16-22

Signalling pathway of hepatocyte growth factor action. Diagram of the proposed intracellular signaling pathway involved in hepatocyte growth factor (HGF)–mediated tubulogenesis. Although HGF is perhaps the best-characterized of the growth factors involved in epithelial cell-branching tubulogenesis, very little of its mechanism of action is understood. However, recent evidence has shown that the HGF receptor (c-Met) is associated with Gab-1, a docking protein believed to be involved in signal transduction [96]. Thus, on binding to c-Met, HGF activates Gab-1–mediated signal transduction, which, by an unknown mechanism, affects changes in cell shape and cell movement or cell-cell–cell-matrix interactions. Ultimately, these alterations lead to epithelial cell–branching tubulogenesis.

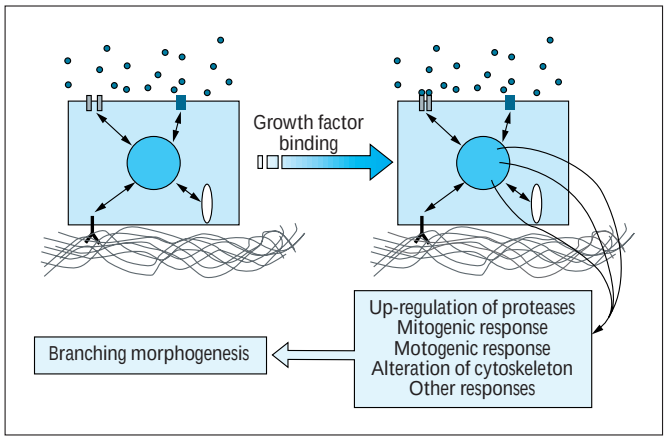


FIGURE 16-23

Mechanism of growth factor action. Proposed model for the generalized response of epithelial cells to growth factors, which the depends on their environment. Epithelial cells constantly monitor their surrounding environment via extracellular receptors (*ie*, integrin receptors) and respond accordingly to growth factor stimulation. If the cells are in the appropriate environment, growth factor binding induces cellular responses necessary for branching tubulogenesis. There are increases in the levels of extracellular proteinases and of structural and functional changes in the cytoarchitecture that enable the cells to form branching tubule structures.

GROWTH FACTORS IN DEVELOPMENTAL AND RENAL RECOVERY

Growth Factor	Expression Following Renal Ischemia	Effect of Exogenous Administration	Branching/Tubulogenic Activity
HGF	Increased [97]	Enhanced recovery [103]	Facilatory [109,110]
EGF	Unclear [98,99]	Enhanced recovery [104,105]	Facilatory [111]
HB-EGF	Increased [100]	Undetermined	Facilatory [111]
TGF-α	Unclear	Enhanced recovery [106]	Facilatory [111]
IGF	Increased [101]	Enhanced recovery [107,108]	Facilatory [112,113]
KGF	Increased [102]	Undetermined	Undetermined
bFGF	Undetermined	Undetermined	Facilatory [112]
GDNF	Undetermined	Undetermined	Facilatory [114]
TGF-β	Increased† [98]	Undetermined	Inhibitory for branching [115]
PDGF	Increased† [98]	Undetermined	No effect [112]

*Increase in endogenous biologically active EGF probably from preformed sources; increase in EGF-receptor mRNA
†Chemoattractants for macrophages and monocytes (important source of growth promoting factors)

FIGURE 16-24

Growth factors in development and renal recovery. This table describes the roles of different growth factors in renal injury or in branching tubulogenesis. A large variety of growth factors have been tested for their ability either to mediate ureteric branching tubulogenesis or to affect recovery of kidney tubules after ischemic or other injury. Interestingly, growth factors that facilitate branching tubulogenesis in vitro also enhance the recovery of injured renal tubules.

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