Pathophysiology of Ischemic Acute Renal Failure: Cytoskeletal Aspects

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Ischemia remains the major cause of acute renal failure (ARF) in the adult population [1]. Clinically a reduction in glomerular filtration rate (GFR) secondary to reduced renal blood flow can reflect prerenal azotemia or acute tubular necrosis (ATN). More appropriate terms for ATN are *acute tubular dysfunction* or *acute tubular injury*, as necrosis only rarely is seen in renal biopsies, and renal tubular cell injury is the hallmark of this process. Furthermore, the reduction in GFR during acute tubular dysfunction can now, in large part, be related to tubular cell injury. Ischemic ARF resulting in acute tubular dysfunction secondary to cell injury is divided into initiation, maintenance, and recovery phases. Recent studies now allow a direct connection to be drawn between these clinical phases and the cellular phases of ischemic ARF (Fig. 13-1). Thus, renal function can be directly related to the cycle of cell injury and recovery.

Renal proximal tubule cells are the cells most injured during renal ischemia (Fig. 13-2) [2,3]. Proximal tubule cells normally reabsorb 70% to 80% of filtered sodium ions and water and also serve to selectively reabsorb other ions and macromolecules. This vectorial transport across the cell from lumen to blood is accomplished by having a surface membrane polarized into apical (brush border membrane) and basolateral membrane domains separated by junctional complexes (Fig. 13-3) [4]. Apical and basolateral membrane domains are biochemically and functionally different with respect to many parameters, including enzymes, ion channels, hormone receptors, electrical resistance, membrane transporters, membrane lipids, membrane fluidity, and cytoskeletal associations. This epithelial cell polarity is essential for normal cell function, as demonstrated by the vectorial transport of sodium from the lumen to the blood (*see* Fig. 13-3). The establishment

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and maintenance of this specialized organization is a dynamic and ATP dependent multistage process involving the formation and maintenance of cell-cell and cell-substratum attachments and the targeted delivery of plasma membrane components to the appropriate domains [5]. These processes are very dependent on the cytoskeleton, in general, and the cytoskeletal membrane interactions mediated through F-actin (*see* Fig. 13-2, 13-3), in particular.

Ischemia in vivo and cellular ATP depletion in cell culture models ("chemical ischemia") are known to produce characteristic surface membrane structural, biochemical, and functional abnormalities in proximal tubule cells. These alterations occur in a duration-dependent fashion and are illustrated in Figures 13-2 and 13-3 and listed in Figure 13-4. Ischemia-induced alterations in the actin cytoskeleton have been postulated to mediate many of the aforementioned surface membrane changes [2,6,7]. This possible link between ischemia-induced actin cytoskeletal alterations and surface membrane structural and functional abnormalities is suggested by several lines. First, the actin cytoskeleton is known to play fundamental roles in surface membrane formation and stability, junctional complex formation and regulation, Golgi structure and function, and cell–extracellular membrane attachment [2,4,5,8]. Second, proximal tubule cell actin cytoskeleton is extremely sensitive to ischemia and ATP depletion [9,10]. Third, there is a strong correlation between the time course of actin and surface membrane alterations during ischemia or ATP depletion [2,9,10]. Finally, many of the characteristic surface membrane changes induced by ischemia can be mimicked by F-actin disassembly mediated by cytochalasin D [11]. Although these correlations are highly suggestive of a central role for actin alterations in the pathophysiology of ischemia-induced surface membrane damage they fall short in providing mechanistic data that directly relate actin cytoskeletal changes to cell injury.

Proximal tubule cell injury during ischemia is also known to be principally responsible for the reduction in GFR. Figure 13-5 illustrates the three known pathophysiologic mechanisms that relate proximal tubule cell injury to a reduction in GFR. Particularly important is the role of the cytoskeleton in mediating these three mechanisms of reduced GFR. First, loss of apical membrane into the lumen and detachment of PTC result in substrate for cast formation. Both events have been related to actin cytoskeletal and integrin polarity alterations [12–15]. Cell detachment and the loss of integrin polarity are felt to play a central role in tubular obstruction (Fig. 13-6). Actin cytoskeletal-mediated tight junction opening during ischemia occurs and results in back-leak of glomerular filtrate into the blood. This results in ineffective glomerular filtration (Fig. 13-7). Finally, abnormal proximal sodium ion reabsorption results in large distal tubule sodium delivery and a reduction in GFR via tubuloglomerular feedback mechanisms [2,16,17].

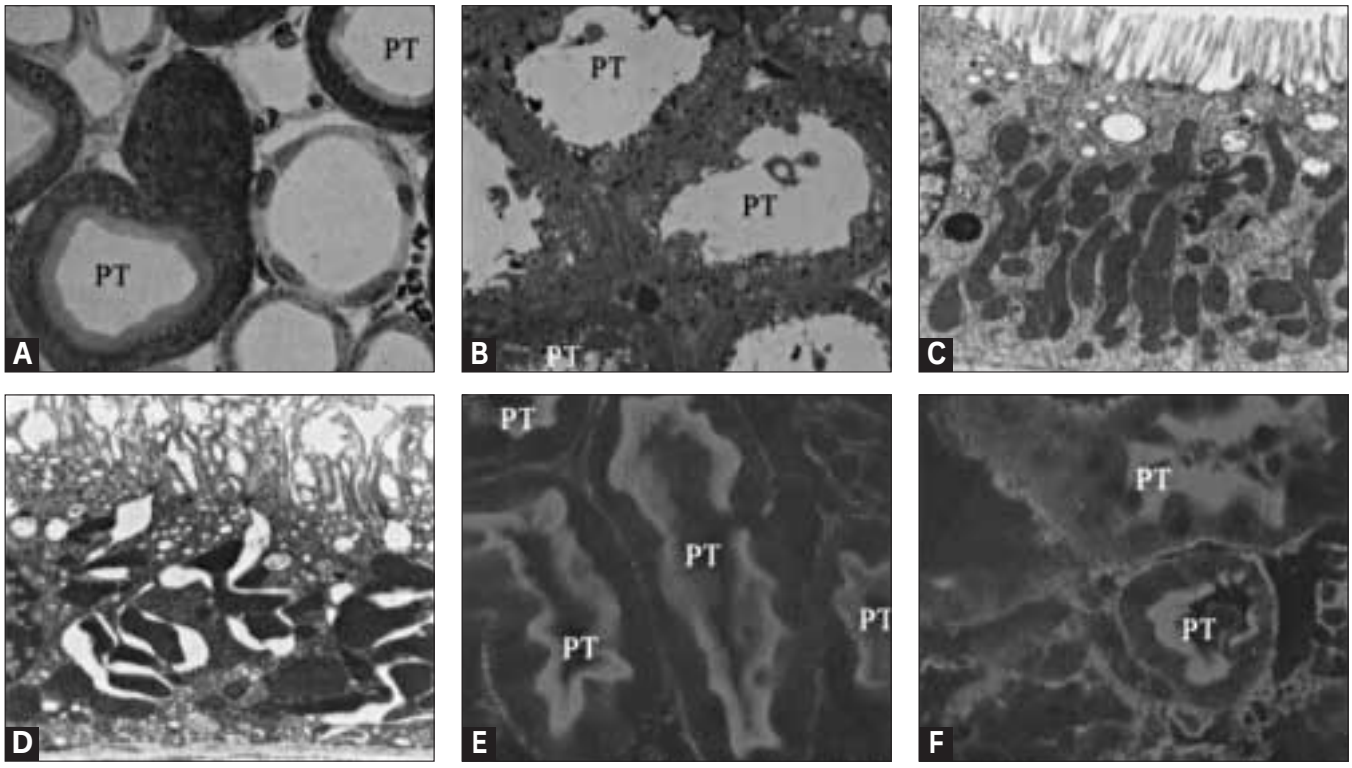
In summary, ischemia-induced alterations in proximal tubule cell surface membrane structure and function are in large part responsible for cell and organ dysfunction. Actin cytoskeletal dysregulation during ischemia has been shown to be responsible for much of the surface membrane structural damage.

RELATIONSHIP BETWEEN THE CLINICAL AND CELLULAR PHASES OF ISCHEMIC ACUTE RENAL FAILURE

Clinical Phases	Cellular Phases
Prerenal azotemia	Vascular and cellular adaptation
↓	↓
Initiation	ATP depletion, cell injury
↓	↓
Maintenance	Repair, migration, apoptosis, proliferation
↓	↓
Recovery	Cellular differentiation

FIGURE 13-1

Relationship between the clinical and cellular phases of ischemic acute renal failure. Prerenal azotemia results from reduced renal blood flow and is associated with reduced organ function (decreased glomerular filtration rate), but cellular integrity is maintained through vascular and cellular adaptive responses. The initiation phase occurs when renal blood flow decreases to a level that results in severe cellular ATP depletion that, in turn, leads to acute cell injury. Severe cellular ATP depletion causes a constellation of cellular alterations culminating in proximal tubule cell injury, cell death, and organ dysfunction [2]. During the clinical phase known as maintenance, cells undergo repair, migration, apoptosis, and proliferation in an attempt to re-establish and maintain cell and tubule integrity [3]. This cellular repair and reorganization phase results in slowly improving cell and organ function. During the recovery phase, cell differentiation continues, cells mature, and normal cell and organ function return [18].



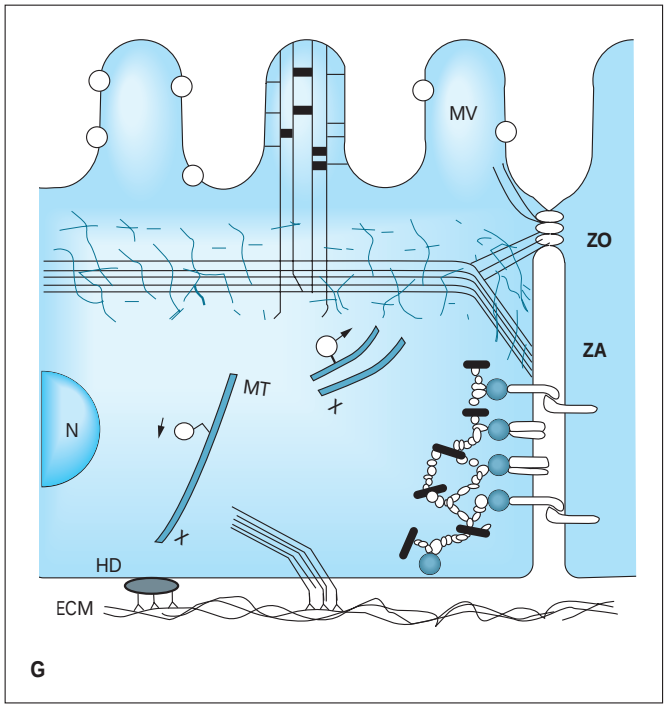


FIGURE 13-2

Ischemic acute renal failure in the rat kidney. Light **A, B**, transmission electron, **C, D**, and immunofluorescence **E, F**, microscopy of control renal cortical sections, **A, C, E**, and after moderate ischemia induced by 25 minutes of renal artery occlusion, **B, D, F**. Note the extensive loss of apical membrane structure, **B, D**, in proximal (PT) but not distal tubule cells. This has been shown to correlate with extensive alterations in F-actin as shown by FITC-phalloidin labeling, **E, F**. **G**, Drawing of a proximal tubule cell under physiologic conditions. Note the orderly arrangement of the actin cytoskeleton and its extensive interaction with the surface membrane at the zonula occludens (ZO, tight junction) zonula adherens (ZA, occludens junction), interactions with ankyrin to mediate Na^+, K^+-ATPase [2] stabilization and cell adhesion molecule attachment [5,8]. The actin cytoskeleton also mediates attachment to the extracellular matrix (ECM) via integrins [12,15]. Microtubules (MT) are involved in the polarized delivery of endocytic and exocytic vesicles to the surface membrane. Finally, F-actin filaments bundle together via actin-bundling proteins [19] to mediate amplification of the apical surface membrane via microvilli (MV). The actin bundle attaches to the surface membrane by the actin-binding proteins myosin I and ezrin [19,20].