Spontaneous electrical and Ca^{2+} signals in the mouse renal pelvis that drive pyeloureteric peristalsis

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Summary

1. Peristalsis in the smooth muscle cell (SMC) wall of the pyeloureteric system is unique in physiology in that the primary pacemaker resides in a population of atypical SMCs situated near the border of the renal papilla.

2. Atypical SMCs display high frequency Ca^{2+} transients upon the spontaneous release of Ca^{2+} from IP_3-dependent stores that trigger cation-selective spontaneous transient depolarizations (STDs). In nifedipine, these Ca^{2+} transients and STDS seldom propagate >100 μm. Synchronization of STDs in neighbouring atypical SMCs into an electrical signal that can trigger action potential discharge and contraction in the typical SMC layer involves a coupled oscillator mechanism dependent on Ca^{2+} entry through L-type voltage-operated Ca^{2+} channels.

3. A population of spindle- or stellate-shaped cells, immuno-positive for the tyrosine receptor kinase kit, are sparsely distributed throughout the pyeloureteric system. Ca^{2+} transients and action potentials of long duration occurring at low frequencies are also recorded in a population of fusiform cells, which we have termed interstitial cells of Cajal-like cells (ICC-like cells).

4. The electrical and Ca^{2+} signals in ICC-like cells are abolished upon blockade of Ca^{2+} release from either IP_3- or ryanodine-dependent Ca^{2+} stores. However, the spontaneous Ca^{2+} signals in atypical SMCs or ICC-like cells are little affected in W/W^{-v} transgenic mice which have extensive lesions of their intestinal ICC networks.

5. In summary, we have developed a model of pyeloureteric pacemaking in which atypical SMCs are indeed the primary pacemakers, the function of ICC-like cells has yet to be determined.

Pyeloureteric peristalsis

Since the earliest descriptions by Englemann (1869)[1] of pyeloureteric peristalsis, it has been recognised that the spontaneous propagating contractions which transport urine expressed in the kidney along the ureter to the bladder originate within the most proximal regions of the renal pelvis. In most mammals, including the mouse, the kidney contains a single papilla which is surrounded by a funnel-shaped calyx or renal pelvis (Figure 1A). This renal pelvis consists of a urothelium-lined lumen and a plexus of long, spindle-shaped 'typical' smooth muscle cells (SMCs) which form an inner layer, originating near the base of the papilla and extending through into the ureter.[2,3] The density of typical SMCs increases with distance from the papilla base resulting in a gradual thickening of the pelvic wall.[3-5] In human and pig, the kidney is multi-papillate so that a number of major and minor calyces fuse to form a separate renal pelvis which extends to the ureter.[2-4,6] Even though there is evidence of extensive networks of parasympathetic and sympathetic nerves within the wall of the pelviureteric system,[7] it has been difficult to demonstrate that these networks play any efferent role in maintaining or modulating pyeloureteric peristalsis. Electrical nerve stimulation evokes in the guinea pig renal pelvis a transient increase in the amplitude and frequency of the spontaneous contractions which is followed by a prolonged negative inotropic effect that are little affected by N^G-nitro-L-arginine, guanethidine or atropine,[1,8,9] but have been attributed to the release of tachykinins and calcitonin gene related peptide (CGRP), respectively, from sensory nerves.[9,10]

The recent use of intracellular microelectrode recording techniques (Figure 1Bi) and fluorescence indicators of Ca^{2+} concentration (Figure 2A) have unequivocally established that nifedipine-sensitive action potentials and Ca^{2+} waves within the smooth muscle wall are responsible for the peristaltic contractions that propagate the length of the upper urinary system.[5,11-15] The spatial temporal map in the lower panel of Figure 2A also illustrates that rises of Ca^{2+} occur instantaneously ('flashes') along the entire length of each typical SMC *in situ* and that propagation of the Ca^{2+} wave occurs in the direction perpendicular to the long axis of individual typical SMCs (Figure 2A upper panel).

Pacemaking in the pyleoureteric system

In both uni- and multi-calyceal kidneys, a single pacemaker region at any one time on the papilla-calyceal border initiates the wave of contraction, which conducts radially across the pelvis to form a crescent shaped wave that then conducts distally. In uni-papillate kidneys, the pacemaker region shifts spontaneously along the pelvi-calyceal border; in multi-papillate kidneys this 'primary' pacemaker shifts spontaneously between calyces.[16,17]

Atypical SMCs, which are shorter in their long axis than typical SMCs, form a relatively sparse outer layer in uni-calyceal kidneys and an inner layer in multi-calyceal kidneys that wraps around the most proximal regions of the papilla and terminates at the pelviureteric junction.[2,3,5,6,18]

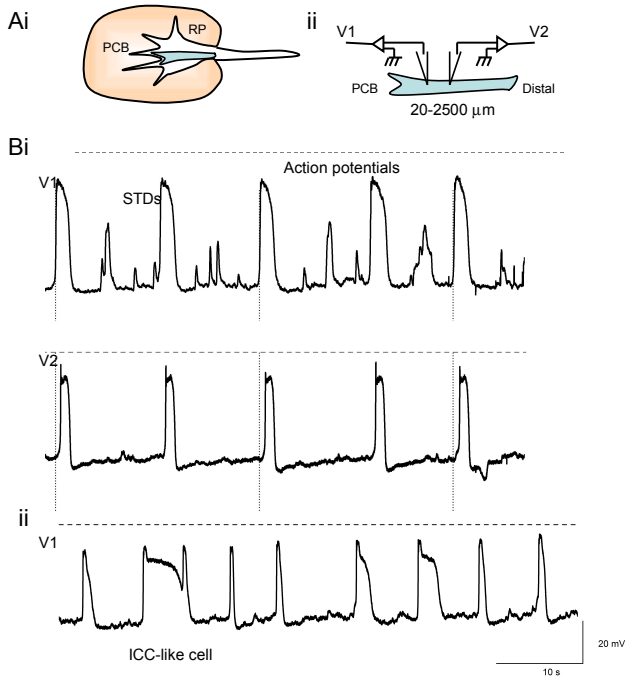

Figure 1. Electrical recordings in the mouse renal pelvis. A. *Schematic representation of the structure of the mouse pyeloureteric system (**Ai**) and the electrophysiological arrangement (**Aii**) when recording from longitudinal strips using 2 intracellular microelectrodes (V_1 and V_2). **Bi.** Typical intracellular microelectrode recordings from the mouse renal pelvis when 2 microelectrodes (V_1 and V_2) are placed 1 mm apart. Propagating action potentials and non-propagating spontaneous transient depolarizations (STDs) are both evident. **Bii.** Long duration action potentials which we believe originate in ICC-like cells are recorded in another preparation. PCB papilla-calyceal border; RP renal pelvis. Dashed lines represent 0 mV.*

These atypical SMCs display many of the morphological features of cardiac sino-atrial pacemaker cells having a small nucleus and many long branching processes that contain relatively few contractile filaments and displaying a light immuno-staining for smooth muscle α-actin.[2,4,5]

Intracellular microelectrode recordings, particularly in the most proximal regions of the renal pelvis of rat,[18] mouse[19] and guinea pig,[5,20] reveal that short spindle-shaped atypical SMCs display high frequency (10-40 min^{-1}) spontaneous transient depolarizations (STDs)(Figure 1Bi) of a simple waveform that are reduced but not blocked by nifedipine. Using two intracellular microelectrodes we have recently established that STDs in the presence of nifedipine do not propagate over distances >100 μm in either the axial or circumferential direction. These STDs are recorded less often in the distal renal pelvis (Figure 1Bi) and never in the ureter.[5] STDs are little affected by the Cl^- channel blocker, DIDS, or upon the removal of 93% of extracellular Cl^- concentration using isethionate as the replacement ion. Instead, STDs are reduced when the extracellular Na^+ is mostly replaced with N-methyl-D-glucamine, indicating that these spontaneous events are arising from the opening of cationic selective channels.[19] However, blockers (La^{3+}, Gd^{3+}) of cationic pacemaker currents in cultured intestinal ICC[21,22] have little effect on STD discharge in the renal pelvis suggesting that they are arising from the opening of different cationic channels whose properties have yet to be established.[19]

When preparations of mouse renal pelvis are loaded with Ca^{2+} fluorophore, Fluo-4 AM, high frequency, transient rises in intracellular Ca^{2+} ($[Ca^{2+}]_i$) are readily recorded in a population of short spindle-shaped atypical SMCs, particularly in the presence of nifedipine (Figure 2B).[15] These atypical SMCs form small randomly-orientated bundles that were not in the same plane of focus as the typical SMC layer, nor uniformly distributed throughout the proximal regions of the preparation. Even though distinct Ca^{2+} waves of relatively slow velocity are observed in individual atypical SMCs in nifedipine (Figure 2B), these waves are only occasionally observed to propagate into neighbouring cells (Figure 2Ba,b). Moreover Ca^{2+} transients in neighbouring atypical SMCs show little correlation in time when they are separated by distances >30 μm.[15] Thus we have proposed that this inability of both Ca^{2+} waves and STDs to propagate over long distances in nifedipine indicates that both phenomena are occurring in the same cell population, atypical SMCs.

Mechanism of STD discharge

All STD activity and Ca^{2+} transients recorded in atypical SMCs bathed in nifedipine are blocked upon removal of Ca^{2+} from the bathing solution or upon preventing Ca^{2+} store uptake by blocking the sarco-endoplasmic reticulum Ca^{2+} ATPase (SERCA) with cyclopiazonic acid (CPA).[19,23] Both treatments cause membrane depolarization of some 10-20 mV. However, Ca^{2+}-free solution reduces intracellular $[Ca^{2+}]_i$ while CPA transiently raises $[Ca^{2+}]_i$.[19] The membrane depolarization evoked by the fall of $[Ca^{2+}]_i$ in Ca^{2+} free solution presumably arises from a reduction of a Ca^{2+}-activated membrane conductance for K^+, or from a cationic conductance activated upon lowering the $[Ca^{2+}]_i$ as suggested in intestinal ICC.[22] However the lack of effect of DIDS, La^{3+} or Gd^{3+} on the membrane potential or STD discharge in the renal pelvis suggests that such cationic channels are not involved. The most likely K^+ channels closed during a fall of $[Ca^{2+}]_i$ would be large conductance Ca^{2+}-activated K^+ (BK_{Ca}) channels expressed in typical SMCs but not ICC-like cells of the mouse ureteropelvic junction.[24] This notion is perhaps confirmed by our recent observations that renal pelvis strips exposed to the selective BK_{Ca} channel blocker iberiotoxin (100 nM for 10 min N=4) display a membrane depolarization of 5-10 mV which did not block STD discharge (MA Tonta & RJ Lang unpublished observations). Blockade of SERCA would be expected to result in a rise of $[Ca^{2+}]_i$ suggesting that the membrane depolarization in CPA may be arising from the induction of a cation- or Cl^--selective conductance that is

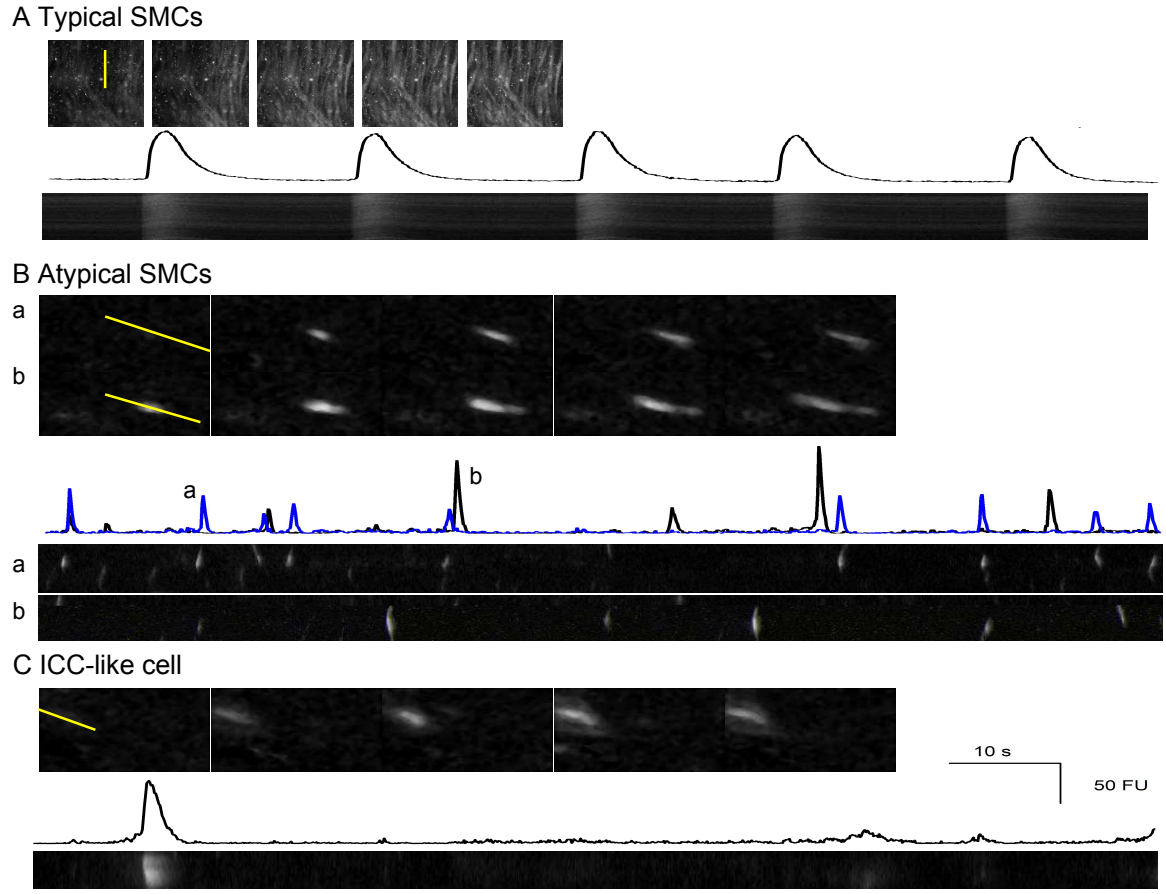

Figure 2 Spontaneous Ca^{2+} transients in the renal pelvis. A upper panel*: Sequential Ca^{2+} fluorescence intensity micrographs of a fluo-4 loaded TSMC layer displayed at time intervals of 400 ms at ×20 magnification. A Ca^{2+} wave is seen as a transient rise in Ca^{2+} intensity (**middle panel**) which propagates across the field of view perpendicular to the long axis of each typical SMC (modified from Lang* et al. *2007[15]). **Lower panel** A spatial temporal map of the cell indicated by the bar in the upper left panel illustrates that the Ca^{2+} rise occurred synchronously along the entire length of the cell (Ca^{2+} flash). **B upper panel**: Sequential Ca^{2+} fluorescence intensity micrographs of 2 atypical SMCs (a and b) displayed at time intervals of 100 ms at ×60 magnification, tissue bathed in 1 μM nifedipine. **Middle panel**: Fluorescence intensity for cells a and b have been plotted against time for comparison. **Lower panels**: Spatial temporal maps of cells a and b illustrate that slow velocity Ca^{2+} waves travel down each atypical SMC and that the site of initiation and direction of these Ca^{2+} waves in the each cell can change randomly. **C upper panel**: Sequential Ca^{2+} fluorescence intensity micrographs of an ICC-like cell in the same preparation as B displayed at time intervals of 500 ms at ×60 magnification. **Middle panel**: Fluorescence intensity for this ICC-like cell has been plotted against time for comparison. **Lower panel**: Spatial temporal map of this ICC-like cell illustrates that the Ca^{2+} rise occurred near synchronously along the cell body. However Ca^{2+} transients in projections of these ICC-like cells could not be detected.*

Ca^{2+} activated. It is interesting that rat, but not guinea pig, ureteric myocytes express Ca^{2+}-activated Cl^- channels[25] which, if present in mouse ureteropelvic typical SMCs could also contribute to this membrane depolarization in CPA.

Role of Ca^{2+} stores

Urogenital ICC-like cells and many smooth muscles display transient rises in $[Ca^{2+}]_i$ which are achieved upon the release of Ca^{2+} from internal stores *via* Ca^{2+} release channels coupled to ryanodine or inositol trisphosphate (IP_3) receptors. Both receptor populations are sensitive to cytoplasmic Ca^{2+} so that Ca^{2+} entry and (or) Ca^{2+} release *via* one receptor population can stimulate the Ca^{2+} release from the other receptor population *via* Ca^{2+}-induced Ca^{2+} release (CICR) mechanisms (Figure 3).[26] In many smooth muscles except the ureter, the pool of Ca^{2+} within ryanodine- and IP_3-sensitive stores appears to be functionally coupled *via* the co-localization of their receptors within the sarco-endoplasmic reticulum membrane.[27,28] A species dependence on the expression of each functioning Ca^{2+} store is evident in the SMCs of the ureter, such that only the ryanodine sensitive store appears to be functional in the guinea pig,[29] while only the IP_3

receptor-sensitive Ca^{2+} store can be detected in the rat.[29,30]

Blockers of, phospholipase C (PLC) function and IP_3 formation (neomycin, U73122, xestospongin C) or IP_3 receptors (2-APB and heparin) have all been shown to block or reduce electrical oscillations in urogenital[23,31] and gastrointestinal[32-35] preparations. However, the most compelling evidence establishing the essential role of IP_3-sensitive Ca^{2+} release channels has come from the absence of slow waves in mice which lack IP_3 type 1 receptors in their smooth muscle.[36] In our experiments, STDs and Ca^{2+} transient discharge in atypical SMCs are blocked by 2-APB and reduced by blockers of PLC function, both in a manner associated with membrane depolarization of some 5-10 mV.[19]

2-APB has been suggested to both deplete Ca^{2+} stores by blocking store-operated channels without affecting IP_3 dependent Ca^{2+} release[37] and block SERCA in a manner similar to CPA.[38] However, the disconnect between membrane potential and Ca^{2+} levels seen with 2-APB, when compared with CPA, and the lack of effect of TRP channels blockers in our hands suggest that 2-APB is indeed blocking IP_3 receptors. These observations are consistent with our previous notion that the intrinsic release of prostaglandins and tachykinins provides a constant excitatory PLC drive of IP_3 production in our pacemaker cells, which is essential in maintaining pelviureteric peristalsis.[39,40]

Blockade of ryanodine receptors, using concentrations (<1 mM) thought to block ryanodine-sensitive Ca^{2+} release channels without causing store depletion,[41] abolishes spontaneous electrical and Ca^{2+} signalling in ICC-like cells in the rabbit urethra[42] and guinea-pig corporal tissue,[43] but not the spontaneous electrical activity in portions of the mouse small intestine[44] or guinea renal pelvis.[23] In mouse renal pelvis, the amplitude and ½ width of STD recorded *in situ* are significantly reduced to 582% and 746%, respectively, after 20 min exposure to 100 μM ryanodine. However the Ca^{2+} transients recorded in individual atypical SMCs *in situ* are not significantly affected, even after 30-60 min exposure.[19]

Model of pyeloureteric pacemaking

We have interpreted our data to suggest that STDs are likely to represent the summation of the electrical activity generated by more than one atypical SMC. We envisage that, in the presence of nifedipine, the spontaneous release of Ca^{2+} from IP_3-dependent Ca^{2+} stores in individual atypical SMCs underlies the generation of the Ca^{2+} transients and 'unitary' STDs, while the subsequent activation of neighbouring Ca^{2+} release channels and CIRC mechanisms represent the fundamental mechanism underlying the slow velocity Ca^{2+} waves (Figure 3).[26]

In the absence of nifedipine, STD discharge in an individual atypical SMC induces the entry of Ca^{2+} through 'L-type' voltage-operated Ca^{2+} channels (VOCCs) which leads to the accelerated activation (entrainment) of neighbouring IP_3-primed Ca^{2+} stores both within the same cell and in neighbouring atypical SMCs (Figure 3). This depolarization-dependent entrainment couples neighbouring atypical SMCs within a pacemaker bundle to fire (release Ca^{2+} and generate STDs) synchronously *via* a coupled oscillator mechanism (as described in the stomach by van Helden & Imtiaz[45]) to create a depolarizing signal sufficiently rapid and large enough to propagate into the typical SMC layer and trigger action potential discharge and contraction (Figure 3). The atypical SMC bundle firing STDs at the highest frequency would presumably drive neighbouring atypical and typical smooth muscle bundles so that only one pacemaker region would be apparent at any one time. However, as each atypical SMC is capable of either initiating or contributing to an entrainment signal generated by its neighbours, the site of the dominant pacemaker region can spontaneously move within the most proximal region of the uni-calyceal renal pelvis, even between the minor calyces in multi-calyceal kidneys.[26,45]

Action potential discharge in the SMC wall of the pyeloureteric system is characterized by a prolonged refractory period of many seconds so that contraction frequency of the muscle wall is generally ¼ to ½ of the frequency of atypical SMC STD discharge (10-40 min^{-1}).[5,11,46] Moreover, the frequency of contraction of circumferentially-cut muscle strips decreases with distance from the base of papilla[12,47,48] which we have previously correlated with an increasing ratio of typical SMCs, compared to atypical SMCs.[5] This difference in frequencies between atypical and typical SMCs has recently been explained in terms of the refractory mechanisms within typical SMCs that are dependent on the state of the internal Ca stores.[11,26] It has been suggested that Ca^{2+} entry through VOCCs during typical SMC action potential discharge and contraction is sequestered into the endoplasmic reticulum, which, once loaded, sensitizes its ryanodine receptors to generate Ca^{2+} sparks. This release of stored Ca^{2+}, in turn, activates plasmalemmal BK_{Ca} channels which hyperpolarize the membrane of the typical SMCs creating the refractory periods observed. Ca^{2+} release declines as the Ca^{2+} levels in the store returns to normal which results in a decreased BK_{Ca} channel activity and a repolarization of the membrane into a voltage range that is responsive to the STD drive coming from neighbouring atypical SMCs (Figure 3).[26]

The role of the mitochondria in the generation of the atypical SMC STDs or in the control of smooth muscle contraction remains unclear. Whether the mitochondria play a passive role, temporarily and rapidly storing Ca^{2+} before slowly releasing it so that it can be taken up into the endoplasmic reticulum by SERCA,[26] or whether it has a more active involvement, lowering Ca^{2+} concentrations in micro-domains near the plasmalemma membrane so that the cationic conductance underlying STD generation is actually activated by a fall[33,49] in Ca^{2+} has yet to be established. However, preliminary experiments have demonstrated that disruption of the mitochondrial function with CCCP (1 μM) abolishes Ca^{2+} transients in the typical SMC layer in the absence of nifedipine (H. Hashitani unpublished observations). The role of the mitochondria in atypical and typical SMC function is under intensive

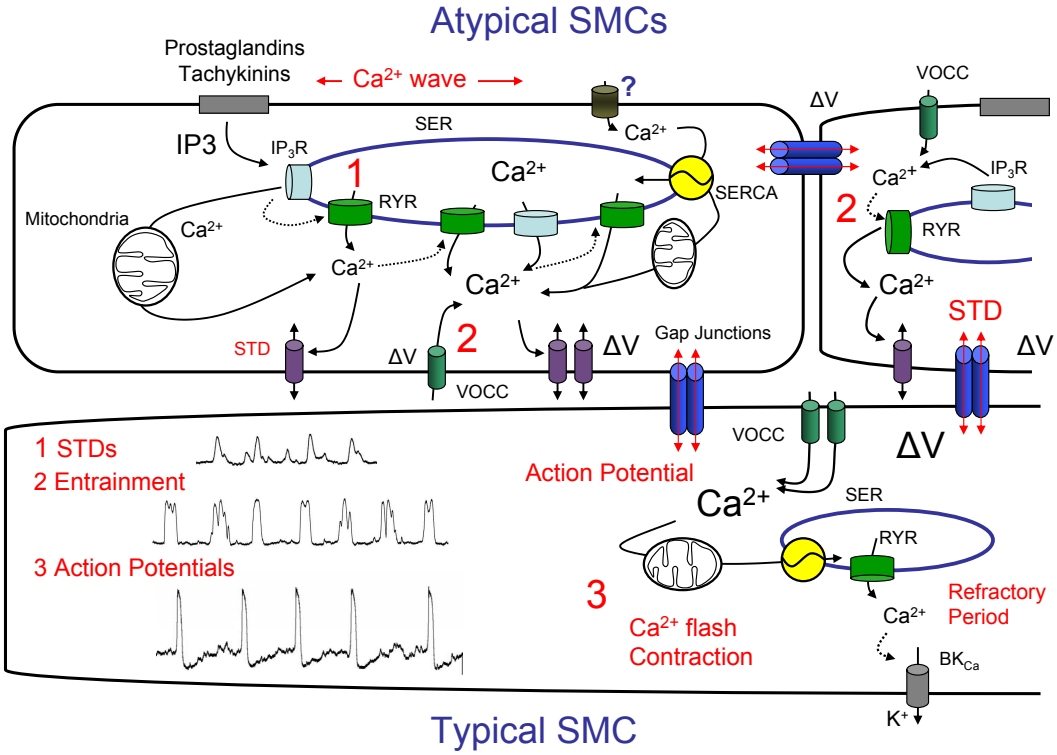

Figure 3 Model of atypical SMC pacemaking. *Under a constant IP_3 drive arising from the intrinsic release of prostaglandins and sensory nerve tachykinins, the spontaneous release of Ca^{2+} from IP_3-dependent Ca^{2+} stores in individual atypical SMCs activates the firing of 'unitary' cation-selective STDs. In the presence of nifedipine, the sequential activation of neighbouring Ca^{2+} release channels* via *CICR mechanisms gives rises to the slow velocity intercellular Ca^{2+} waves observed in atypical SMCs (1). In the absence of nifedipine, STD evoked membrane depolarization triggers the influx of Ca^{2+} through L-type Ca^{2+} channels which accelerates the entrainment of neighbouring Ca^{2+} release channels in the same cell, as well as in neighbouring atypical SMCs (2) as current flows through gap junctions. Synchronicity of atypical SMCs occurs* via *this depolarization-dependent coupled oscillator mechanism[26,45] until the amplitude of the current flow underlying the STDs is large enough to discharge the membrane of neighbouring typical SMCs to cause membrane depolarization, action potential discharge and contraction (3). SER sarco-endoplasmic reticulum; RyR ryanodine receptor; VOCC voltage operated Ca^{2+} channels; IP_3R IP3 receptor; ΔV membrane depolarization.*

investigation.

Interstitial cells of Cajal (ICC)-like cells

The essential role of ICC in the generation of autorhythmicity in many smooth muscles has been established from the use of W/W^{-v} mice which have a mutation of the dominant *white spotting* (*W*) locus (kit receptor) that results in a marked reduction of kit tyrosine kinase-dependent signalling, essential for the development and survival of mast cells, haematopoiesis and the cell division and network formation of ICC. W/W^{-v} mice have a profound loss of ICC number located near the myenteric border in many regions of the gastrointestinal tract which results in a blockade of rhythmic muscle activity, as well as a loss of intramuscular ICC which selectively blocks nitrergic and cholinergic neurotransmission.[50,51]

In uni-calyceal and multi-calyceal mammals, including mouse,[15,52,53] rat[54] and human,[55-57] sparse networks of spindle- and stellate-shaped cells that are immuno-positive for antibodies raised against kit have been described within the lamina propria and the muscle layer of the renal pelvis and proximal ureter. Indeed, ICC-like cells, immuno-reactive to kit, appear in the mouse embryonic ureter in culture at the same time as coordinated unidirectional peristaltic contractions in a manner blocked by the kit antibody, ACK45.[58] In 1999,[5] we reported a population of electrically-active, but kit-negative, ICC-like cells in guinea pig renal pelvis that, when viewed with an electron microscope, had many of the 'gold standard' morphological features used to distinguish intestinal ICC from fibroblasts. These ICC-like cells formed close appositions with themselves and with both atypical and typical SMCs, suggesting electrical connectivity and therefore possibly having an influence on pyeloureteric peristalsis.

In the mouse renal pelvis, fusiform interstitial cells with a distribution similar to the stellate-shaped kit-positive cells fire nifedipine-insensitive Ca^{2+} signals with frequencies and temporal characteristics similar to the nifedipine-insensitive long-plateau action potentials

recorded with intracellular microelectrodes (Figure 1Bii).[15] However, we have not been able to demonstrate that the interstitial cells pre-labelled with a kit antibody that binds at an extracellular site(s) are in fact the interstitial cells displaying low frequency Ca^{2+} signals. Moreover, we cannot find any noticeable difference between the renal pelvis obtained from wild type and W/W^{-v} mice, particularly in terms of their Ca^{2+} signalling, contractility or responses to electrical nerve stimulation. Finally, the kidneys of W/W^{-v} mice did not display any changes in their shape or size, consistent with the lack of any hydronephrosis in this phenotype (RJ Lang & H Hasihtani unpublished observations).

Autorhythmicity in ICC-like cells

The major limitation of recording the membrane potential of the renal pelvis using intracellular microelectrodes is that ICC-like cells and atypical and typical SMCs are likely to be in electrical continuity and, therefore, it is difficult to ascertain the cellular origin of our electrical signals. The infrequency of impaling an ICC-like cell with an intracellular microelectrode with confidence, rather than a cell electrically close to an ICC-like cell, has prevented the examination of the effects of our modifiers of Ca^{2+} mobilization on their electrical behaviour. However it has been possible to examine the effects of these agents on the low frequency Ca^{2+} transients in fusiform interstitial cells at the same time as our examination of the Ca^{2+} transients in atypical SMCs.[15] In Figure 2C, it is evident that, in contrast to atypical SMCs, the Ca^{2+} signal in ICC-like cells appears as a long-lasting signal along the entire length of the cell body. Interestingly, even though this Ca^{2+} transient in nifedipine is relatively larger and longer than the Ca^{2+} wave in atypical SMCs, it fails to propagate into any neighbouring cells, be they other ICC-like cells or atypical and typical SMCs. In addition, even though some electrical coupling between the cells firing the long lasting action potentials and the typical SMC layer is often evident, we have not yet seen an individual interstitial cell trigger a typical SMC Ca^{2+} transient or muscle contraction.[15]

The agents described above that interrupted store uptake and IP_3-dependent release of Ca^{2+} in atypical SMCs were equally effective on Ca^{2+} signalling in ICC-like cells in the mouse renal pelvis. However, there was a clear difference in the action of ryanodine in atypical SMCs and ICC-like cells within the same preparation. In comparison to the relatively small effect on Ca^{2+} transients in atypical SMCs in nifedipine, ryanodine completely blocked Ca^{2+} transient discharge in ICC-like cells within several minutes exposure.[19]

We have previously suggested that ICC-like cells in guinea pig renal pelvis could well be acting as integrators of the atypical SMC pacemaker drive.[5,8] In our present experiments, we have been unable to demonstrate any synchronicity between neighbouring ICC-like cells, or between any atypical SMCs and ICC-like cells in preparations bathed in 1 μM nifedipine.[15] This situation may well be quite different in the absence of nifedipine when Ca^{2+} influx through VOCCs could again be the agent of entrainment in ICC-like cells such that Ca^{2+} transients generated in the cell body or neighbouring cells rapidly travel down the interstitial cell projections *via* CICR from Ca^{2+} release channels as described above.[26]

Our measurements of $[Ca^{2+}]_i$ in cells of the renal pelvis are at a relatively low resolution so that we have not been able to detect Ca^{2+} transients in the stellate projections of our ICC-like cells. However, we envisage that the long processes of ICC-like cells could serve an integrative role to rapidly distribute a pacemaker signal over a relatively wide area of the renal pelvis. This may well be critical in the proximal regions of the renal pelvis where the distribution of both atypical and typical SMCs is relatively sparse and not as well organized as in the more distal regions. Even though the intrinsically low frequency of Ca^{2+} transient discharge in ICC-like cells bathed in nifedipine makes it is unlikely that these cells are acting as a primary pacemaker, it is tempting to suggest that ICC-like cells could provide a secondary drive which can take over pacemaking during conditions that dislocate the ureter from the proximal pacemaker drive, e.g. during ureteric obstruction[56] or after kidney transplantation.

Conclusions

Except for regions distal of the ureteropelvic junction it is likely that atypical SMCs and ICC-like cells both influence the spontaneous electrical and contractile activity of the upper urinary tract. We believe that atypical SMCs are indeed the primary pacemakers for pyeloureteric peristalsis, the function of ICC-like cells has yet to be determined. We have demonstrated that the mechanisms of Ca^{2+} transient generation in these 2 cell populations can be pharmacologically distinguished on the basis of their (in)sensitivity to ryanodine. Elucidation of the fundamental mechanisms by which ICC-like cells and atypical SMCs control muscle wall contractility will lead to the development of functional/ electrical models that combine the autorhythmicity of atypical SMCs and ICC-like cells with the ultrastructure and electrical coupling of all of the cells involved under normal physiological and pathological conditions. To date, non-surgical pharmaceutical treatments to alleviate the consequences of obstruction-induced remodelling in the upper urinary system have not been developed due to a lack of a basic understanding of the fundamental physiology underlying the initiation and propagation of pyeloureteric contractions. Thus pyeloureteric ICC-like cells and atypical SMCs with their unique distribution and autorhythmicity may well provide selective pharmacological targets when considering non-surgical interventions to alleviate hydronephrosis arising from pelviureteric remodelling during and after ureteric blockade or pyeloplasty.

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