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Review

Renal epithelial choices in ischemia: The unfolded protein response in acute kidney injury

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ABSTRACT

Acute kidney injury (AKI) occurs in 5-7% of hospitalized patients in the US. Treatment has largely remained limited to renal replacement measures and supportive care, and mortality remains disturbingly high. Over the last three decades, several pathways have been suggested to account for the derangements in renal function associated with AKI as well as the failure of some individuals to recover. The unfolded protein response (UPR) is a complex intracellular stress response triggered by misfolded proteins within the endoplasmic reticulum. The UPR is a key cellular mechanism that can mediate either cell death or recovery. Due to the large amount of directional transcellular transport, renal tubule cells of the proximal tubule and thick ascending limb of Henle have a high metabolic demand for regulated protein synthesis and trafficking. This makes the renal tubule cells particularly susceptible to ischemic or toxic damage. Factors that induce AKI may initiate the UPR within these cells and lead to a cascade of pro-apoptotic processes. The UPR also contributes to changes in cell polarity and in the location of tubular transport proteins that give rise to abnormal urinary excretory function. Far less is known about the role of the UPR in recovery from AKI but studies indicate that the UPR is involved in preconditioning responses and regeneration of tubular epithelium after injury.

KEYWORDS: unfolded protein response, acute kidney injury

ABBREVIATIONS

AKI	:	acute kidney injury
ARF	:	acute renal failure
ATF4	:	activating transcription factor 4
ATF6	:	activating transcription factor 6
CHOP	:	C/EBP homologous protein-10 also known as GADD153
eIF	:	eukaryotic initiation factor
ERK	:	extracellular signal-regulated kinase
ERSE	:	ER stress response element
GRP78	:	78 kDa glucose-related protein, also referred to as immunoglobulin heavy chain binding protein or BIP
HIF	:	hypoxia-inducible factor
IRE1	:	'inositol requiring 1'
JIK	:	c-Jun N-terminal inhibitory kinase inhibitory kinase
JNK	:	c-Jun N-terminal inhibitory kinase
MDCK	:	Madin-Darby canine kidney
ORP150	:	oxygen-regulated protein 150
PERK	:	protein kinase R-like endoplasmic-reticulum kinase

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TRAF2	:	tumor necrosis factor receptor-associated factor 2
UPR	:	unfolded protein response
XBP1	:	X-box binding factor protein 1

INTRODUCTION

Acute kidney injury (AKI) is a broad category of pathological processes characterized by rapid reduction in GFR glomerular filtration rate (GFR) over hours or days and diagnosed based on changes in serum creatinine [1, 2]. AKI is a major [3] and increasing [4] cause of morbidity and mortality in the US and throughout the world. In the US alone, the cost of treating AKI has risen to over ten billion dollars annually [5], and AKI now affects 5-7% of all patients admitted to US hospitals [3, 6]. The epidemiology, pathogenesis, and treatment of AKI are all areas of active research interest, but treatment is currently limited to renal replacement therapy and supportive measures. The mortality rate of AKI has remained a disturbingly consistent 50% for over three decades [7]. The failure of numerous potential therapeutic agents [8-10] in the face of intensive research suggests there may be a fundamental lack of understanding about cellular mechanisms which mediate AKI.

The unfolded protein response (UPR) is a complex intracellular stress response triggered by misfolded protein within the endoplasmic reticulum. The UPR is known to be active in cell culture [11] and animal models [12, 13] of AKI and UPR markers are present in biopsies from human patients with AKI [14]. The UPR may mediate either cell death or recovery in renal proximal tubule cells after ischemia and reperfusion injury. In this review, we investigate the impact of these conflicting roles for the UPR in AKI.

S3 Proximal tubule cells are selectively vulnerable to ischemic AKI

Definition and etiology of AKI

The definition of AKI by glomerular dysfunction, borne out of confusion about the associated terms ARF (acute renal failure) and ATN (acute tubular necrosis) [15], encompasses varied pre-renal, intra-renal and post-renal etiologies. Ischemia is the most common cause of AKI. Pre-renal or whole-body ischemia is the cause of reduced GFR in about 40% of AKI patients [6]. Pre-renal ischemia occurs on a continuum with ischemia within the kidney [16], and combined pre-renal and intra-renal ischemia cause 75% of all AKI [6]. Ischemic injury also complicates other etiologies such as surgery [17] and vasodilation associated with sepsis [16].

Hemodynamics of AKI

Largely because of differential oxygen supply and demand, the outer medulla of the kidney [18] and specifically the S3 segment of the proximal tubule [19] is selectively vulnerable to AKI. The renal vasculature draws 25% of cardiac output [16], but this blood flow is distributed unevenly throughout the kidney. Only 10% of total renal blood flow perfuses the renal medulla [20]. Low blood flow in the medulla is expected and necessary to drive countercurrent exchange through the loop of Henle. However, this architecture also results in low perfusion of the thick ascending limb and S3 proximal tubule epithelium, tissues with a high metabolic demand. Sodium reabsorption in these cells is the primary oxygen-consuming activity of the kidney [21]. Blood flow to the medulla is supplied by descending vasa recta arteries receiving the efferent flow of juxtamedullary glomeruli. This serial organization of arteries also severely depletes the oxygen content of blood perfusing the medulla, compounding the problem of reduced flow in this region. Partial pressure of oxygen drops from 50 mmHg in the cortex to only 10-20 mmHg in the outer medulla [22].

During AKI, overall renal blood flow can be rapidly reduced by up to 50% [23]. The regional differences in blood flow present in the uninjured kidney are further exacerbated during reperfusion following ischemia. In unilateral renal artery occlusion models of AKI, when blood flow to the renal artery is restored, the majority of the kidney is rapidly and effectively perfused. However, even when blood flow has returned to 100% of pre-ischemic levels in the rest of the kidney, outer medullary blood flow remains at just 10% of its pre-ischemic levels [24].

Histopathology of AKI at the S3 proximal tubule

Both thick ascending limb and S3 proximal tubule cells are disproportionally affected by AKI because of their location in the outer medulla and their high intrinsic metabolic demand. In animal models, specifically the warm ischemia and reperfusion rodent model of AKI, the S3 segment of the proximal tubule is more damaged than the distal tubule epithelium [25]. (Warm ischemia refers to clamping the renal artery of an intact kidney as opposed to cold ischemia which refers to preservation of the kidney in cold physiologic solution in preparation for transplantation.) The S3 is the deepest segment of the proximal tubule and the only proximal tubule segment significantly affected by the altered hemodynamics of the medulla. The proximal tubule reabsorbs over 60% of all the sodium and water filtered through the glomeruli as well as glucose, amino acids, phosphate, and other solutes [26]. Protein turnover is also high in the proximal tubule. Studies using radiolabeled amino acids show that the massive transport capacity of proximal tubule cells requires a massive turnover of 30-42% of proximal tubule cell protein per day [27, 28]. In addition to their unique blood supply, S3 proximal tubule cells may be more susceptible to acute injury because of their reduced glycolytic capacity relative to the distal nephron epithelium [29, 30]. We focus on the proximal tubule in this review because it is heavily studied in cell and animal models of ischemia, but we note that injury and recovery in distal tubule epithelium is also important in ischemic AKI, particularly in stress-induced genetic reprogramming and apoptosis.

Histologically, ischemic AKI manifests as acute tubular necrosis (ATN), a complex but well-characterized process that involves tubule dilation, effacement, loss of brush border, and patchy loss of whole cells [31, 32]. Four major avenues of damage have been proposed to mediate ATN: vasoconstriction and vasodilation imbalance, endothelium and leukocyte-derived vascular congestion, tubuloglomerular feedback, and tubular obstruction.

AKI activates the UPR in the S3 proximal tubule

Growing evidence indicates the UPR, triggered by misfolded protein in the endoplasmic reticulum, mediates cellular dysfunction in a wide variety of human pathologies [33]. The endoplasmic reticulum (ER) functions of protein folding and glycosylation are highly energy dependent and the loss of ATP in cardiac [34], epithelial cell [12], and brain [35, 36] ischemia causes protein misfolding and activates the UPR. Cells with a high protein turnover such as pancreatic beta cells [33] and plasma B cells [37] are known to be particularly susceptible to misfolded ER protein. Activation of the UPR has also been noted in the pathogenesis of many kidney disorders including chronic kidney disease, glomerulonephritis, membranous nephropathy, polycystic kidney disease, and diabetic nephropathy [38-40].

For the past two decades, investigation of the UPR has been a highlight of molecular biology research and is already well-described elsewhere [41-43]. A brief summary of the UPR's initiation and downstream effectors will facilitate our specific discussion of the UPR in AKI (Figure 1). In mammalian cells, UPR is a remarkably complex cellular mechanism but defined by a small group of initial mediators beginning with the 78 kDa glucose-related protein (GRP78). GRP78 is the ER-specific HSP70-family protein folding chaperone. In unstressed cells, GRP78 is active at low levels in the ER lumen where it corrects the misfolding of proteins translated across the rough ER membrane [44]. But the majority of GRP78 protein is bound to the luminal surface of three ER transmembrane proteins, PERK (protein kinase R-like endoplasmic-reticulum kinase), IRE1 ('inositol-requiring 1'), and ATF6 (activating transcription factor 6) in unstressed cells [45-47]. A wide variety of triggers including ER calcium depletion, loss of cellular ATP, inhibition of asparagine (N)-linked glycosylation, disulfide bond reduction, expression of mutant protein, and protein overexpression all culminate in overwhelming ER protein misfolding [48]. Mass action drives GRP78 off of its transmembrane partners to correct protein misfolding of ER luminal targets. The dissociation of GRP78 allows IRE1, PERK, and ATF6 to direct an expanding cascade of gene expression and cell signaling which will either permit the cell to recover from injury or induce apoptosis.

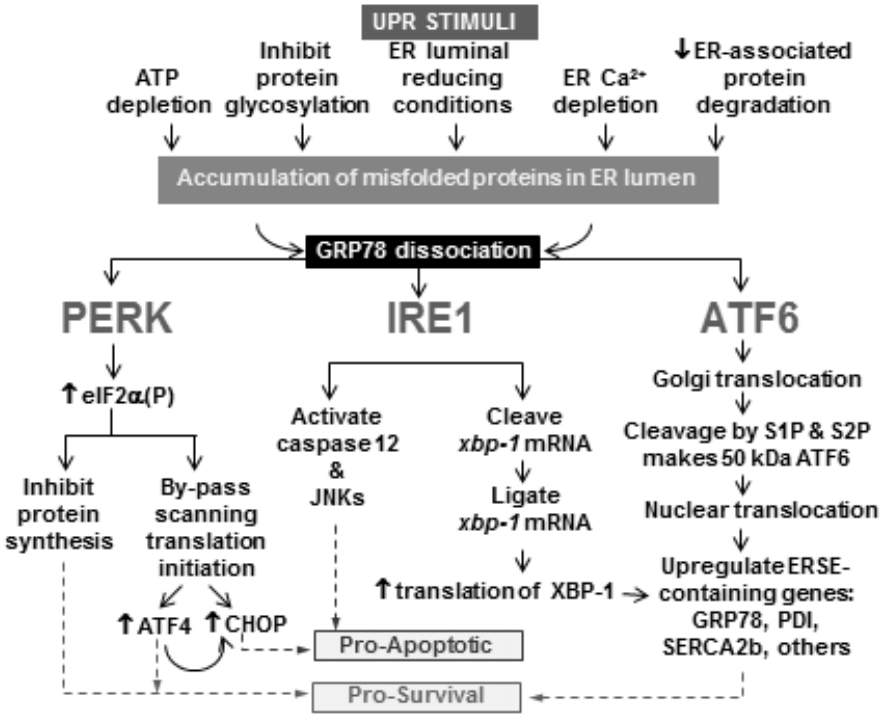

Figure 1. Pathways and elements involved in the unfolded protein response (see text).

In the absence of associated GRP78, both PERK [49] and IRE1 [50] dimerize and auto-phosphorylate, activating their regulatory activity. Activated IRE1 is a site-specific endoribonuclease targeting the X-box binding factor protein 1 mRNA (*XBP1*). IRE1 cleaves a 26-nucleotide sequence of *XBP1* generating a frameshift mutation. The XBP1 protein resulting from this mutation is a functional bZIP transcription factor, inducing genes regulated by an ER stress response element (ERSE) such as GRP78, GRP94, calreticulin, and many others [51]. IRE1 has also been directly implicated in activating the ER-specific caspase-12 to induce apoptosis. When activated by ER stress, the cytoplasmic domain of IRE1 interacts with c-Jun N-terminal inhibitory kinase (JNK) and tumor necrosis factor receptor-associated factor 2 (TRAF2) [52]. This interaction dissociates TRAF2 from caspase-12, allowing the dimerization and activation of caspase-12 [53].

Unlike IRE1, activated PERK functions primarily to inhibit global protein translation while preferentially upregulating the translation of specific transcripts. Protein translation typically begins with the formation of a complex of initiator tRNA (Met-$tRNA_R$) and eukaryotic initiation factor 2 (eIF2) bound to GTP called the ternary complex. Formation of this complex is rate-limiting in translation, and therefore a key regulatory point in protein expression. Initiation of translation leads to hydrolysis of eIF2-associated GTP to GDP. GTP must be restored before a new round of initiation can begin, a process catalyzed by the eIF2B subunit. PERK phosphorylation of the eIF2 alpha at serine-51 blocks guanine exchange by eIF2B and halts translation initiation [54]. eIF2 alpha is not phosphorylated exclusively by PERK, and inhibition of translation initiation through eIF2 alpha is a common mechanism to regulate many forms of cell stress, a process collectively referred to as the integrated stress response [42]. PERK is active and eIF2 alpha is phosphorylated in a rat whole-animal ischemia model of AKI, where eIF2 alpha phosphorylation is most prominent in outer medullary proximal tubule by immunohistochemistry [55].

Suppression of protein synthesis is generally viewed as protective, a mechanism to reduce

energy demands on an ischemic cell by preventing the expression of unnecessary housekeeping genes [56].

However, a small subset of mRNAs are known to escape the translational block, and their translation is upregulated by eIF2 alpha phosphorylation [57, 58]. These transcripts all have multiple upstream open reading frames which increase scanning time at the ribosome when eIF2 alpha is phosphorylated. Consistent with the UPR's overall paradoxical induction of both pro-apoptotic and pro-recovery mechanisms, proteins expressed during the integrated stress response include apoptosis-inducing C/EBP homologous protein-10 (CHOP) and pro-recovery activating transcription factor 4 (ATF4). Nephrin, an essential transmembrane protein in the slit diaphragm of podocytes, is known to escape translational block in this manner during the integrated stress response [59].

The third arm of the UPR is mediated by the 90-kDa ER transmembrane protein ATF6. Dissociation of GRP78 allows ATF6 to migrate to the Golgi apparatus [60] where proteases S1P and S2P cleave ATF6 into a 50-kDA active transcription factor [61] which interacts with the ERSE [62]. ATF6 and XBP1 have overlapping but distinct targets. XBP1 is itself induced by ATF6 [51], and ATF6 must be activated to express *XBP1* for splicing by IRE1 [63].

The UPR in proximal tubule cell models of ischemic injury

In culture, proximal tubule cells have long been used to study cellular injury and stress responses including the UPR [64]. Insults including oxygen and glucose deprivation, halogenated hydrocarbons, ultraviolet light, and depletion of thiols all cause robust induction of the UPR in proximal tubule cells [65]. A variety of nephrotoxic agents which induce AKI have been found to induce ER stress in proximal tubule cell culture including acetaminophen [66], cisplatin [67, 68], gentamicin [69], cyclosporine A [70], and heavy metals [71]. The UPR is also specifically induced in cell culture with a variety of pharmacological and chemical agents including the SERCA inhibitor [72] thapsigargin [73], tunicamycin [74] which is an inhibitor of protein glycosylation [75], calcium ionophores such as A23187 [76], and the secretory pathway inhibitor [77] brefeldin A [78]. All of these agents specifically induce ER stress but have relatively little effect on related pathways such as the heat shock response.

Inhibition of *Grp78* by RNAi in the porcine proximal tubule LLC PK1 cell line resulted in increased cell death from the alkylating agent iodoacetamide [65]. In immortalized rat proximal tubule cells, tunicamycin and brefeldin A induced autophagy, a form of cell death characterized by lysosomal degradation of membrane-bound structures [79].

The UPR in animal models of AKI

The Aoe laboratory has created a mouse with nonfunctional mutant knock-in *Grp78* [80]. Mice bred with homozygous mutant *Grp78* die as neonates from respiratory failure. Heterozygotes for the nonfunctional mutant *Grp78* survived to adulthood and displayed normal kidney function by physiological parameters. However, histological examination of older mice showed marked tubulo-interstitial lesions compared with age-matched controls, suggesting that impairment of the UPR may predispose the kidney to tubulo-interstitial disease such as the acute tubular necrosis seen in AKI [81]. The mutant knock-in mice were also more susceptible to an albumin-overload model of proteinuria and displayed more caspase-12 activity in this model. The specific and localized pathology of acute tubular necrosis in this *Grp78* null model imply that renal tubular epithelium is especially prone to ER dysfunction.

Forty-five minutes of left renal artery occlusion in rats resulted in increased expression of transcripts for GRP78, GRP94, and another ER luminal protein, ERp72 [12], establishing that the UPR was active in animal models of ischemic AKI. Recently, Prachasilchai and colleagues described a precise time course of UPR induction in a mouse 35-minute bilateral renal artery ischemia model which replicated the histological tubular epithelium changes, casts, endothelial cell death, and rise in plasma creatinine seen in human AKI patients [13]. They showed a logical, progressive expression of UPR markers during reperfusion.

XBP1 mRNA was detectable at 2 hours reperfusion, and GRP78 mRNA and protein expressed at 6 hours reperfusion. Immuno-histochemistry showed that GRP78 expression localized to the proximal tubule cells at the corticomedullary junction.

Cisplatin is a nephrotoxic agent used in chemotherapy for many malignancies and is known to induce AKI [67, 68]. Animal models of AKI often employ cisplatin instead of ischemia because cisplatin-induced injury is predictably dose-dependent and results in more consistent outcomes [82]. Induction of the UPR also occurs in cisplatin models of AKI, and this induction is again localized to the proximal tubule. Five mg per kg cisplatin in Wistar rats significantly increased plasma creatinine and BUN. Increases in cleaved *XBP1* mRNA by qPCR and XBP1 protein by western blot were seen in the proximal tubule 7 days post-administration [69]. Additionally, these rats showed changes consistent with m-calpain activation indicating cytoplasmic calcium-induced proteolysis.

A study of UPR gene induction in mice stressed with hypobaric hypoxia presents an interesting challenge to the general consensus that lack of oxygen upregulates the UPR in kidney cells. After up to 12 hours of hypobaric hypoxia, microarray and PCR showed that *Grp78* and *Grp94* expression and *XBP1* splicing were all significantly downregulated in whole-kidney isolated mRNA from mouse [83]. In contrast, cardiac tissue from the same animals showed expected increases in *Grp78*, *Grp94*, and *XBP1* splicing. In a related finding, podocyte cell culture ischemia, but not hypoxia or hyperglycemia induce the UPR [84]. These findings underscore the differences between hypoxic and ischemic injuries as well as differential regulation of the UPR in different tissues.

UPR interactions with other cellular pathways in AKI

There is a confluence between the downstream pathways active after protein misfolding and those found in models of AKI. Free radical damage, increased intracellular calcium, proteolysis and lipolysis, loss of cell polarity, and cell death are all common between UPR and AKI models.

Polarity and localization

Polymerized actin is necessary to maintain the proximal tubule epithelium apical brush border. AKI causes actin polymer disassembly and re-localization of actin to the cytoplasm, a process mediated by cofilin/ADF [85]. The cofilin/ADF family of proteins, which disassemble actin to monomers, are normally sequestered away from the apical surface of proximal tubule epithelium. During AKI, ADF/cofilin re-localizes to the apical surface and disassembles actin polymers, disrupting the brush border. Depolarization in AKI also re-localizes basolateral Na/K-ATPase to the apical surface. Reduced sodium reabsorption caused by abnormal targeting of Na/K-ATPase to the apical membrane may alter tubuloglomerular feedback at the macula densa increasing sodium excretion. The resultant increase in tubular sodium enhances the polymerization of Tamm-Horsfall proteins that are synthesized and secreted by the thick ascending limb of Henle, thereby contributing to tubular obstruction and backleak due to the formation of dense tubular casts. Chemical chaperones have shown a surprising ability to restore subcellular localization by correcting misfolded protein as discussed below.

Calcium

Impaired calcium sequestration is an important mediator of cellular injury in both AKI and in the UPR. Calcium depletion in the ER can induce the UPR and the UPR can induce expression of proteins involved in calcium sequestration such as SERCA. In AKI, ATP depletion results in impaired intracellular calcium sequestration. Increased intracellular calcium activates the calpain family of calcium-dependent cysteine proteases which can cleave procaspase-12 to active caspase-12 [86]. Calcium can also directly activate phospholipases damaging cell membranes. In rabbit renal proximal tubule cells, 90% ATP depletion by the mitochondrial inhibitor antimycin increased cytosolic calcium from 20 to 100 nM [87]. Importantly, pretreatment of these cells with a low dose of thapsigargin prevented antimycin-induced cell death, presumably by preventing the depletion of ER calcium.

Cell death

Cell death by apoptosis and necrosis are both present after AKI. Cell death from AKI is typically differentiated by nephron segment with apoptotic cells more prominent in the distal tubule and necrotic cells more in the proximal tubule [88]. But it is now accepted that cell death in both proximal and distal nephron exist on a broad continuum where apoptosis and necrosis represent only the most extreme cases [32]. Pro-apoptotic factors known to be active in AKI include Bcl-2, cytochrome c, caspase-9, p53, NF-kB (activated by TNF receptor stimulation), Fas, FADD, p38, and pJNKs [32].

In renal ischemia, cells throughout the nephron phosphorylate JNKs, activating the extrinsic pathway of apoptosis. Thick ascending limb and distal tubule cells also produce extracellular-signal-regulated kinases (ERKs) in response to ischemia [89]. ERKs can inhibit activation of the pro-apoptotic JNK kinases in renal ischemia, and it is thought that the balance of ERK to JNK production confers protection from apoptosis in response to ischemia. Proximal tubule cells produce only pJNKs and not ERKs, presumably making proximal tubule cells more susceptible to apoptosis. However, we have already noted that necrosis is the predominant form of cell death observed morphologically in the proximal tubule in animal models of I/R and in human biopsies [90]. Counter intuitively it is the distal tubule where ERKs are produced that experiences relatively more cell death by apoptosis. Thus, factors controlling the balance of apoptotic and necrotic pathways in the nephron during I/R remain wholly unquantified.

The UPR can induce apoptosis through a variety of mechanisms. All three arms of the UPR can ultimately lead to increased expression of the pro-apoptotic factor C/EBP homologous protein-10 (CHOP). CHOP can induce insertion of pro-apoptotic Bcl-2 related proteins into the ER membrane resulting in calcium release and into the outer mitochondrial membrane allowing the release of cytochrome c. As mentioned above, IRE1 association with JIK and TRAF2 can activate caspase-12. The UPR can also induce anti-apoptotic transcripts. Bax inhibitor 1 is an ER-localized apoptosis inhibitor induced by the UPR. Mice with knockout of the Bax inhibitor 1 gene, *Bi-1*, had increased susceptibility to ER stress in a renal artery occlusion model of AKI [91].

Before and after- the UPR in preconditioning and regeneration

Preconditioning

In multiple organs and injury modalities, exposure to a brief ischemic episode confers protection to a subsequent, larger ischemic insult, a process known as preconditioning. This pretreatment is often accomplished with a specific UPR-inducing agent. For example, pretreatment with tunicamycin or A23187 protected 80% of Madin-Darby canine kidney (MDCK) cells from cell death from antimycin-induced ATP depletion [92]. Preconditioning of tubule cells may confer protection by preserving cytoarchitecture and junctional complexes. Again using MDCK cells pretreated with tunicamycin and ATP-depleted with antimycin, George, *et al.* found the UPR prevented re-localization of Na/K-ATPase and the apical marker gp135 and increased cell adhesion 5-fold over cells that were not preconditioned [92].

LLC-PK1 cells treated with nonlethal doses of thapsigargin, tunicamycin, or A23187 all expressed increased GRP78, and GRP78 expression was protective against oxidative injury, an effect abolished by antisense RNA against *Grp78* [93]. Preconditioning also prevented the injury-induced increase in cytoplasmic calcium and shifted the kinase pathways to favor ERK activation over JNK, the same mechanisms proposed to confirm protection of thick ascending limb cells to apoptosis. Preconditioning has also been effective in preventing damage in an animal model of AKI. Mice pretreated with tunicamycin two days before bilateral renal artery occlusion had reduced cast formation, tubule dilation, and epithelial cell death in both the cortex and outer medulla [13].

An emerging strategy to impart protection from the UPR is the use of nontoxic, specific UPR inducers. Pretreatment with one such nontoxic inducer, DTTox, protected rat kidneys from nephrotoxic AKI as measured by BUN [94].

The same effect was seen in LLC-PK1 cells which had significantly reduced lactate dehydrogenase release.

Pro-recovery gene expression

Genes induced by the UPR vary by tissue and stress, an area of active investigation [95]. The UPR induces a wide variety of genes regulated by the poorly defined ERSE sequence. These genes can be either pro-apoptosis or pro-recovery. A number of UPR-regulated pro-recovery genes have been implicated in recovery from AKI. Oxygen regulated protein 150 (ORP150) contains an ERSE [96] and is upregulated in rat 1-hour renal artery occlusion models and human AKI biopsies [14]. ORP150 is an ER-specific chaperone, but its precise molecular function is unknown. Constitutive expression of ORP150 protected MDCK cells from hypoxic stress, and cells with siRNA knockdown of ORP150 showed increased caspase-3 production and cell death during hypoxia [14]. Consistent with the selective vulnerability of S3 proximal tubule, thick ascending limb cells expressed more ORP150 than proximal tubule cells.

Regeneration of tubular epithelium

While cytoplasmic calcium can damage renal epithelium by activating proteases and phospholipases, unsequestered calcium also induces the expression of proliferative proteins. In a mouse nephrotoxic model of AKI, tubule cell regeneration correlated to increased expression of calcium-binding proteins S100A6 and annexin A2, and expression of these proteins correlated to expression of proliferating cell nuclear antigen, a cell proliferation marker [97].

Chemical chaperones

The primary function of GRP78 and related HSP70-family proteins is to chaperone other proteins into a proper folding conformation. It has been proposed that small molecules could be added to cells to perform as chemical chaperones and assist normal protein folding [98, 99]. Small molecules functioning to correct protein misfolding have been studied in the treatment of several kidney pathologies. In congenital nephrotic syndrome, missense mutations result in ER retention of misfolded nephrin in podocytes and stimulate the UPR [100]. The chemical chaperone sodium 4-phenylbutyrate restored localization and functionality in some mutants [101]. Podocytes with a mutation in the NPHS2 gene express mutant podocin, causing autosomal recessive type steroid-resistant nephrotic syndrome. When glycerol, trimethylamine-N-oxide, or dimethylsulfoxide was added to podocyte culture with mutated *NPHS2*, these chemicals corrected podocin misfolding allowing podocin to migrate through the secretory pathway to the cell membrane [102]. For now, chemical chaperones have only found use in podocytes and not in tubular epithelium, but these findings suggest that such studies would be warranted.

SUMMARY

Ischemia and toxic injury to the kidney lead to apoptosis and, if severe, necrosis of renal tubular epithelium. The S3 segment of the proximal tubule and the thick ascending limb in the outer medulla are particularly susceptible to these insults. The resultant ATP depletion, ER calcium depletion, changes in redox potential and other factors within these cells may initiate the UPR leading to a cascade of pro-apoptotic processes. The UPR also contributes to changes in cell polarity and the location of tubular transport proteins both of which give rise to abnormal urinary excretory function. Far less is known about the role of the UPR in recovery from AKI but studies indicate that the UPR is involved in preconditioning responses and regeneration of tubular epithelium after injury. Given the large number of individuals at risk for AKI and the dismal prognosis of this disorder, the role of the UPR in the initiation and recovery from acute kidney injury merits continued investigation.

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