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Effects of nerve stimulation on the spontaneous action potentials recorded in the proximal renal pelvis of the guinea-pig

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Abstract The effects of nerve stimulation on the electrical and mechanical activity of the smooth muscle of the proximal renal pelvis of the guinea-pig were investigated using standard tension and microelectrode recording techniques. Spontaneous action potentials were deemed to have been recorded from three cell types: (1) "pacemaker" cells (9 of >120) had membrane potentials (MPs) of -42.1 ± 2.9 mV and fired action potentials of a simple waveform; (2) "driven" cells (>100) had more stable MPs of -56.1 ± 1.2 mV ($n = 36$) and more complex "ureter-like" action potentials; (3) the remaining cells had MPs of -45.5 ± 1.7 mV ($n = 15$) and action potentials with a waveform "intermediate" to groups (1) and (2). Nifedipine (0.1–1 μM) and Cd^{2+} (0.1–1 mM) blocked all spontaneous action potential discharge and depolarized the membrane to near -40 mV. Intramural nerve stimulation (10–50 Hz for 1–10 s) increased both the amplitude and frequency of the spontaneous contractile activity, this increase peaked in about 30 s and decayed slowly over several minutes. Nerve stimulation depolarized pacemaker and driven cells 9.1 ± 3.5 ($n = 3$) and 1.6 ± 0.7 ($n = 6$) mV, respectively; the frequency of their action potential discharge increased from 7.6 ± 2.7 and 9.9 ± 1.1/min to 17.3 ± 0.5 and 11.1 ± 1.4/min, respectively. The duration of the action potentials in driven cells also increased significantly for several minutes. All these effects were blocked by tetrodotoxin (TTX) (1.6 μM). It was concluded that the positive chronotropic and inotropic effects of nerve stimulation on renal pelvis contractility can be correlated with the changes in the frequency and duration of the action potentials recorded in driven cells.

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The renal pelvis has long been suggested to be the pacemaker for ureteric peristalsis [1]. This pacemaker activity has been thought to originate either from the proximal portions of the pelvicalyceal region [3] or from the pelviureteral junction [22], depending on the animal species and on whether they have multi- or unicalyceal kidneys. Generally, however, there is a gradient in frequency of the spontaneous rhythmic contractions of circumferentially cut strips of the renal pelvis, reducing as strips were taken from regions more distal to the renal fornix [3, 9, 19]. The ureter in most mammals is quiescent unless a portion of the pelviureteral junction is attached or if it is stimulated by excitatory agents such as α-adrenoceptor agonists [8]. These slower or latent pacemaker regions in the distal pelvis and ureter are usually dominated by the proximal pacemaker regions, but can drive peristalsis, and sometimes antiperistaltic contractions, after the primary pacemakers have been destroyed, e.g. after partial nephrectomy [18].

Anatomically, the renal pelvis in man and many mammals consists of two morphologically and histochemically distinct types of smooth muscle cells. "Typical" smooth muscle cells, similar to those found in the ureter, are spindle shaped and grouped into bundles which lie in various directions, branch and form connections with adjacent bundles to form a single plexiform of interconnecting muscle bundles. "Atypical" smooth muscle cells form a thin subepithelial layer which extends from the region of pelvic attachment to the renal parenchyma to the pelviureteric junction. These cells are characterized by their distinctive

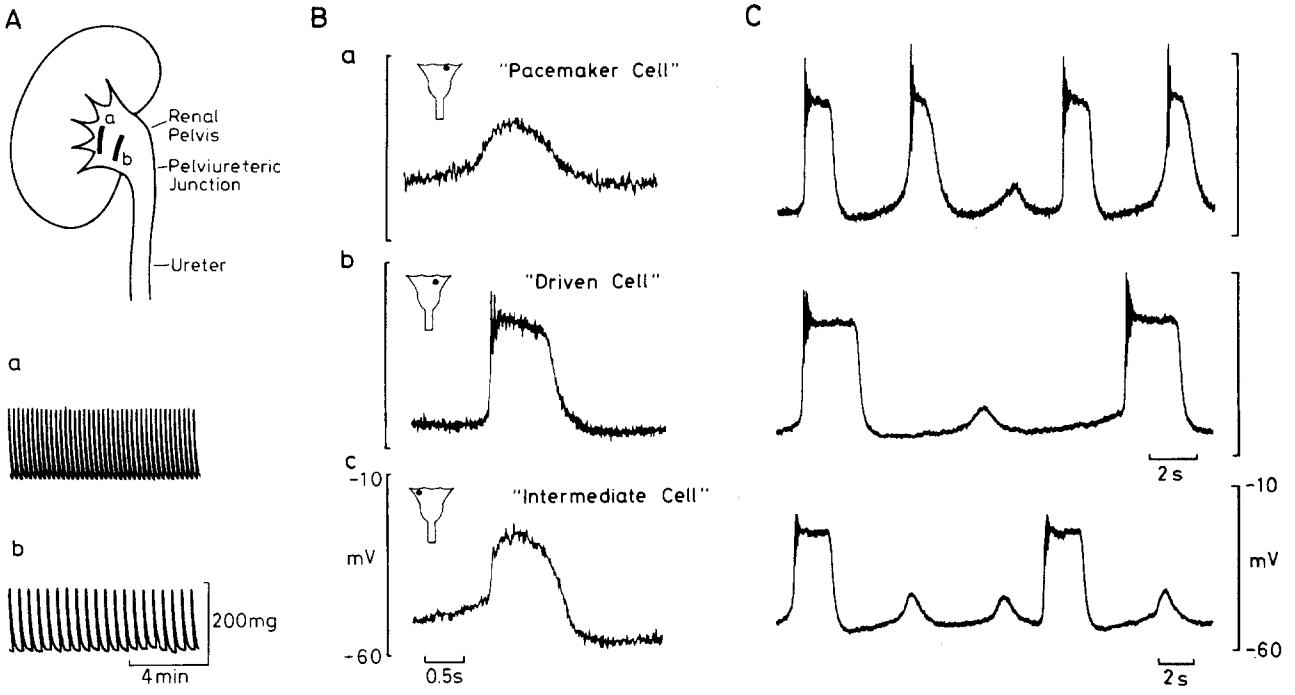

Fig. 1A–C Circumferentially orientated strips cut from the proximal (**A***a*) and mid (**A***b*) regions of the guinea-pig renal pelvis developed spontaneous contractions after 30–60 min incubation. The frequency of contraction of the proximal renal pelvis ($4.8 \pm 0.3\ min^{-1}$) was significantly greater than that of the mid renal pelvis (1.8 ± 0.2/min; $P < 0.5$, $n = 10$). **B** The spontaneous action potentials recorded in the proximal renal pelvis were divided into three groups, designated as being recorded from "pacemaker" (**B***a*), "driven" (**B***b*) and "intermediate" (**B***c*) cells. *Inserts* indicate the approximate position of each recording. **C** Three examples of driven cells with varying refractory periods, during which subsequent action potentials were either of a reduced duration or failed to fire

irregular morphology and histology and by the absence of non-specific cholinesterase staining [4]. In multicalyceal mammals (man, pig), the major and minor caliceal fornices are covered and interconnected by these atypical cells; they appear less frequently in the distal regions of the renal pelvis of all mammals studied, and are generally not present in the ureter [4, 6, 7]. These cells, although separated from each other for much of their length by connective tissue, have areas of close apposition with other atypical cells and with bundles of typical smooth muscles, suggesting that they may form specialized preferential pathways of electrical conduction [4]. It has also been suggested that the pacemaker activity of the renal pelvis arises from the intrinsic electrical activity of these atypical smooth muscle cells [6, 7]. The decrease in the number of these interconnecting atypical cells in more distal regions of the renal pelvis has been thought to explain both the gradient of spontaneous activity in the upper urinary tract and the ability of more proximal regions of the renal pelvis to drive ureteric peristalsis.

The nature of the ionic channels underlying the electrical activity in the upper urinary tract has been extensively studied only in the ureter, in the intact tissue [12, 13] and at the single cell level [10, 11, 14, 15]. In contrast, there have only been a few attempts to record the electrical activity in the guinea-pig renal pelvis, although some preliminary data has been obtained with the sucrose gap technique [23] and with intracellular microelectrodes [5, 20, 21, 26]. In the present experiments, we have further characterized the nature of the electrical activity recorded in the smooth muscle cells of the proximal renal pelvis, examining the effects of intramural nerve stimulation on the spontaneous electrical and mechanical activity. Some of these results have been presented previously in brief [24, 25].

Materials and methods

Tension recordings

Guinea-pigs (300–600 g) of either sex were stunned and bled. Kidneys were removed through an abdominal incision and placed in a pre-oxygenated physiological saline at room temperature. The renal pelvis was exposed by careful dissection of the parenchyma and surrounding adipose and connective tissues. The renal pelvis was then opened and the resultant sheet of tissue pinned out in the dissecting dish. Circumferentially cut strips (2 mm wide, 10–15 mm long) were cut from the proximal and mid portions of the renal pelvis (Fig. 1A) and suspended in a 2-ml organ bath (at 35 °C). One end of each strip was attached to the bottom of the organ bath; the other end was attached to a Grass force displacement transducer (Grass FT03C) connected to a Grass Polygraph (Model 79D).

Intracellular recordings

Electrophysiological recordings were made from strips (5 mm wide, 5 mm long) of proximal renal pelvis pinned, serosal side up, in an electrophysiological recording chamber, which was placed on an inverted microscope. The recording electrode and tissue movements were viewed on a TV monitor. Cells were impaled with fine borosillicate glass microelectrodes filled with 2 M KCl and having resistances of 50–150 MΩ. Membrane potentials were recorded with a standard unity-gain pre-amplifier and stored on a video cassette recorder using a digital data recorder (List VR-10). Portions of data were later digitized using a Labmaster analog-to-digital interface (Axon Instruments), a personal computer (NEC Powermate 286 Plus) and Axotape 1.1 (Axon Instruments) software. In both electrophysiological and tension recording experiments, intramural nerves were stimulated using silver-silver chloride wires placed on either side of the muscle strips and connected to a Grass (S88) stimulator. Stimulating parameters (pulses 0.3 ms in duration, 100–150 V at 10–50 Hz for 1–10 s) were chosen to avoid direct muscle stimulation.

Data analysis

The waveforms of the spontaneous electrical events recorded in these preparations varied considerably in their time course (see below). Various parameters of these spontaneous events were measured: (1) the membrane potential (MP) was measured 600 ms preceding the peak of a spontaneous event; (2) the upstroke amplitude of the initial spike, when present, and (3) the half-amplitude duration of these events. The half-amplitude durations were measured from the time the upstroke of either the simple action potential or the initial spike of the more complex action potentials was half maximal. In each cell, these parameters were averaged from four control action potentials and compared with the averaged parameters from four action potentials recorded after 2–5 min exposure to a test drug. These parameters were then averaged for a number of cells as indicated. Tests for significant changes of action potential parameters during an experiment were made using a paired Student's *t*-test, unless otherwise stated.

Solutions and drugs used

The bicarbonate-buffered physiological saline contained (mM): Na^+ 146; K^+ 5; Ca^{2+} 2.5, Mg^{2+} 1, Cl^- 132, HCO_3^- 25, $H_2PO_4^-$ 1, glucose 11. This solution had a pH of 7.3–7.4 (at 35°C) after being bubbled with 95% O_2:5% CO_2. Atropine sulphate, histamine dihydrochloride, tetrodotoxin (TTX) (all Sigma) and tetraethylammonium (TEA) (Eastman Kodak) were freshly prepared in distilled water as stock solutions (0.1–10 mM); nifedipine (10 mM) was dissolved in 100% ethanol. Drugs were diluted with physiological saline to their final concentrations indicated. Solutions were rigorously bubbled before use to restore any changes in pH.

Results

Tension recordings

Strips of proximal and mid renal pelvis developed spontaneous contractile activity after some 30–60 min equilibration in the organ bath (at 35 °C); during this time these contractions grew in amplitude and became regular. The frequency of contraction of the proximal strips (4.8 ± 0.3/min) was significantly greater than strips taken from the mid region (1.8 ± 0.2/min; $P < 0.05$, $n = 10$ preparations) of the renal pelvis (Fig. 1Aa, b) [3, 9]. The frequency of these contractions was particularly dependent on temperature, decreasing approximately 50% when the temperature was lowered 5 °C. Spontaneous contractions were observed for 2–4 h, after which they decreased in frequency and occurred irregularly.

We have confirmed previous reports [16, 17] that the repetitive stimulation of the intramural nerves (at 5–50 Hz for 1–10 s) produced a frequency-dependent positive inotropic and chronotropic effect on the spontaneous contractions of the renal pelvis. In the proximal renal pelvis, nerve stimulation at 5, 10, 20 and 50 Hz (100 V for 2 s), respectively, induced a 18 ± 4 ($n = 4$), 25 ± 5 ($n = 4$), 42 ± 6 ($n = 11$) and 43 ± 5 ($n = 7$)% increase in the contraction amplitude over their controls. In the mid renal pelvis, the percentage increases in contraction amplitude to these stimulation parameters were 14 ± 6 ($n = 4$), 16 ± 7 ($n = 4$), 32 ± 6 ($n = 10$) and 38 ± 8 ($n = 7$)%, respectively. These increases in contraction amplitude peaked 30–60 s after nerve stimulation and slowly returned to control levels over 2–5 min. In contrast, the increases in contraction frequency (approximately two to threefold) to nerve stimulation lasted for only 10–30 s, and were therefore more difficult to quantify. These effects of nerve stimulation were unaffected by the addition of atropine (3 μ*M*) and/or guanethidine (3 μ*M*), but were abolished by the previous application of TTX (1.6 μ*M*) or capsaicin (10 μ*M*) [16, 17, 26].

Electrical recordings

Recently, we have suggested that the action potentials recorded in the proximal renal pelvis could be designated to come from three separate cell populations: "pacemaker", "intermediate" and "driven" cells [26]. Figure 1B shows typical examples of these three types of spontaneous action potentials recorded in the proximal renal pelvis, often from the same region. The majority of cells (>120 cells successfully impaled) were driven cells with stable MPs of -56.1 ± 1.2 mV ($n = 36$ cells measured). The spontaneous action potentials recorded in these cells consisted of an initial rapid upstroke which triggered a characteristic spike, followed by a period of fast potential oscillation and then a plateau phase of 200–1200 ms duration (Fig. 1Bb). These "ureter-like" action potentials [12, 14] often occurred abruptly, but were sometimes preceded by a small depolarizing pre-potential. Action potential repolarization was often followed by a period of undershoot or "afterhyperpolarization", 5–10 mV in amplitude and 2–5 s in duration (Fig. 1Bc). The most negative membrane potential reached during the afterhyperpolarization was -64.4 ± 3 mV ($n = 6$). These driven action potentials were also followed by a distinct

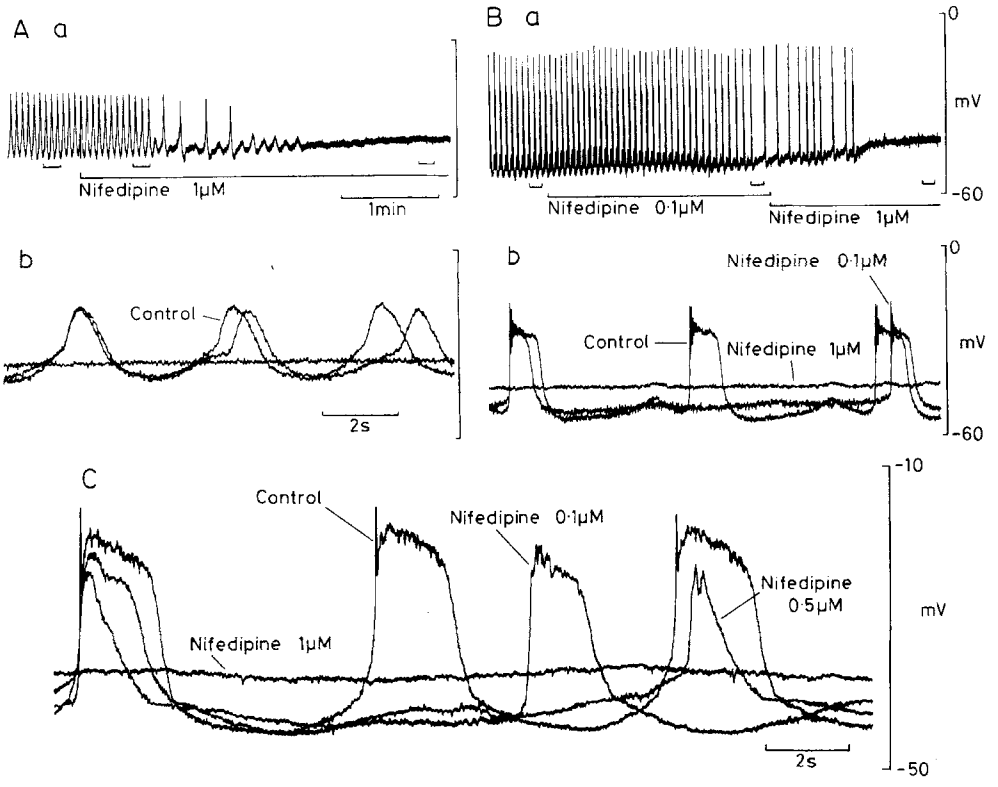

Fig. 2A–C Effects of nifedipine (0.1–1 μ*M*) on the spontaneous action potentials recorded in a pacemaker (**A**), driven (**B**) and intermediate cell (**C**) of the proximal renal pelvis. The effects of nifedipine on the action potentials *indicated by the horizontal brackets* (**A***a*, **B***a*) were best observed on an expanded time course (**A***b*, **B***b*). **C** Action potentials were recorded in an intermediate cell bathed in control saline and after 3 min exposure to the concentrations of nifedipine indicated

refractory period (duration 5–20 s), during which subsequent action potentials either failed to fire or were of reduced amplitude and duration. When action potentials failed to fire during this refractory period, the underlying triggering events had a simple "pacemaker" wave form (Fig. 1C). This period between action potential discharge, however, did not represent an absolute refractory period. Direct electrical stimulation (10 V for 200 ms) of the smooth muscle usually triggered an action potential of similar amplitude and duration when applied at > 4 s after a preceding spontaneous event. If this electrical stimulation was applied within shorter periods, however, only truncated action potentials could be triggered (data not shown).

"Pacemaker" cells fired spontaneous action potentials with a simple waveform, consisting of a slowly developing pre-potential, a relatively slow upstroke and a simple repolarizing phase. The MP of these pacemaker cells was -42.1 ± 2.9 mV ($n = 9$) (Fig. 1Ba). "Intermediate" cells were identified as cells with action potentials displaying properties of both the simple and complex action potentials. These cells had MPs of -45.5 ± 1.7 mV ($n = 15$) and action potentials with an initial spike, often occurring after a slow pre-potential. The initial spike was then followed by a plateau and a slow repolarizing phase (Fig. 1Bc). The averaged parameters of these action potentials are shown in Table 1, where it can be seen that: (1) the MP of driven cells was significantly more negative than either pacemaker or intermediate cells; (2) the initial spike amplitude increased significantly with more negative MPs, and (3) the frequency of action potential discharge in driven cells was not significantly different to that recorded in pacemaker cells, but was significantly smaller than the frequency observed in intermediate cells. Spontaneous action potentials were generally recorded for 3–4 h, after which they occurred less frequently and finally ceased to fire, establishing a stable MP of about -50 mV. Action potential discharge could be re-established, however, by raising the extracellular K^+ concentration (to 26 mM) or upon the addition of the prostaglandin $F_{2\alpha}$ agonist, dinoprost (100 nM).

Effects of Ca^{2+} entry blockers

Application of the dihydropyridine, nifedipine (0.1–1 μ*M*), blocked all spontaneous contractions in both proximal and mid portions of renal pelvis ($n = 4$). This was associated with a blockade of action potential discharge in all three cell populations. Figure 2 illustrates the effects of nifedipine (0.1–1 μ*M*) on the action potentials recorded in a pacemaker (Fig. 2A), driven (Fig. 2B) and intermediate (Fig. 2C) cell. Onset of nifedipine blockade was characterized by a gradual reduction of the action potential duration and frequency; the MP in pacemaker, intermediate and driven cells depolarized to settle, respectively, at -36 mV ($n = 1$) (Fig. 2A), -39 mV ($n = 1$) (Fig. 2C) and -43.8 ± 2.6 mV ($n = 4$) (Fig. 2B), from their control MPs of

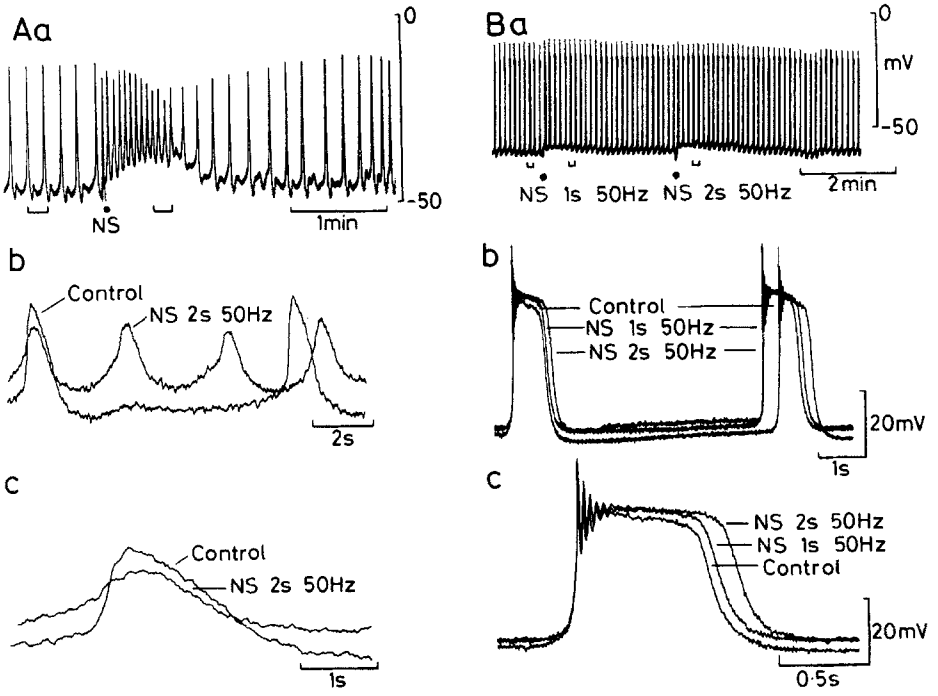

Fig. 3 Nerve stimulation (NS) (50 Hz for 1–2 s) induced a membrane depolarization in both pacemaker ($n = 3$) (**A***a, b*) and driven ($n = 6$) (**B***a, b*) cells. Such depolarization in pacemaker cells increased the frequency of action potential discharge, but reduced their amplitude (**A***c*). In driven cells, nerve stimulation induced a significant increase in only the half-amplitude duration of the spontaneous action potentials (**B***c*)

-42, -47 and -52.8 ± 1.7 mV ($P < 0.05$). Cd^{2+} (0.1 mM), an inorganic Ca^{2+} entry blocker, produced a similar blockade of action potential discharge and membrane depolarization (data not shown).

Effects of stimulating intramural nerves

In the pacemaker cell shown in Fig. 3Aa, nerve stimulation (50 Hz for 2 s) induced a membrane depolarization of 11 mV (control MP -51 mV), which peaked after 30 s and slowly declined over 60 s. This depolarization increased the frequency of action potential discharge from 5.8 to 17.6/min, but decreased their amplitude from 31 mV to 16 mV (Fig. 3Ab, c). Similar results were recorded in two other pacemaker cells, where nerve stimulation induced membrane depolarizations of 2 and 14 mV respectively (average depolarization 9.1 ± 3.5 mV, $n = 3$).

In six driven cells, nerve stimulation (50 Hz for 2 s) induced a membrane depolarization of 2 mV to -53.6 ± 1.3 mV (control MP -55.6 ± 1.6 mV; $P < 0.05$) (Fig. 3Ba, b), which peaked in 10–30 s and declined over 1–3 min (Fig. 3Ba). The duration of the first spontaneous action potential after nerve stimulation was always shorter than the control action potentials (data not shown). Subsequent action potentials, however, increased to a maximum half-amplitude duration of 908 ± 106 ms (control duration 812 ± 95 ms; $P < 0.05$, $n = 6$) (Fig. 3Bc) and a peak frequency of 11.1 ± 1.4/min (control frequency 9.9 ± 1.1/min). Both parameters then declined over the same time course as the change in membrane potential. The afterhyperpolarizations following each action potential were also reduced during this period. All these effects of nerve stimulation were all blocked by TTX (1.6 μM) (data not shown).

The membrane depolarization induced upon repetitive nerve stimulation was most clear after action potential blockade. In Fig. 4, nerve stimulation (50 Hz for 2 s) applied to a pacemaker cell, previously arrested with nifedipine (1.0 μM), induced a biphasic depolarization with an initial transient component of 5 mV, followed by a more slowly developing component which peaked in about 30 s and then decayed slowly over 1–2 min (Fig. 4Aa). Both components increased in amplitude when a more intense stimulation (50 Hz for 5 s) was used (Fig. 4Ab). They were also both blocked by TTX (1.6 μM) (Fig. 4Ba, b), confirming they were indeed nerve-evoked responses. Nifedipine-arrested driven cells exhibited a similar TTX-sensitive transient depolarization (of about 5 mV), followed by a long-lasting depolarization (1–2 mV in amplitude) (data not shown).

Discussion

The present experiments have confirmed our recent suggestion that the spontaneously occurring action potentials recorded in the proximal renal pelvis can be designated as coming from three groups of cells which we have termed pacemaker, intermediate and driven cells

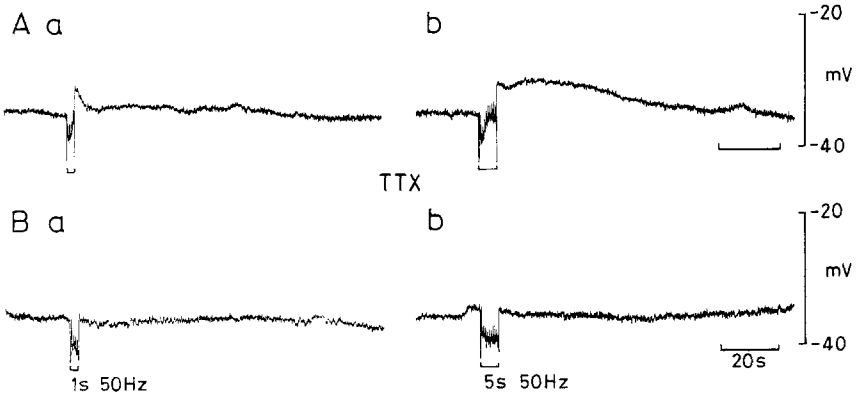

Fig. 4 Biphasic membrane depolarizations evoked in a nifedipine (1 μ*M*)-arrested pacemaker cell upon repetitive intramural nerve stimulation (50 Hz), applied for 2 s (**A***a*) and 5 s (**A***b*), were blocked upon the application of TTX (1.6 μ*M*) (**B***a*, *b*)

Table 1 Properties of the action potentials recorded in smooth muscle cells of the guinea-pig proximal renal pervis

	Membrane potential (mV)	Frequency (/min)	Spike amplitude (mV)	Half-amplitude duration (ms)	Number of cells (*n*)
Pacemaker cells	− 42.1 ± 2.9	8.0 ± 1.3	19.2 ± 2.0	864 ± 80	9
Intermediate cells	− 45.5 ± 1.7	10.8 ± 1.0	24.6 ± 1.3 c	938 ± 50	15
Driven cells	− 56.1 ± 1.2 a, b	6.3 ± 0.5 b	35.4 ± 1.3 a, b	1249 ± 112	36

a, *b*, *c* denotes a significant difference between pacemaker and driven cells, intermediate and driven cells, and pacemaker and intermediate cells, respectively (unpaired *t*-test, $P < 0.05$)

[26]. It has often been proposed that "atypical" smooth muscle cells, which lie near the epithelial surface, act as the "pacemaker cells" of the renal pelvis [6, 7]. This is perhaps supported in the present experiments as pacemaker-like action potentials were recorded from cells "deep" within the preparation and not near the serosal surface. It is tempting to suggest, therefore, that there is a gradient of MPs (from − 40 to − 55 mV) across the wall of the renal pelvis. The pacemaker cells lying near the epithelial border are presumably coupled to driven cells at the serosal surface by intermediate cells, the MPs of these intermediate cells being determined, to some extent, by the degree of this coupling. Alternatively, intermediate and driven cells may well represent recordings from typical smooth muscles which are located relatively more distal to the pacemaker cells, so that the gradient of MP would, in fact, be along the longitudinal axis of the renal pelvis. This coupling of pacemaker cells to driven cells could also be demonstrated both electrically and pharmacologically, pacemaker potentials being recorded in driven cells during their "effective" refractory periods and in the presence of indomethacin [26]. Driven and intermediate cells may well be coupled to more than one pacemaker region, as evident by the significantly greater frequency of discharge in intermediate cells (Table 1) and by the number of subthreshold pacemaker potentials in driven cells (Fig. 1C). The regular frequency of contraction and action potential discharge in driven cells, however, suggests that this frequency is determined by either the "strongest" pacemaker region, or by the effective refractory period of the driven cells (Fig. 1C).

The organic Ca^{2+} entry blocker nifedipine (0.1–1 μ*M*) reduced the frequency, amplitude and duration of the action potentials recorded in pacemaker, intermediate and driven cells of the renal pelvis in a concentration- and time-dependent manner (Fig. 2). This reduction and final blockade of action potential discharge was associated with a slow reduction and finally abolition of contractile activity, and with membrane depolarization to between − 35 and − 40 mV (Fig. 2). These results suggest that the influx of Ca^{2+} through voltage-activated "L-type" Ca^{2+} channels [15] is responsible for the upstroke of the three action potential types, which leads to a rise in the internal Ca^{2+} concentration and normal muscle contraction. The apparent membrane depolarization in the presence of nifedipine presumably arises from the loss of time- and voltage-dependent K^+ conductances associated with the downstroke and refractory period of each blocked action potential so that the membrane settles at its "true" membrane potential.

The absence of a functional sympathetic or parasympathetic nerve supply to the renal pelvis has been previously well established. Transmural electrical stimulation activates capsaicin-sensitive sensory nerves which increase both the strength and frequency of the spontaneous contractions in the renal pelvis. The tachykinins, substance P and neurokinin A [17], induced similar

positive chronotropic and inotropic effects as nerve stimulation. The addition of MEN 10376, a neurokinin A antagonist, suppresses the effects of neurokinin A and nerve stimulation to reveal a nerve-mediated negative inotropic response which was blocked by the C-terminal fragment of human α-calcitonin gene-related peptide (hCGRP) [17]. Repetitive nerve stimulation therefore releases both excitatory and inhibitory peptides from capsaicin-sensitive sensory nerves, with the predominant effect being excitatory. In the present experiments, nerve stimulation depolarized pacemaker cells approximately 10 mV which increased the frequency of their action potential discharge for about 1 min. Driven cells were only depolarized 2 mV upon nerve stimulation, which was associated with a small increase in their frequency of action potential discharge. There was, however, a prolonged increased in the half-amplitude duration of the action potentials recorded in driven cells which was associated with a blockade of the afterhyperpolarization. This increase in duration lasted for several minutes, the same period over which the contraction amplitude was increased. Given this correlation of strength of contraction with the length of the plateau phase of the action potential in driven cells upon nerve stimulation, we would like to suggest that driven smooth muscle cells represent the major contractile structures in the renal pelvis.

The nerve-induced depolarizations in cells of the renal pelvis, recorded in both the absence or presence of nifedipine, consisted of an initial rapid transient component, followed by a slowly developing, slowly decaying component (Fig. 4). These membrane depolarizations presumably arise from a modulation of the channels responsible for normal action potential discharge, or from the opening/closing of additional cationic or chloride channels. At present, we can only conclude that it is unlikely that a modulation of Ca^{2+} channels is responsible. In nifedipine, all "L-type" channels would have been blocked. Moreover, any nifedipine-resistant Ca^{2+} channels should have been mostly inactivated at MPs of about -35 to -40 mV (Fig. 2) and therefore unavailable to contribute to the nerve-induced depolarizations [2]. We can speculate, however, that the initial transient depolarization is likely to be responsible for the short-term acceleration of action potential discharge in pacemaker cells and for the transient reduction in the duration of the driven cell action potential recorded during, or immediately after, nerve stimulation. Clearly, the effects of nerve stimulation require further study; it would be of particular interest to record the electrophysiological effects of nerve stimulation in the presence of either a tachykinin, or CGRP, antagonist [17].

In summary, we have confirmed the presence of three populations of spontaneously active smooth muscle cells in the guinea-pig proximal renal pelvis which we believe are responsible for the generation and propagation of peristaltic contractions in the upper urinary tract. We have speculated that the "pacemaker" action potentials were recorded from subepithelial "atypical" smooth muscle cells, the frequency of these pacemaker action potentials depending on their membrane potential and, in some way, on their number. The frequency of action potential discharge in driven cells was not particularly dependent on their membrane potential suggesting that, while they were being driven by pacemaker cells, their frequency of discharge was mostly determined by their refractory period. We have also correlated the long-term positive inotropic effects of nerve stimulation with a significant increase in the duration of the action potentials in driven cells.

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ANNOUNCEMENTS

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The 1996 Duke Urologic Assembly Meeting
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Vienna, Austria
3rd European Chlamydia Meeting
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