Minireview

Store-operated calcium entry: Pivotal roles in renal physiology and pathophysiology

Sarika Chaudhari, Robert T Mallet, Parisa Y Shotorbani, Yu Tao and Rong Ma

Department of Physiology and Anatomy, University of North Texas Health Science Center, Fort Worth, TX 76107, USA
Corresponding author: Sarika Chaudhari. Email: Sarika.Chaudhari@unthsc.edu

Impact statement

In the last two decades, SOCE has emerged as a major Ca^{2+} signaling mechanism in myriad cells, yet its complex regulation and the membrane channels involved are not fully understood. SOCE plays diverse roles in different renal cell types, even in the same organ system. SOCE disturbances are pivotal to the pathogenesis of many renal disorders, including renal fibrosis in chronic kidney disease, polycystic kidney disease, water imbalance, and acute-to-chronic kidney disease transition. Our knowledge about SOCE-associated renal disorders is growing exponentially. This review integrates recent and current research in the physiology and pathophysiology of renal SOCE. Discussed in detail are several putative SOCE-regulated pathways that modulate synthesis of extracellular matrix, with direct relevance to fibrotic kidney diseases.

Abstract

Research conducted over the last two decades has dramatically advanced the understanding of store-operated calcium channels (SOCC) and their impact on renal function. Kidneys contain many types of cells, including those specialized for glomerular filtration (fenestrated capillary endothelium, podocytes), water and solute transport (tubular epithelium), and regulation of glomerular filtration and renal blood flow (vascular smooth muscle cells, mesangial cells). The highly integrated function of these myriad cells effects renal control of blood pressure, extracellular fluid volume and osmolality, electrolyte balance, and acid–base homeostasis. Many of these cells are regulated by Ca^{2+} signaling. Recent evidence demonstrates that SOCCs are major Ca^{2+} entry portals in several renal cell types. SOCC is activated by depletion of Ca^{2+} stores in the sarco/endoplasmic reticulum, which communicates with plasma membrane SOCC via the Ca^{2+} sensor Stromal Interaction Molecule 1 (STIM1). Orai1 is recognized as the main pore-forming subunit of SOCC in the plasma membrane. Orai proteins alone can form highly Ca^{2+} selective SOCC channels. Also, members of the Transient Receptor Potential Canonical (TRPC) channel family are proposed to form heteromeric complexes with Orai1 subunits, forming SOCC with low Ca^{2+} selectivity. Recently, Ca^{2+} entry through SOCC, known as store-operated Ca^{2+} entry (SOCE), was identified in glomerular mesangial cells, tubular epithelium, and renovascular smooth muscle cells. The physiological and pathological relevance and the characterization of SOCC complexes in those cells are still unclear. In this review, we summarize the current knowledge of SOCC and their roles in renal glomerular, tubular and vascular cells, including studies from our laboratory, emphasizing SOCE regulation of fibrotic protein deposition. Understanding the diverse roles of SOCE in different renal cell types is essential, as SOCC and its signaling pathways are emerging targets for treatment of SOCE-related diseases.

Keywords: Store-operated Ca^{2+} channels, STIM1, Orai1, TRPC, extracellular matrix, kidney disease, mesangial cells

***Experimental Biology and Medicine* 2021; 246: 305–316. DOI: 10.1177/1535370220975207**

Introduction

The complex physiological functions of mammalian kidneys require functional integration of diverse, highly specialized cells including vascular endothelium and smooth muscle, glomerular mesangial cells (MCs), podocytes, and tubular epithelial cells. The discovery of store-operated Ca^{2+} channels (SOCC) in these diverse renal cells has ignited intense research effort to delineate the contributions of store-operated Ca^{2+} entry (SOCE) to renal function and the complex neuroendocrine regulation of renal blood flow, glomerular filtration, and tubular handling of water and electrolytes. In particular, SOCE has emerged as a crucial regulator of extracellular matrix (ECM) synthesis and deposition by glomerular MCs, and a convergence point of neuroendocrine and paracrine signaling mechanisms. Moreover, SOCE dysregulation is implicated in the pathogenesis of mesangial and interstitial fibrosis in diabetic nephropathy, polycystic kidney disease, glomerular hemodynamic disorders, acute-to-chronic kidney disease transition, and other chronic conditions afflicting the renal system. This article summarizes the recent and ongoing research that has characterized SOCE

and delineated its pivotal contributions to renal function and disease.

In 1986, Putney proposed the novel phenomenon of capacitative Ca^{2+} entry, wherein the reduction of intracellular Ca^{2+} stores activates Ca^{2+} influx through plasma membrane Ca^{2+} channels.[1] Three decades later, the molecular mediators and mechanisms regulating this phenomenon are the focus of intense research effort. Activation of phospholipase C coupled receptors provokes hydrolysis of phosphatidylinositol 4,5-bisphosphate, generating inositol 1,4,5-trisphosphate (IP_3) and diacylglycerol. Acting on its receptors in the sarco/endoplasmic reticulum (SR/ER), IP_3 releases Ca^{2+} from these intracellular stores. Depletion of these Ca^{2+} stores triggers opening of plasma membrane Ca^{2+} channels allowing Ca^{2+} influx from the extracellular compartment. Because of its dependence on Ca^{2+} store depletion, this Ca^{2+} influx was termed SOCE, and the plasma membrane channels mediating this Ca^{2+} entry were dubbed SOCC. Subsequent studies demonstrated SOCE to be a pivotal Ca^{2+} entry mechanism for several non-excitable and excitable cell types.

Proposed SOCE mechanisms include diffusible messengers,[2] vesicle fusion/exocytosis,[3] and direct coupling between ER-IP_3 receptor channels and plasma membrane Ca^{2+} channels[4] including those of the Transient Receptor Potential Canonical (TRPC) channel family. However, only within the last 15 years have gene array and RNA interference technology enabled identification of stromal interaction molecule 1 (STIM1) as the ER Ca^{2+} sensor that signals plasma membrane Orai1 Ca^{2+} channels to execute SOCE.[5–7] Various TRPC channels are also known to be activated by ER-STIM1, whereupon they form complexes with Orai subunits to regulate SOCE.[8] Although the subunit composition of SOCC has been debated, it is now widely accepted that homo- or hetero-multimeric Orai subunits form highly Ca^{2+}-selective SOCC, while Orai subunits which interact with TRPC channels form the nonselective SOCC.[9,10] TRPC channels are characterized by their relatively low selectivity for Ca^{2+}, while the Ca^{2+} release-activated Ca^{2+} channel, the classical SOCC, has a high Ca^{2+} selectivity.[11] TRPC channels can also independently act as receptor-operated calcium channels (ROCC) which are activated by various agonists stimulating diverse G-protein-coupled receptors, and subsequent increase in diacylglycerol and decrease in phosphatidylinositol 4,5-bisphosphate. ROCC activation is independent of SR/ER Ca^{2+} store depletion.

The renal architecture organizes multiple cell types, each serving unique functions, into functional units that effect electrolyte homeostasis, extracellular fluid volume and blood pressure regulation, and excretion of waste materials. SOCE plays an essential role in a wide variety of physiological functions including exocytosis, enzymatic activity, gene transcription, cell proliferation, and apoptosis.[12] In this review, we focus on recent studies demonstrating SOCE in different renal cell types, identifying the contributing channels, defining SOCE's physiological relevance and pathways involved, and assessing the pathological potential of altered SOCE.

SOCE in the renal corpuscle

SOCE in glomerular MCs

Renal corpuscles harbor several cell types including MCs, podocytes, fenestrated capillary endothelium, interstitial fibroblasts, and squamous parietal epithelium. MCs, podocytes, and capillary endothelial cells act in an integrated manner to effect robust regulation of glomerular filtration.[13–16] MCs are readily isolated and maintained in cell culture,[17] permitting study of their many crucial functions. MCs provide structural support for the glomerular capillary loops, generate the mesangial extracellular matrix, control matrix turnover by regulating matrix metalloproteinase-catalyzed degradation, and clear debris by phagocytosis. In a reciprocal fashion, the matrix components influence MC growth and proliferation as well as matrix-cell signaling to optimize glomerular function.[18] ECM secreted by MCs can bind and store growth factors. Moreover, MCs secrete numerous growth factors, vasoactive agents, cytokines, and matrix metalloproteinases. MC hypertrophy and proliferation provoke excessive mesangial matrix accumulation in chronic kidney diseases with fibrosis including diabetic nephropathy.[19] The matrix components are tightly controlled in the normal physiological environment, while many acute and chronic renal diseases are associated with altered protein abundance or abnormal release from the matrix scaffold.[19,20] MCs exert another important function: their contractile properties enable fine tuning of glomerular capillary flow and the surface area available for glomerular filtration. However, the technical challenges of accessing these cells limit the study of their contractile function in vivo. MCs also serve as sources and targets of growth factors and various cytokines.

Because MC activity is essential for glomerular health, and Ca^{2+} signaling modulates the function of these cells, the role of SOCE in MC has attracted considerable research attention. In 1994, Mene *et al.* reported the first evidence of SOCE in human MCs. Using Fura2 fluorescence to monitor $[Ca^{2+}]_i$, they demonstrated that Ca^{2+} store depletion with angiotensin II, thapsigargin (TG), or ionomycin increased Ca^{2+} influx upon addition of Ca^{2+} to the initially Ca^{2+} free medium, in a manner independent of plasma membrane depolarization.[21] Subsequent studies reinforced and extended these findings.[22,23] Using patch clamp technique, Ma *et al.* demonstrated that the single channel currents in human MCs had the same properties as SOCC currents.[11] When pre-incubated with Ca^{2+} chelator BAPTA and treated with TG, these cells showed a non-voltage activated, inwardly rectifying current that was remarkably selective for Ca^{2+} and could be blocked by low concentrations of La^{3+}, a SOCC blocker.[22]

The direct evidence of SOCC in MCs raised the question, if and how SOCE affects physiological signaling in these cells. Initial studies revealed that vasoactive factors like angiotensin II and thromboxane-A_2 activate SOCE through G protein-coupled receptors via the classical phospholipase C/diacylglycerol/IP_3 pathway in many cells, including MCs (Figure 1(a)).[21] Additionally, receptor tyrosine kinases, the high-affinity cell surface receptors for many polypeptide growth factors, cytokines, and hormones, also stimulated

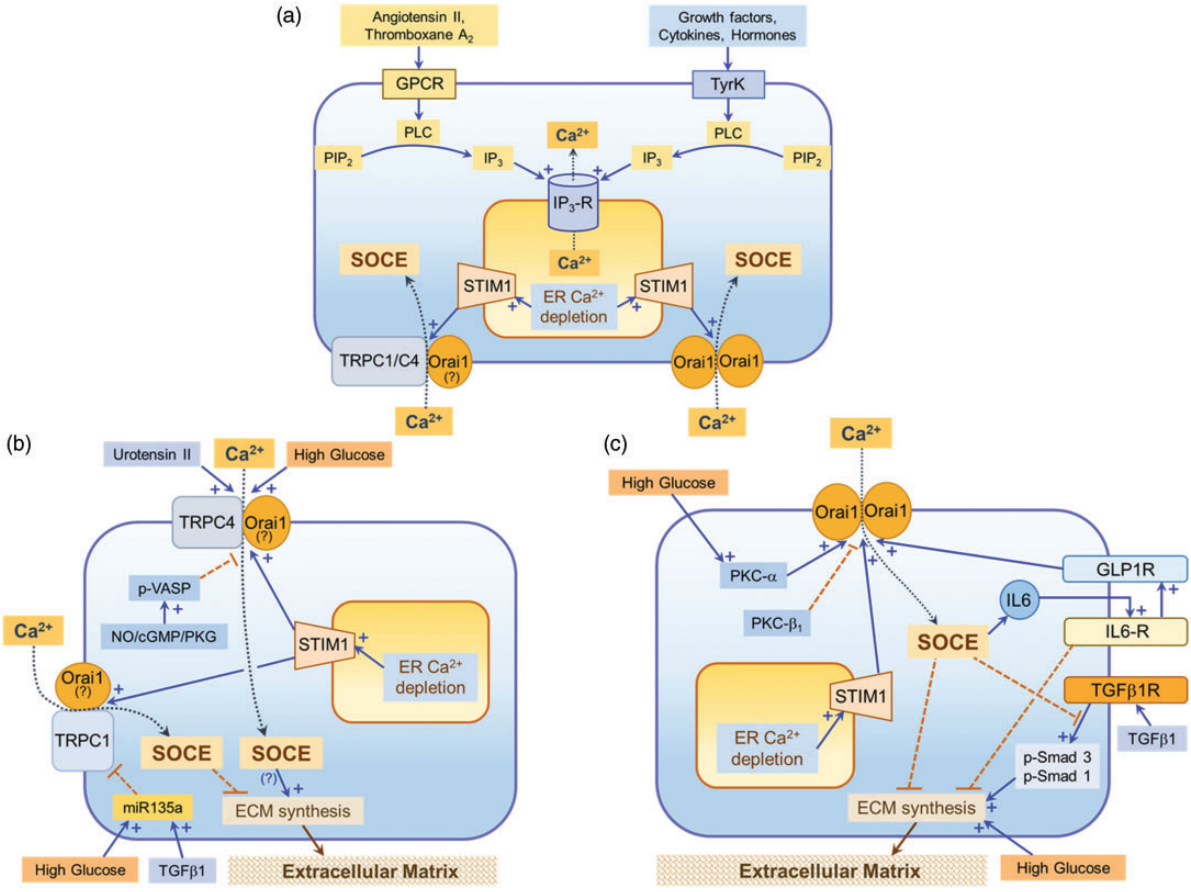

Figure 1. Mediators and modulators of store-operated Ca^{2+} entry in glomerular mesangial cells. **Panel (a)**: Endoplasmic reticular (ER) Ca^{2+} depletion activates store-operated Ca^{2+} entry (SOCE) via STIM1. Activation of G-protein coupled receptors (GPCR) or receptor tyrosine kinases activates phospholipase C (PLC), which hydrolyzes phosphatidylinositol bisphosphate (PIP_2), generating inositol trisphosphate (IP_3). Interaction of IP_3 with its ER receptor provokes Ca^{2+} release, depleting the ER Ca^{2+} store. ER Ca^{2+} depletion causes conformational changes in STIM1 that activate Orai1:Orai1 homodimeric and/or Orai1: transient receptor potential canonical (TRPC) heterodimeric store-operated Ca^{2+} channels in the plasma membrane and, thus, SOCE. **Panel (b)**: Factors modulating SOCE via TRPC channels. STIM1-activated TRPC1- and TRPC4-mediated Ca^{2+} entry are suppressed, respectively, by micro RNA 135a (miR135a) and the nitric oxide (NO)—cyclic GMP (cGMP)—protein kinase G (PKG)—vasodilator-stimulated phosphoprotein (VASP) cascade. TRPC1-mediated SOCE inhibits extracellular matrix (ECM) synthesis, while available evidence suggests urotensin II-induced TRPC4-mediated SOCE activates ECM synthesis. TRPC:Orai1 heterodimers are shown, but the association of TRPC and Orai1 has not been established unequivocally; thus, SOCE may proceed via TRPC1 or TRPC4 alone. Transforming growth factor $\beta 1$ (TGF$\beta 1$) and high extracellular glucose concentrations suppress TRPC1-mediated SOCE by activating miR135a expression, while urotensin II and high glucose activate TRPC4-mediated SOCE. **Panel (c)**: Factors modulating SOCE via Orai1 homodimers, and SOCE-inhibited ECM synthesis. Orai1-mediated SOCE is activated by STIM1, protein kinase C (PKC) α, and glucagon-like peptide-1 receptor (GLP-1R), and inhibited by PKC β_1. SOCE suppresses ECM synthesis directly, and indirectly by (1) activating release of interleukin-6 (IL6) which, upon binding its receptor (IL6-R), activates GLP-1R, and (2) inhibiting TGF$\beta 1$-receptor (TGF$\beta 1$R) activation of smad 1 and 3 phosphorylation.

SOCC-mediated Ca^{2+} entry in MCs. Interestingly, agonists like epidermal growth factor acting through the tyrosine kinase receptors failed to trigger detectable SR/ER Ca^{2+} release and seemed to activate the SOCE in an IP_3-independent manner with phospholipase C as the crucial element.[24] Unlike G protein-coupled receptors, tyrosine kinase receptors control a diverse array of downstream cellular pathways, affording complex regulation of SOCE.

Because protein kinase C (PKC) is activated by the phospholipase C/diacylglycerol pathway, it too might activate SOCC. Indeed, PKC differentially controls SOCE in diverse cells. In our study, Calphostin C, a specific PKC inhibitor, diminished TG-activated SOCE in cultured human MCs. Furthermore, PKC activation or introduction of PKC catalytic subunits prevented SOCE rundown in inside-out patches. This PKC-mediated SOCE activation was attributed to the PKCα isoform.[25] However, other studies contradicted these findings. Mene *et al*.[26] demonstrated that PKC inhibited the SOCE induced by angiotensin II. The finding that STIM1's interaction with Orai1 mediates SOCE has prompted research focused on these two proteins. Kawasaki *et al.* used RNA interference, transfection with green fluorescent protein-tagged PKC isoforms and Flag-tagged wild type or mutant Orai1 to demonstrate that phosphorylation of Orai1's Ser-27 and Ser-30 residues by the PKC$\beta 1$ isoform was responsible for SOCE suppression in HEK293 cells.[27] The apparent discrepancies among these studies might be due to differences in the SOCC machinery among cell types, the activating stimuli, or activation of different PKC subtypes.

TRPC channels in MCs

TRPC channels, those most closely related to the transient receptor potential superfamily of Drosophila, exist throughout the kidneys and are likely SOCC candidates. Species variations are reported in the renal distribution of

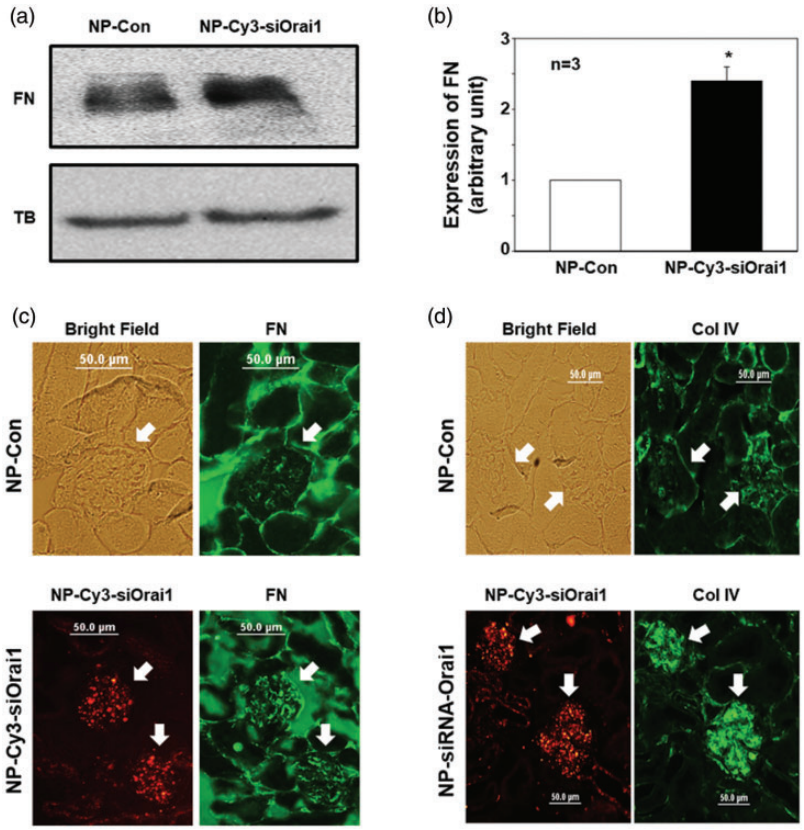

Figure 2. Increased glomerular ECM protein content in mice after *in vivo* knockdown of Orai1 in MCs using targeted nanoparticle (NP) delivery system with Cy3 tagged siOrai1. Panels (a), (b): Immunoblot analysis of renal cortex extracts showing fibronectin (FN) content in the cortex of kidney from the mice treated with control NP (NP-Con) and NP-Cy3-siOrai1 (knockdown of Orai1). Tubulin-α (TB) is a loading control. Panel (a): representative immunoblot; panel (b): summary data. *$P < 0.05$ vs. NP-Con. Panels (c) and (d): immunohistochemistry showing abundance of (c) FN and (d) collagen IV (Col IV) in glomeruli of the mice treated with NP-Con and NP-Cys-siOrai1. Both FN and Col IV are stained green. In NP-Con, a bright-field image was captured to show the glomerulus. In NP-Cy3-siOrai1, the distribution of NP-Cy3-siOrai1 is indicated by Cy3 signals (red). Arrows indicate glomeruli. Original magnification $\times$200. (Adapted from Wu *et al.*[37]).

TRPC isoforms. Out of seven known isoforms, TRPC1 and TRPC4-α are found in mouse MCs, where TRPC4-α functions as SOCC.[28] On the other hand, cultured human MCs express TRPC1, 3, 4, and 6, and TRPC1 interacts with TRPC4 and TRPC6.[29] Only TRPC1 is reported in rat MC cultures.[30] However, the mere presence of TRPC does not prove it operates as SOCC or regulates SOCE. In cultured human MCs, the TRPC1/TRPC4 complex may interact with STIM1 to regulate SOCE as demonstrated by the inhibition of TG-induced membrane currents after knockdown of STIM1 or either TRPC1 or TRPC4.[31] Interestingly, the gene expression approach revealed that Orai1 was essential for functional activation of SOCE by the TRPC1-STIM1 interaction in HEK293 cells.[32,33] On the other hand, protein kinase G-mediated phosphorylation of vasodilator-stimulated phosphoprotein (VASP), a focal adhesion molecule highly expressed in MCs, causes VASP to associate with TRPC4 to inhibit the associated SOCE (Figure 1(b)).[34] Although the precise mechanism of this inhibition is not clear, phosphorylated VASP is hypothesized to inhibit SOCE by dissociating TRPC4 from the SOCC complex.

Orai1 and STIM1 in MCs

Discovered in 2005 and 2006, respectively, Orai1 and STIM1 are the central elements of SOCE-mediated signaling. Researchers have manipulated these proteins with molecular tools to elucidate the role of SOCC in different organ systems. Although diverse roles of SOCE were observed in different cell types like lymphocytes, T cells, vascular smooth muscle cells, and cardiomyocytes, the physiological and pathophysiological significance of SOCE in kidneys initially was obscure. The abundance of Orai1 was reported in rat[35] and human[36] MCs several years after Orai channels were discovered.

Over the last decade, our laboratory has worked extensively to unveil the role of SOCC in MCs. In cultured human MCs, our studies of SOCC regulation of ECM proteins like fibronectin and collagen IV demonstrated that TG-induced SOCE suppresses ECM protein synthesis and deposition (Figure 1(c)). STIM1 overexpression reduced, and Orai1 knockdown increased, ECM protein formation.[37] These findings, which were confirmed by other investigators,[38] are discussed further in the next paragraph. The absence of specific MC markers prevents selective manipulation of gene expression in MCs. After a novel targeted nanoparticle delivery system was used to knockdown the Orai1 channel protein specifically in mouse MCs, the contents of glomerular fibronectin and collagen IV increased (Figure 2)[37] causing significant mesangial expansion and a fibrotic glomerular phenotype. The principal limitation of this technique is that the nanoparticles are too small to

accommodate the target protein's overexpression plasmid. Hence, the nanoparticles cannot be used to evaluate the possible protective effects of increased SOCE in renal diseases associated with increased ECM deposition. Nevertheless, these studies show SOCE downregulates MC synthesis of ECM proteins. Under physiological conditions, SOCE in MCs could conceivably be activated to maintain the balance of ECM protein synthesis and degradation.

Pathophysiological role of SOCC in MCs

SOCE impairment has been implicated in numerous disorders such as immunodeficiency, myopathy, vascular diseases, and many types of cancer.[39–41] Aging related disorders like Alzheimer's disease present with STIM1/STIM2 downregulation and reduced neuronal SOCE. Dysfunction of SOCE-mediated Ca^{2+} signaling resulted in dendritic spine deformity in a mouse model of the PS1-M146V mutation of familial Alzheimer disease.[42,43] However, another study in mice reported that TRPC6-associated SOCE was increased by the PSEN1ΔE9 mutation, found in familial Alzheimer disease in the Finnish population.[44] In aged rats, STIM1 and Orai1 contents were attenuated with reduced SOCE in MCs. The decreased MC proliferation in aged rats was attributed to reduced STIM1-Orai1 interaction and SOCE in the Ca^{2+} depleted MCs.[35]

Imbalances in matrix protein synthesis and degradation leading to fibrotic protein accumulation in the glomerular mesangium initiate the pathogenesis of renal fibrosis in chronic kidney disease.[20] Numerous extracellular stimuli including the vasoactive peptides endothelin-1, angiotensin II, and urotensin II can stimulate pro-fibrotic signaling in MCs. Diabetic nephropathy is the leading cause of chronic kidney disease involving ECM protein accumulation. High extracellular glucose (HG) impacts Ca^{2+} signaling in various cells, including MCs. Mene *et al.* demonstrated inhibition of resting and vasopressin-induced SOCE in isolated rat MCs exposed to 30 mM glucose for five days, modeling the hyperglycemic diabetic milieu. Reversal of SOCE inhibition by a PKC activator, phorbol myristate acetate, implicated PKC inactivation in HG suppression of SOCE.[45] Nutt *et al.* reported that exposure of cultured rat MCs to 30 mM glucose for up to seven days depressed endothelin-induced receptor-operated Ca^{2+} influx, although SOCE was unimpaired.[23] While 8–24 h exposure to 25 mM glucose decreased Orai1 content in rat and human MCs,[46] we found that prolonged (7 days) HG exposure enhanced TG-induced SOCE in cultured human MCs and in parallel increased abundance of STIM1 and Orai1 proteins.[36] This HG-induced SOCE was inhibited by GSK-7975A, a specific blocker of SOCC, and by knockdown of Orai1 expression. Whole-cell patch-clamp experiments showed that chronic HG markedly augmented store-operated Ca^{2+} currents. Interestingly, chronic HG attenuated angiotensin II-induced receptor-operated Ca^{2+} entry, measured after depletion of intracellular Ca^{2+} stores, and re-addition of Ca^{2+} to induce SOCE. Concordantly, diabetic rats showed increased STIM1 and Orai1 contents in the renal cortices and glomeruli.

Divergent findings in the above-described studies might be ascribable to different SOCE measurement methods and/or species differences. For example, Mene *et al.*[45] assessed agonist-induced SOCE as the Ca^{2+} influx after addition of Ca^{2+} to nominally Ca^{2+}-free media. However, this Ca^{2+} entry could be mediated by both ROCC and SOCC. Nutt *et al.*[23] measured Ca^{2+} influx in cultured rat MCs treated with endothelin-1 or TG in the presence of 1 mM Ca^{2+} or Ca^{2+} free solution. While our findings are concordant with the reports of increased Orai1 content in diabetic rat renal cortex, the role of this enhanced SOCE in MCs remains enigmatic.

Our lab further examined the role of SOCC in human MCs. Inhibition of SOCC with 2-aminoethyl diphenylborinate augmented angiotensin II-induced fibronectin content, whereas SOCC activation with TG abrogated HG- and transforming growth factor β1 (TGF-β1)-stimulated fibronectin and collagen IV matrix accumulation.[37] The inhibition of collagen IV and fibronectin abundance by SOCE is mediated through suppression of pro-fibrotic smad1 and smad3 phosphor-activation (Figure 1(c)). Accordingly, TG treatment of human MCs attenuated angiotensin II-, HG-, and TGF-β1-induced smad1 phosphorylation as well as TGF-β1-induced smad3 phosphorylation. Also, targeted downregulation of SOCE in MCs in mice, using the nanoparticle incorporated small-interfering RNA (siRNA) against Orai1, increased the contents of both smad1 and smad3 in the glomeruli.[47,48] However, the mechanism whereby SOCE inhibits smad1 or smad3 phosphorylation (Figure 1(c)) is not yet clear. SOCE may inhibit the kinase activity of a Type-II TGF-β1 receptor dimer that phosphorylates the Type-I dimer, or either activate a Ca^{2+}-dependent phosphatase or inhibit a protein kinase that phosphorylates smad1/3.

Another interesting study demonstrated that liraglutide, an agonist of the G protein-coupled glucagon-like peptide-1 receptor (GLP-1R), inhibits HG-induced ECM production by MCs through activation of SOCC[49] and Wnt/β-catenin signaling pathways.[50] Of note, GLP-1R's natural ligand glucagon-like peptide-1 is used clinically to promote insulin secretion to treat type 2 diabetes. The Wnt/β-catenin pathway is downregulated in diabetic kidneys, and liraglutide is renoprotective in diabetic nephropathy.[51,52] Crosstalk between the SOCE and Wnt/β-catenin pathways in MCs is not known. Micro-RNAs like miR-29a promoted Wnt/β-catenin signaling in MCs.[53] An important question is whether SOCE activates miR-29a to augment the Wnt/β-catenin signaling machinery to downregulate ECM proteins.

Upregulation of GLP-1R in MCs may constitute another mechanism, whereby SOCE inhibits ECM protein deposition. Recently we demonstrated that TG-induced SOCE increased the content of the cytokine interleukin-6 in human MCs.[54] Interleukin-6 is a pleiotropic cytokine that can be pro- or anti-inflammatory depending on the specific pathological stimulus, the cells affected, the molecular signaling pathways activated, and/or its interactions with other cytokines and immune factors. We found that SOCE

favors interleukin-6 production in human MCs in which overexpression of interleukin-6 with its receptor partially increased GLP-1R abundance and also attenuated fibronectin and collagen IV production (Figure 1(c)).[54] Collectively, these studies established that the accentuated SOCC and SOCE in MCs chronically exposed to HG is very likely a defense mechanism to nullify the effects of angiotensin II, advanced glycation end products, and other pro-fibrotic factors that might predominate in the diabetic kidneys.

In MCs, TRPC1 and TRPC4 can mediate SOCE,[28,31] while nitric oxide inhibited TRPC4-associated SOCE via the protein kinase G/VASP pathway (Figure 1(b)).[34] The downstream mechanism of SOCE inhibition by VASP would be particularly interesting since nitric oxide and its metabolites are well-known mediators of inflammatory renal diseases[55] and endothelial cell preservation and regeneration attenuated nephrotoxic nephritis in VASP-null mice.[56]

Like Orai1-mediated SOCE,[37] the TRPC1-mediated SOCE also inhibited HG induction of ECM proteins in human MCs.[57] The micro RNA miR-135a was markedly upregulated in renal tissue from diabetic db/db mice and patients with diabetic nephropathy, and in human MCs and proximal tubular epithelial cell exposed to HG and TGFβ. miR-135a attenuated SOCE in human MCs by reducing TRPC1 abundance and, consequently, augmented fibronectin and collagen I synthesis (Figure 1(b)).[57] However, urotensin II activation of TRPC4-mediated SOCE (Figure 1(b)) promoted MC proliferation and ECM protein accumulation in mouse MCs exposed to HG.[58] These variances in the TRPC4-mediated SOCE in MCs might be due to Ca^{2+} influx through other channels like ROCCs, because selective TRPC4 channel inhibition could not completely abolish the Ca^{2+} influx upon re-addition of Ca^{2+}, and an L type Ca^{2+} channel blocker, nimodipine, did not prevent Ca^{2+} influx through the ROCCs. Figure 1 summarizes the signaling mechanisms upstream and downstream of SOCC which may modulate ECM protein synthesis in MCs, and the points where HG and other factors impact these mechanisms.

SOCE in podocytes

Podocytes, the highly specialized visceral epithelial cells of the renal corpuscle that surround the glomerular capillaries, partner with the capillary endothelium to generate and maintain the glomerular basement membrane. Collectively, the capillary endothelium, basement membrane, and podocyte epithelium comprise the interface that effects glomerular filtration. The diaphragms spanning the narrow slits between the interdigitating podocyte pedicels comprise a selective barrier imposing steric hindrance on filtration of protein-size molecules. Various TRPC channels are found in the podocytes with some variations among species. Rat podocytes express TRPC3 and TRPC6,[30] while mRNAs for TRPC1, 2, 5, and 6 are expressed in cultured murine podocytes.[59] The most significant and widely studied are the TRPC6 channels present across the podocyte cell body, processes, and pedicles,[59] which are associated with many glomerular disorders including focal segmental glomerulonephritis and diabetic nephropathy. Although the role of podocyte TRPC6 in SOCE is unclear, podocytes demonstrated TG-inducible SOCE which was unaltered by HG. Moreover, TRPC6 knockdown had no effect on SOCE but diminished receptor-operated Ca^{2+} influx.[60] It is not yet known if other TRPC channels are associated with podocyte SOCC complexes. Readers are referred to a recent review on the role of podocyte TRPC channels in chronic kidney disease.[61] Another recent study demonstrated STIM in podocytes and showed that STIM overexpression is associated with increased epithelial-mesenchymal transition of podocytes in diabetic nephropathy.[62] Further evidence of STIM and Orai channels in podocytes might open up new avenues of research since epithelial-mesenchymal transition mobilizes many signaling pathways associated with renal fibrosis.[63]

SOCE in renal tubular cells

Orai1 and STIM1 in renal tubular cells

The divergent impacts of SOCC in different cell types exemplify the remarkable diversity of cell types in kidney. In sharp contrast to MCs, Mai *et al.* demonstrated that Orai1 knockdown prevented TGF-β1- or angiotensin II-induction of fibrotic proteins in cultured human proximal tubular epithelial (HK2) cells, renal peritubular interstitium of mice subjected to unilateral ureteric obstruction, and $ApoE^{-/-}$ mice consuming a high-fat diet.[38] Kidney biopsies from human patients with fibrotic nephropathies, e.g. focal proliferative sclerosis and tubule-interstitial nephritis showed increased Orai1 staining in proximal tubular epithelial cells, while $ApoE^{-/-}$ mice consuming a high-fat diet had increased Orai1 content in the renal cortex. In addition, studies in HK2 cells showed that Orai1 knockdown prevented TGF-β1 driven epithelial-mesenchymal transition, and implicated suppression of smad2/3 phosphorylation as the possible mechanism for this effect. Those studies also established that Orai1 knockdown increases ECM proteins in human MCs, as our study demonstrated,[37] confirming the cell specificity of SOCE's effects. Some differences in experimental design and mouse models in Mai *et al.*'s study vs. ours should be noted. While we used the targeted nanoparticle delivery to suppress Orai1 expression specifically in MCs of C57BL6 mice, Mai *et al.* used systemic knockdown of Orai1, hence the findings *in vivo* might not be attributed specifically to changes in the proximal tubules. Also, smad2 and smad3 may have diametrically opposite downstream effects, as smad3 is profibrotic and smad2 antifibrotic.[64,65] Whether the decrease in phosphorylated smad2/3 in this study represented decreased phosphorylation of one or both isoforms was not clear. Another possibility is that the effects of SOCE are cell type- or context-specific. Recently we demonstrated that SOCE behaves differently in human MCs and proximal tubular epithelial cells where the two cell types showed opposite responses to overexpression of a fusion protein of interleukin-6 and its receptor.[54]

Reabsorption of albumin by the proximal tubular cells, a crucial mechanism to preserve plasma colloid osmolality, is

impaired in chronic kidney diseases like diabetic nephropathy.[66] Orai channels play a major role in this receptor-mediated albumin endocytosis, in which apical membrane Orai1/STIM1 complexes colocalize with clathrin to mediate the endocytosis.[67] Kidney sections of diabetic nephropathy patients demonstrated downregulation of Orai 1–3, while treatment of human proximal tubular epithelial cells with 25 mM HG blunted Orai protein content. STIM–Orai3-mediated SOCE also contributed to the pathogenesis of crystal nephropathy. When insoluble crystals in the tubular fluid, which are normally excreted in urine, accumulate in the tubular lumen, they may promote acute or chronic kidney injury. Some crystals, particularly those containing Ca^{2+} salts, adhere to the tubular wall and are taken up by the epithelial cells where they may initiate cellular injury. Uptake of Ca^{2+} salt crystals into proximal tubule-derived HK2 cells elicited a sustained increase in intracellular Ca^{2+} via STIM–Orai3, provoking an ER stress response, oxyradical-induced cell damage, and apoptosis.[68] Suppressing SOCC gene expression reversed these changes.

The pathogenesis of autosomal dominant polycystic kidney disease (ADPKD) involves loss of function mutations in the PKD1 or PKD2 genes encoding polycystin-1 and -2, respectively, which disrupt Ca^{2+} homeostasis and provoke cyclic AMP-dependent cyst formation in renal tubular cells. P100, a cleavage product of polycystin-1, suppresses SOCE by inhibiting STIM1 translocation to the plasma membrane after ER Ca^{2+} depletion,[69] and may be a protective mechanism against cell proliferation by reducing the plasma membrane STIM1 and subsequent SOCE. Increased abundance of STIM1 and IP_3 receptor in polycystin-1-null proximal tubular cells and polycystin-1 knockout mice modeling ADPKD leads to SOCE-related Ca^{2+} dysregulation. Knockdown of STIM1 reduced cyclic AMP and ER Ca^{2+} release, and suppressed cyst formation and growth.[70] A prototypical member of a TRP protein subfamily, polycystin-2 can function independently as a non-selective cation channel. It can also modulate cellular Ca^{2+} responses by regulating other molecules or channels participating in intracellular Ca^{2+} homeostasis like IP_3 receptors, STIM1, TRPC1, TRPV4.[71–73] Mutation of polycystin-2 provokes imbalances of Ca^{2+} and cyclic AMP, in turn producing a proliferative, secretory proximal tubular epithelial phenotype.[74]

Dysfunctional SOCE may contribute to impairments in water reabsorption in collecting ducts characterizing nephrogenic diabetes insipidus. In spontaneously hypertensive SHR-A3 rats in which SOCE is disrupted by a STIM1 mutation, collecting duct epithelium lacked the vasopressin-induced sustained elevation of intracellular Ca^{2+} necessary for aquaporin AQP2 expression and translocation to the apical membrane to effect water reabsorption.[75] $Stim1^{fl/fl}$ Ksp-cre mice with specific knockout of STIM1 in the epithelial cells of collecting ducts and thick ascending limbs of Henle's loop and fed a high protein (40%) diet demonstrated increased Ca^{2+} excretion and impaired urinary concentrating ability vs. wild type mice. STIM 1 may modulate vasopressin signaling by regulating Ca^{2+}-dependent adenylyl cyclase activity and cyclic AMP formation in collecting duct epithelium, or by altering vasopressin receptor abundance or AQP2 translocation to the apical membrane.[76] These studies indicate a pivotal role of SOCE in water homeostasis, and delineating the mechanisms mobilized by SOCE could identify potential therapeutic targets for nephrogenic diabetes insipidus.

TRPC channels in renal tubular cells

TRPC channels are selectively and differentially positioned along the renal tubules[30] implying they play physiological and pathophysiological roles in phospholipase C-dependent renal cellular responses. TRPC1 channel protein is expressed in the apical membrane of proximal tubular epithelial cells where it co-localizes with aquaporin channel AQP1, while TRPC3 and TRPC6 in collecting duct principal cells co-localize with vasopressin-activated AQP2. TRPC3 resides in the apical membrane both *in vivo* and in cultured cells, whereas TRPC6 exists in both the apical and basolateral membranes of principal cells. The role of TRPC channels in the regulation of SOCE in the tubular cells in different segments of nephron is unclear at present. Interestingly, although Orai1 knockdown attenuated TGF-β- or angiotensin II-induced fibronectin and collagen IV expression in HK2 cells,[38] HG- or TGF-β-induced miR-135a attenuated SOCE in HK2 cells by reducing TRPC1 expression and subsequently augmenting fibronectin and collagen I synthesis.[57] TRPC1-associated SOCC complexes could possibly activate downstream pathways distinct from Orai-mediated SOCE.

SOCE in renal microvasculature

In addition to the well-known voltage-operated Ca^{2+} channel (VOCC), vascular smooth muscle cells (VSMC) rely on SOCC and ROCC for their Ca^{2+} signaling. Studies in pulmonary artery, aorta, and portal vein identified a crucial role for SOCE in determining the basal tone of the VSMCs and their contractile responses to agonists. Although this section focuses on studies conducted in renal vasculature, research in other vascular beds has identified molecular mediators of Ca^{2+} signaling that also may operate in the renal vasculature. Regarding the complex role of SOCE in different types of muscle cells, the reader is referred to a recent review and the original literature cited therein.[39] TRPC channels contribute to signaling pathways initiated by G protein coupled receptors that modify vasomotion and cellular proliferation. However, the TRPC subunits of SOCC may vary in different blood vessels, so caution is warranted when extrapolating studies in specific vasculature to other vascular beds and organs.

In the renal microvasculature SOCE have been identified by several research groups.[77,78] The renal microcirculation, which comprises two arterioles and two capillary beds in series, is unique in its anatomic and physiological characteristics. In the cortex, the hemodynamics of the afferent and efferent arterioles govern glomerular filtration. The efferent arterioles give rise to the peritubular capillary plexus, which, due to its proximity to the cortical tubular segments, returns to the systemic circulation the enormous amounts of filtered water and solutes reabsorbed by the

tubular epithelium. The vascular resistances of the afferent and efferent arterioles exert powerful regulation of renal vascular resistance, glomerular filtration, and peritubular capillary blood flow. The first hint of divergent Ca^{2+} signaling mechanisms in the afferent and efferent arterioles was the finding that membrane depolarization-induced Ca^{2+} entry was abolished by the Ca^{2+} channel antagonist nifedipine in the afferent but not efferent arterioles.[79] Confirming this heterogeneity, Nagahama *et al.* demonstrated in perfused hydronephrotic rat kidneys that angiotensin II-induced constriction of efferent arterioles was abrogated by a PKC inhibitor yet was refractory to VOCC inhibition, while constriction of afferent arterioles was eliminated by a VOCC inhibitor but only partially attenuated by a PKC inhibitor.[78] Further experiments confirmed the dominance of the VOCC in the afferent and SOCE in the efferent arterioles. Angiotensin II-induced Ca^{2+} influx was suppressed by nifedipine in isolated afferent but not efferent arterioles, while depolarization-induced Ca^{2+} influx was only detected in afferent arterioles. The sarcoplasmic reticular Ca^{2+} ATPase inhibitor cyclopiazonic acid induced a sustained Ca^{2+} influx in efferent arteriolar VSMCs that was unaltered by nifedipine.[80]

In contrast, studies by Fellener and Arendshorst[77] in VSMC freshly isolated from rat preglomerular vessels contradicted the apparent distinctions between the afferent and efferent arteriolar VSMCs. In these studies, the depletion of internal Ca^{2+} stores with cyclopiazonic acid or TG increased appreciably the Ca^{2+} entry in the presence of VOCC blockers. Nifedipine or verapamil only partially attenuated the vasopressin-induced rise in intracellular Ca^{2+} concentration. Their finding of SOCC in the afferent arterioles gained pathological significance when Fellener and Arendshorst reported that SOCE elicited by the vasopressin-1 receptor agonist, [Phe^2, Ile^3, Orn^8]-vasopressin, was exaggerated in preglomerular VSMC from young salt-sensitive hypertensive rats, while VOCC was not affected.[81] Concordantly, Fallet *et al.* attributed the VSMC contractile response to vasopressin to Ca^{2+} release from intracellular stores in both afferent and, more substantially, efferent arterioles. Furthermore, only in afferent arterioles do VOCC mediate vasopressin-induced vasoconstriction.[82] SOCE also was identified by fura-2 ratiometric fluorescence in interlobular arteries, where cyclopiazonic acid- and TG-induced Ca^{2+} store depletion activated robust Ca^{2+} influx. These responses were dose-dependently suppressed by SOCC antagonists Gd^{3+} and 2-aminoethyl diphenylborinate.[83] Guan *et al.* reported that sphingosine-1-phosphate, a bioactive sphingolipid metabolite and signaling molecule, elicited segment-specific vasoconstriction of renal arcuate and interlobular arteries and afferent arterioles, yet without affecting the efferent arterioles.[84] Sphingosine-1-phosphate's effect on the afferent arterioles was specifically mediated by the VOCC-dependent rho/rho kinase pathway.[85]

The divergent findings from those studies might represent variations among the segments of the preglomerular vasculature, the diameter of the vessels, variability of cell types or receptor distribution within different arteriolar segments in the same bed, different protocols and buffers for isolating the microvessels, and/or use of different vasoconstrictors. Nevertheless, many vasoactive compounds have been shown to confer distinct actions on afferent vs. efferent arterioles attributable to differences in Ca^{2+} channel mechanisms.

Members of the TRPC channel family have been found to mediate receptor-operated Ca^{2+} entry and SOCE in various cell types, including vascular endothelial and smooth muscle cells.[86] Both aorta and renal preglomerular resistance vessels expressed mRNA for TRPC1, 3, 4, 5, and 6, while TRPC2 and TRPC7 transcripts were not detected. TRPC3, TRPC1, and TRPC6 protein contents are greater in preglomerular resistance vessels than in aorta.[87] TRPC4 channels are also found in the descending vasa recta within the renal medulla.[88] The possibility that TRPC channels function as SOCC in vascular smooth muscle and other tissues continues to inspire debate. Recently, using a systematic approach with patch clamp recording, GFP-PLCδ1-PH imaging, protein co-localization analysis, and RNA interference, Shi *et al.* confirmed in VSMCs that TRPC1 interacted with STIM1 after Ca^{2+} store depletion and activated Ca^{2+} channels via the G protein αq subunit/phospholipase C-$_{\beta 1}$/PKC pathway.[89] TRPC3 and TRPC6 are not generally known to participate in SOCE. The associations of STIM, Orai, and other TRPC channels, and their role as SOCC, merit evaluation in the renal vasculature. Also, as SOCE is robust in vascular endothelial cells and its Ca^{2+} influx can increase endothelium-dependent myogenic tone, the influence of endothelial SOCE in studies with endothelium-intact vessels cannot be excluded.

SOCE in intrarenal immune cells

SOCE is a crucial Ca^{2+} entry pathway in diverse immune cells like lymphocytes, macrophages, dendritic cells[90,91] and mutation of Orai1 elicits a severely immunodeficient state.[5] Activation of immune cells may be beneficial or detrimental depending on the degree of activation, immune cell types contributing their specific cytokines to the response, and the nature and severity of tissue injury.[92] Recently, T lymphocyte activation was shown to contribute to the transition from acute kidney injury to chronic kidney disease.[93] In rats, accumulation of $CD4^+$/IL-17^+/$Orai1^+$ cells (Th17 cells) secreting IL-17 was observed within the first three days after renal ischemia-reperfusion, and resolved over seven days. Treatment of these acute kidney injury-primed $CD4^+$ cells with Angiotensin II or elevated Na^+ increased the percentage of cells expressing IL-17 and their abundance of IL-17 mRNA. SOCE pathway inhibitors completely blocked this increase in IL-17. Subsequent insult with high salt diet in rats recovered from renal ischemia/reperfusion intensified chronic kidney disease including increased renal interstitial fibrosis, urinary albumin excretion, and kidney injury molecule-1 (KIM-1) abundance. These changes were abolished when the rats were treated with inhibitors of SOCE pathway YM58483/BPT2.[93] SHR-A3 rats with truncated STIM1 demonstrated increased glomerular and interstitial fibrosis, albuminuria, urinary renal injury markers, e.g. neutrophil gelatinase-associated lipocalin (NGAL), osteopontin (OPN), and KIM-1, and also

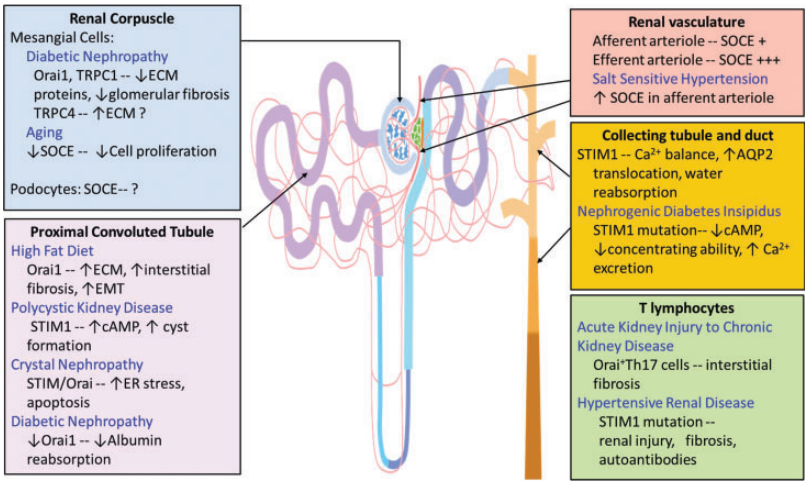

Figure 3. Tubular and microvascular locations of dysfunctional SOCE in renal diseases. The boxes represent different locations within the nephron and renal microvasculature. Blue text identifies the clinical pathologies related to SOCE in each location, and black text lists possible effects of SOCE in the different renal cell types. TRPC: transient receptor potential canonical; ECM: extracellular matrix; SOCE: store-operated Ca^{2+} entry; STIM: stromal interaction molecule; EMT: epithelial-mesenchymal transition; cAMP: cyclic AMP; ER: endoplasmic reticulum; AQP: aquaporin.

extensive T cell and B cell infiltration in kidneys. STIM1 mutation in T cells of these SHR-A3 rats contributed to defective proliferation and cytokine production by T cells, reduced T_{reg} numbers as well as increased IgM and IgG immunoglobulin deposition in the kidneys indicating autoantibody formation. STIM1 gene rescue restored the lymphocyte function and reversed the pathological hallmarks of renal injury except albuminuria and IgG deposition.[94]

Clinical implications of SOCE in kidney

SOCC channels are ubiquitous, existing throughout the body. SOCE is implicated in diverse renal conditions (Figure 3). For example, Orai1-mediated SOCE suppresses ECM synthesis in MCs in the setting of diabetic nephropathy, while Orai1 increases ECM abundance in proximal tubular cells and renal fibrosis in mice consuming high-fat diets. While STIM1 is important for physiological water handling in the collecting tubules, it might be detrimental in ADPKD in proximal tubular epithelium. SOCE inhibition prevents Th17 cell-mediated transition of acute to chronic kidney disease, while loss of STIM1 function in SHR-A3 rats is associated with renal injury and autoantibody formation. These diverse effects of SOCE in different renal cells and settings make treatment of SOCE-related renal conditions extraordinarily challenging. Global inhibition/activation of SOCE might prove beneficial in one renal cell type but produce detrimental effects in other cell types, so SOCE must be carefully evaluated in the context of the specified disease. Cell specific knockdown or overexpression of SOCC proteins or their modulators, or targeted delivery of pharmacological activators/inhibitors of SOCE, might be effective approaches for evaluating SOCE. Strategies like use of targeted nanoparticle delivery of Orai1 siRNA to knockdown Orai1 specifically in MCs *in vivo* may have implications for eventual application in humans.

Concluding remarks

The growing body of evidence reviewed here unequivocally demonstrates that SOCC is essential to signaling mechanisms controlling diverse physiological functions of several distinct renal cell types. SOCE is regulated by multiple agonists and intracellular messengers affecting these pathways in physiological and pathological settings. However, regulation of SOCE varies among renal cell types. As renal cells are highly specialized, the molecular components of their signaling cascades vary, such that SOCE elicits diverse responses. Variations in SOCC protein composition likely contribute to the diversity of cell type-specific responses. Variations among species, experimental approaches, and tools for assessing SOCE also likely contribute to divergent literature reports of these phenomena. When analyzing SOCE, it is essential that other Ca^{2+} channels are masked or silenced. Furthermore, the contributions of TRPC channels must be interpreted carefully since Ca^{2+} entry through SOCC itself can activate individual TRPC channels independent of intracellular Ca^{2+} store depletion. The possibilities that mitochondrial dysfunction affects SOCE in renal cells, or that SOCE regulates renal electrolyte and acid–base balance, merit investigation. siRNA approaches and specific cell-targeted gene knockdown strategies have enabled the role and regulation of SOCC in ECM protein dynamics, renal fibrosis, and water homeostasis to be deciphered. Increasing understanding of SOCE molecular components will permit interrogation of the therapeutic potential of these pathways for treatment of kidney diseases. Since SOCE affects renal cell types differentially, cell-specific targeted delivery of agents to modulate SOCC holds particular promise for addressing SOCE-related disorders.

AUTHORS' CONTRIBUTIONS

All authors contributed equally to the writing of this manuscript. SC prepared the first draft of the manuscript. PYS and

YT contributed to the later drafts. SC and RTM prepared the figures. Finally, RTM and RM provided critical additions and revisions to the manuscript. All authors edited the manuscript and reviewed and approved the final version for submission.

DECLARATION OF CONFLICTING INTERESTS

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

FUNDING

The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: The work was supported by National Institutes of Health (NIH/NIDDK) [Grant number 5RO1DK079968-01] to R. Ma and a Postdoctoral Fellowship from American Heart Association Southwestern Affiliate [Grant number 20POST35210685] to S. Chaudhari.

ORCID IDS

Sarika Chaudhari https://orcid.org/0000-0002-2590-6365
Robert T Mallet https://orcid.org/0000-0001-7388-9419

REFERENCES

1. Putney Jr JW. A model for receptor-regulated calcium entry. *Cell Calcium* 1986;**7**:1–12
2. Randriamampita C, Tsien RY. Emptying of intracellular Ca2+ stores releases a novel small messenger that stimulates Ca^{2+} influx. *Nature* 1993;**364**:809–14
3. Fasolato C, Hoth M, Penner R. A GTP-dependent step in the activation mechanism of capacitative calcium influx. *J Biol Chem* 1993;**268**:20737–40
4. Irvine R. Quanta'Ca^{2+} release and the control of Ca2+ entry by inositol phosphates – a possible mechanism. *FEBS Lett* 1990;**263**:5–9
5. Feske S, Gwack Y, Prakriya M, Srikanth S, Puppel S-H, Tanasa B, Hogan PG, Lewis RS, Daly M, Rao A. A mutation in Orai1 causes immune deficiency by abrogating CRAC channel function. *Nature* 2006;**441**:179–85
6. Roos J, DiGregorio PJ, Yeromin AV, Ohlsen K, Lioudyno M, Zhang S, Safrina O, Kozak JA, Wagner SL, Cahalan MD. STIM1, an essential and conserved component of store-operated. Ca^{2+} channel function. *J Cell Biol* 2005;**169**:435–45
7. Vig M, Beck A, Billingsley JM, Lis A, Parvez S, Peinelt C, Koomoa DL, Soboloff J, Gill DL. Fleig A. CRACM1 multimers form the ion-selective pore of the CRAC channel. *Curr Biol* 2006;**16**:2073–9
8. Yuan JP, Zeng W, Huang GN, Worley PF, Muallem S. STIM1 heteromultimerizes TRPC channels to determine their function as store-operated channels. *Nat Cell Biol* 2007;**9**:636–45
9. Desai PN, Zhang X, Wu S, Janoshazi A, Bolimuntha S, Putney Jw, Trebak M. Multiple types of calcium channels arising from alternative translation initiation of the Orai1 message. *Sci Signal* 2015;**8**:ra74
10. Vaeth M, Yang J, Yamashita M, Zee I, Eckstein M, Knosp C, Kaufmann U, Jani PK, Lacruz RS, Flockerzi V. ORAI2 modulates store-operated calcium entry and T cell-mediated immunity. *Nat Commun* 2017;**8**:1–17
11. Hoth M, Penner R. Depletion of intracellular calcium stores activates a calcium current in mast cells. *Nature* 1992;**355**:353–6
12. Parekh AB, Putney Jr JW. Store-operated calcium channels. *Physiol Rev* 2005;**85**:757–810
13. Floege J, Eitner F, Alpers CE. A new look at platelet-derived growth factor in renal disease. *J Am Soc Nephrol* 2008;**19**:12–23
14. Kikkawa Y, Virtanen I, Miner JH. Mesangial cells organize the glomerular capillaries by adhering to the G domain of laminin α5 in the glomerular basement membrane. *J Cell Biol* 2003;**161**:187–96
15. Leveen P, Pekny M, Gebre-Medhin S, Swolin B, Larsson E, Betsholtz C. Mice deficient for PDGF B show renal, cardiovascular, and hematological abnormalities. *Genes Dev* 1994;**8**:1875–87
16. Quaggin SE, Schwartz L, Cui S, Igarashi P, Deimling J, Post M, Rossant J. The basic-helix- loop-helix protein pod1 is critically important for kidney and lung organogenesis. *Development* 1999;**126**:5771–83
17. Kreisberg J, Venkatachalam M, Troyer D. Contractile properties of cultured glomerular mesangial cells. *Am J Physiol Renal Physiol* 1985;**249**:F457–63
18. Abboud HE. Mesangial cell biology. *Exp Cell Res* 2012;**318**:979–85
19. Mason RM, Wahab NA. Extracellular matrix metabolism in diabetic nephropathy. *J Am Soc Nephrol* 2003;**14**:1358–73
20. Bülow RD, Boor P. Extracellular matrix in kidney fibrosis: more than just a scaffold. *J Histochem Cytochem* 2019;**67**:643–61
21. Menè P, Teti A, Pugliese F, Cinotti GA. Calcium release-activated calcium influx in cultured human mesangial cells. *Kidney Int* 1994;**46**:122–8
22. Ma R, Smith S, Child A, Carmines PK, Sansom SC. Store-operated Ca2+ channels in human glomerular mesangial cells. *Am J Physiol Renal Physiol* 2000;**278**:F954–61
23. Nutt LK, O'neil RG. Effect of elevated glucose on endothelin-induced store-operated and non-store-operated calcium influx in renal mesangial cells. *J Am Soc Nephrol* 2000;**11**:1225–35
24. Li W-P, Tsiokas L, Sansom SC, Ma R. Epidermal growth factor activates store-operated Ca2+ channels through an inositol 1, 4, 5-trisphosphate-independent pathway in human glomerular mesangial cells. *J Biol Chem* 2004;**279**:4570–7
25. Ma R, Kudlacek PE. Sansom SC Protein kinase Cα participates in activation of store-operated Ca^{2+} channels in human glomerular mesangial cells. *Am J Physiol Cell Physiol* 2002;**283**:C1390–98
26. Mene P, Pugliese F, Cinotti GA. Regulation of capacitative calcium influx in cultured human mesangial cells: roles of protein kinase C and calmodulin. *J Am Soc Nephrol* 1996;**7**:983–90
27. Kawasaki T, Ueyama T, Lange I, Feske S, Saito N. Protein kinase C-induced phosphorylation of Orai1 regulates the intracellular Ca^{2+} level via the store-operated Ca^{2+} channel. *J Biol Chem* 2010;**285**:25720–30
28. Wang X, Pluznick JL, Wei P, Padanilam BJ, Sansom SC. TRPC4 forms store-operated Ca2+ channels in mouse mesangial cells. *Am J Physiol Cell Physiol* 2004;**287**:C357–64
29. Sours S, Du J, Chu S, Ding M, Zhou XJ, Ma R. Expression of canonical transient receptor potential (TRPC) proteins in human glomerular mesangial cells. *Am J Physiol Renal Physiol* 2006;**290**:F1507–15
30. Goel M, Sinkins WG, Zuo C-D, Estacion M, Schilling WP. Identification and localization of TRPC channels in the rat kidney. *Am J Physiol Renal Physiol* 2006;**290**:F1241–52
31. Sours-Brothers S, Ding M, Graham S, Ma R. Interaction between TRPC1/TRPC4 assembly and STIM1 contributes to store-operated Ca^{2+} entry in mesangial cells. *Exp Biol Med* 2009;**234**:673–82
32. Cheng KT, Liu X, Ong HL, Ambudkar IS. Functional requirement for Orai1 in store-operated TRPC1-STIM1 channels. *J Biol Chem* 2008;**283**:12935–40
33. Kim MS, Zeng W, Yuan JP, Shin DM, Worley PF, Muallem S. Native store-operated Ca^{2+} influx requires the channel function of Orai1 and TRPC1. *J Biol Chem* 2009;**284**:9733–41
34. Wang X, Pluznick JL, Settles DC, Sansom SC. Association of VASP with TRPC4 in PKG-mediated inhibition of the store-operated calcium response in mesangial cells. *Am J Physiol Renal Physiol* 2007;**293**:F1768–76
35. Shen B, Zhu J, Zhang J, Jiang F, Wang Z, Zhang Y, Li J, Huang D, Ke D, Ma R. Attenuated mesangial cell proliferation related to store-operated Ca^{2+} entry in aged rat: the role of STIM 1 and orai 1. *Age* 2013;**35**:2193–202
36. Chaudhari S, Wu P, Wang Y, Ding Y, Yuan J, Begg M, Ma R. High glucose and diabetes enhanced store-operated Ca^{2+} entry and increased expression of its signaling proteins in mesangial cells. *Am J Physiol Renal Physiol* 2014;**306**:F1069–80

37. Wu P, Wang Y, Davis ME, Zuckerman JE Chaudhari S, Begg M, Ma R. Store–operated Ca^{2+} channels in mesangial cells inhibit matrix protein expression. *J Am Soc Nephrol* 2015;**26**:2691–702
38. Mai X, Shang J, Liang S, Yu B, Yuan J, Lin Y, Luo R, Zhang F, Liu Y, Lv X. Blockade of Orai1 store-operated calcium entry protects against renal fibrosis. *J Am Soc Nephrol* 2016;**27**:3063–78
39. Avila-Medina J, Mayoral-Gonzalez I, Dominguez-Rodriguez A, Gallardo-Castillo I, Ribas J, Ordoñez A, Rosado JA, Smani T. The complex role of store operated calcium entry pathways and related proteins in the function of cardiac, skeletal and vascular smooth muscle cells. *Front Physiol* 2018;**9**:257
40. Chen Y-F, Lin P-C, Yeh Y-M, Chen L-H, Shen M-R. Store-operated Ca^{2+} entry in tumor progression: from molecular mechanisms to clinical implications. *Cancers* 2019;**11**:899
41. Feske S, Picard C. Fischer Immunodeficiency due to mutations in ORAI1 and STIM1. *Clin Immunol* 2010;**135**:169–82
42. Tong BC-K, Lee CS-K, Cheng W-H, Lai K-O, Foskett JK, Cheung K-H. Familial Alzheimer's disease-associated presenilin 1 mutants promote γ-secretase cleavage of STIM1 to impair store-operated Ca^{2+} entry. *Sci Signal* 2016;**9**:ra89
43. Zhang H, Wu L, Pchitskaya E, Zakharova O, Saito T, Saido T, Bezprozvanny I. Neuronal store-operated calcium entry and mushroom spine loss in amyloid precursor protein knock-in mouse model of Alzheimer's disease. *J Neurosci* 2015;**35**:13275–86
44. Chernyuk D, Zernov N, Kabirova M, Bezprozvanny I, Popugaeva E. Antagonist of neuronal store-operated calcium entry exerts beneficial effects in neurons expressing PSEN1ΔE9 mutant linked to familial Alzheimer disease. *Neuroscience* 2019;**410**:118–27
45. Mene P, Pugliese G, Pricci F, Di Mario U, Cinotti G, Pugliese F. High glucose level inhibits capacitative Ca2+ influx in cultured rat mesangial cells by a protein kinase C-dependent mechanism. *Diabetologia* 1997;**40**:521–7
46. Jiang H, Zou S, Chaudhari S, Ma R. Short-term high-glucose treatment decreased abundance of Orai1 protein through posttranslational mechanisms in rat mesangial cells. *Am J Physiol Renal Physiol* 2018;**314**: F855–63
47. Chaudhari S, Li W, Wang Y, Jiang H, Ma Y, Davis ME, Zuckerman JE, Ma R. Store- operated calcium entry suppressed the TGF-β1/Smad3 signaling pathway in glomerular mesangial cells. *Am J Physiol Renal Physiol* 2017;**313**:F729–39
48. Wu P, Ren Y, Ma Y, Wang Y, Jiang H, Chaudhari S, Davis ME, Zuckerman JE, Ma R. Negative regulation of Smad1 pathway and collagen IV expression by store-operated Ca^{2+} entry in glomerular mesangial cells. *Am J Physiol Renal Physiol* 2017;**312**:F1090–100
49. Huang L, Ma R, Lin T, Chaudhari S, Shotorbani PY, Yang L, Wu P. Glucagon-like peptide-1 receptor pathway inhibits extracellular matrix production by mesangial cells through store-operated Ca^{2+} channel. *Exp Biol Med* 2019;**244**:1193–201
50. Huang L, Lin T, Shi M, Chen X, Wu P. Liraglutide suppresses production of extracellular matrix proteins and ameliorates renal injury of diabetic nephropathy by enhancing wnt/β-catenin signaling. *Am J Physiol Renal Physiol* 2020;**319**:F458–468
51. Mann JF, Ørsted DD, Brown-Frandsen K, Marso SP, Poulter NR, Rasmussen S, Tornøe K, Zinman B, Buse JB. Liraglutide and renal outcomes in type 2 diabetes. *N Engl J Med* 2017;**377**:839–48
52. Wang Y, Zhou CJ, Liu Y. Wnt signaling in kidney development and disease. *Prog Mol Biol Transl Sci* 2018;**153**:181–207
53. Hsu Y-C, Chang P-J, Ho C, Huang Y-T, Shih Y-H, Wang C-J, Lin C-L. Protective effects of miR-29a on diabetic glomerular dysfunction by modulation of DKK1/wnt/β-catenin signaling. *Sci Rep* 2016;**6**:1–12
54. Chaudhari S, Yazdizadeh Shotorbani P, Tao Y, Davis ME, Mallet RT, Ma R. Inhibition of interleukin-6 on matrix protein production by glomerular mesangial cells and the pathway involved. *Am J Physiol Renal Physiol* 2020;**318**:F1478–88
55. Ciuntu A. Role of metabolites of nitric oxide and arginase in the pathogenesis of glomerulonephritis. *Curr Health Sci J* 2016;**42**:221–5
56. Hohenstein B, Kasperek L, Kobelt D-J, Daniel C, Gambaryan S, Renné T, Walter U, Amann KU, Hugo CP. Vasodilator-stimulated phosphoprotein–deficient mice demonstrate increased platelet activation but improved renal endothelial preservation and regeneration in passive nephrotoxic nephritis. *J Am Soc Nephrol* 2005;**16**:986–96
57. He F, Peng F, Xia X, Zhao C, Luo Q, Guan W, Li Z, YX, Huang F. MiR-135a promotes renal fibrosis in diabetic nephropathy by regulating TRPC1. *Diabetologia* 2014;**57**:1726–36
58. Soni H, Adebiyi A. Urotensin II-induced store-operated Ca^{2+} entry contributes to glomerular mesangial cell proliferation and extracellular matrix protein production under high glucose conditions. *Sci Rep* 2017;**7**:1–14
59. Reiser J, Polu KR, Möller CC, Kenlan P, Altintas MM, Wei C, Faul C, Herbert S, Villegas I, Avila-Casado C. TRPC6 is a glomerular slit diaphragm-associated channel required for normal renal function. *Nat Genet* 2005;**37**:739–44
60. Yang H, Zhao B, Liao C, Zhang R, Meng K, Xu J, Jiao J. High glucose-induced apoptosis in cultured podocytes involves TRPC6-dependent calcium entry via the RhoA/ROCK pathway. *Biochem Biophys Res Commun* 2013;**434**:394–400
61. Dryer SE, Roshanravan H, Kim EY. TRPC channels: regulation, dysregulation and contributions to chronic kidney disease. *Biochim Biophys Acta Mol Basis Dis* 2019;**1865**:1041–66
62. Jin J, Ye M, Hu K, Gong J, He Q. STIM promotes the epithelial-mesenchymal transition of podocytes through regulation of FcγRII activity in diabetic nephropathy. *Histo Histopathol* 2018;**34**:671–82
63. Ying Q, Wu G. Molecular mechanisms involved in podocyte EMT and concomitant diabetic kidney diseases: an update. *Ren Fail* 2017;**39**:474–83
64. Duan W-J, Yu X, Huang X-R, Yu J-W, Lan HY. Opposing roles for Smad2 and Smad3 in peritoneal fibrosis in vivo and in vitro. *Am J Pathol* 2014;**184**:2275–84
65. Meng XM, Huang XR, Chung AC, Qin W, Shao X, Igarashi P, Ju W, Bottinger EP, Lan HY. Smad2 protects against TGF-β/Smad3-mediated renal fibrosis. *J Am Soc Nephrol* 2010;**21**:1477–87
66. Russo LM, Sandoval RM, Campos SB, Molitoris BA, Comper WD, Brown D. Impaired tubular uptake explains albuminuria in early diabetic nephropathy. *J Am Soc Nephrol* 2009;**20**:489–94
67. Zeng B, Chen G-L, Garcia-Vaz E, Bhandari S, Daskoulidou N, Berglund LM, Jiang H, Hallett T, Zhou L-P, Huang L. ORAI channels are critical for receptor-mediated endocytosis of albumin. *Nat Commun* 2017;**8**:1–12
68. Gombedza FC, Shin S, Kanaras YL, Bandyopadhyay BC. Abrogation of store-operated Ca^{2+}. Entry protects against crystal-induced ER stress in human proximal tubular cells. *Cell Death Discov* 2019;**5**:1–13
69. Woodward OM, Li Y, Yu S, Greenwell P, Wodarczyk C, Boletta A, Guggino WB, Qian F. Identification of a polycystin-1 cleavage product, P100, that regulates store operated Ca^{2+} entry through interactions with STIM1. *PLoS One* 2010;**5**:e12305
70. Yanda MK, Liu Q, Cebotaru V, Guggino WB, Cebotaru L. Role of calcium in adult onset polycystic kidney disease. *Cell Signal* 2019;**53**:140–50
71. Brill AL, Ehrlich BE. Polycystin 2: a calcium channel, channel partner, and regulator of calcium. Homeostasis in ADPKD. *Cell Signal* 2020;**66**:109490
72. Somlo S, Ehrlich B. Human disease: calcium signaling in polycystic kidney disease. *Curr Biol* 2001;**11**:R356–60
73. Tsiokas L, Arnould T, Zhu C, Kim E, Walz G, Sukhatme VP. Specific association of the gene product of PKD2 with the TRPC1 channel. *Proc Natl Acad Sci U S A* 1999;**96**:3934–9
74. Cowley Jr B. Calcium, cyclic AMP, and MAP kinases: dysregulation in polycystic kidney disease. *Kidney Int* 2008;**73**:251–3
75. Mamenko M, Dhande I, Tomilin V, Zaika O, Boukelmoune N, Zhu Y, Gonzalez-Garay ML, Pochynyuk O, Doris PA. Defective store-operated calcium entry causes partial nephrogenic diabetes insipidus. *J Am Soc Nephrol* 2016;**27**:2035–48
76. Cebotaru L, Cebotaru V, Wang H, Arend LJ, Guggino WB. STIM1fl/fl Ksp-Cre mouse has impaired renal water balance. *Cell Physiol Biochem* 2016;**39**:172–82
77. Fellner SK, Arendshorst WJ. Capacitative calcium entry in smooth muscle cells from preglomerular vessels. *Am J Physiol Renal Physiol* 1999;**277**:F533–42

78. Nagahama T, Hayashi K, Ozawa Y, Takenaka T, Saruta T. Role of protein kinase C in angiotensin II-induced constriction of renal microvessels. *Kidney Int* 2000;**57**:215–23
79. Carmines PK, Fowler Bc, Bell PD. Segmentally distinct effects of depolarization on intracellular [Ca^{2+}] in renal arterioles. *Am J Physiol Renal Physiol* 1993;**265**:F677–85
80. Loutzenhiser K, Loutzenhiser R. Angiotensin II–induced Ca^{2+} influx in renal afferent and efferent arterioles: differing roles of voltage-gated and store-operated Ca^{2+} entry. *Circ Res* 2000;**87**:551–7
81. Fellner SK, Arendshorst WJ. Store-operated Ca^{2+} entry is exaggerated in fresh preglomerular vascular smooth muscle cells of SHR. *Kidney Int* 2002;**61**:2132–41
82. Fallet RW, Ikenaga H, Bast JP, Carmines PK. Relative contributions of Ca^{2+} mobilization and influx in renal arteriolar contractile responses to arginine vasopressin. *Am J Physiol Renal Physiol* 2005;**288**:F545–51
83. Facemire CS, Arendshorst WJ. Calmodulin mediates norepinephrine-induced receptor-operated calcium entry in preglomerular resistance arteries. *Am J Physiol Renal Physiol* 2005;**289**:F127–36
84. Guan Z, Singletary ST, Cook AK, Hobbs JL, Pollock JS, Inscho EW. Sphingosine-1-phosphate evokes unique segment-specific vasoconstriction of the renal microvasculature. *J Am Soc Nephrol* 2014;**25**:1774–85
85. Guan Z, Wang F, Cui X, Inscho EW. Mechanisms of sphingosine-1-phosphate-mediated vasoconstriction of rat afferent arterioles. *Acta Physiol* 2018;**222**:1–23
86. Earley S, Brayden JE. Transient receptor potential channels in the vasculature. *Physiol Rev* 2015;**95**:645–90
87. Facemire CS, Mohler PJ, Arendshorst WJ. Expression and relative abundance of short transient receptor potential channels in the rat renal microcirculation. *Am J Physiol Renal Physiol* 2004;**286**:F546–51
88. Lee-Kwon W, Wade JB, Zhang Z, Pallone TL, Weinman EJ. Expression of TRPC4 channel protein that interacts with NHERF-2 in rat descending vasa recta. *Am J Physiol Cell Physiol* 2005;**288**:C942–C49
89. Shi J, Miralles F, Birnbaumer L, Large WA, Albert AP. Store-operated interactions between plasmalemmal STIM1 and TRPC1 proteins stimulate PLCβ1 to induce TRPC1 channel activation in vascular smooth muscle cells. *J Physiol* 2017;**595**:1039–58
90. Gwack Y, Feske S, Srikanth S, Hogan PG, Rao A. Signalling to transcription: store-operated. Ca^{2+} entry and NFAT activation in lymphocytes. *Cell Calcium* 2007;**42**:145–56
91. Shaw PJ, Feske S. Physiological and pathophysiological functions of SOCE in the immune system. *Front Biosci* 2012;**4**:2253–68
92. Dellepiane S, Leventhal JS, Cravedi PT. Cells and acute kidney injury: a two-way relationship. *Front Immunol* 2020;**11**:1546
93. Mehrotra P, Sturek M, Neyra JA, Basile DP. Calcium channel Orai1 promotes lymphocyte IL-17 expression and progressive kidney injury. *J Clin Invest* 2019;**129**:4951–61
94. Dhande IS, Zhu Y, Kneedler SC, Joshi AS, Hicks MJ, Wenderfer SE, Braun MC, Doris PA. Stim1 polymorphism disrupts immune signaling and creates renal injury in hypertension. *J Am Heart Assoc* 2020;**9**:e014142