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Actions of Prostaglandins and Indomethacin on the Electrical and Mechanical Properties of Smooth Muscle Cells of the Guinea-Pig Ileocecal Junction

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Abstract. The effects of prostaglandins (PGs) and indomethacin on the mechanical and membrane properties of the smooth muscle cells of the guinea-pig ileocecal junction were studied using microelectrode and tension recording techniques and radioimmunoassay to determine levels of PGs. In the guinea-pig ileocecal junction, we found two distinct cell populations-cells with, and without spontaneous electrical and mechanical activities. PGs (PGE_1, PGE_2, $PGF_{2\alpha}$) in low concentrations suppressed spontaneous mechanical activity. Correspondingly, PGs ($>10^{-7}$ M) suppressed both spontaneously generated spikes and evoked spikes, presumably due to an increase in the threshold for generation of action potential. On the contrary, indomethacin evoked rhythmic spontaneous contractions in the mechanically quiescent muscle preparations and reduced the PGE and PGF content of the muscle. Spontaneous spike discharges occurred during the indomethacin-induced contractions. The spontaneous electrical and mechanical activities induced by indomethacin were suppressed by PGs, in low concentrations. These results indicate that under physiological conditions, the endogenous PGs in the muscle may play an important role in regulating muscle tone as well as the membrane properties, thereby contributing to the regulation of motility of the intestine and possibly the sphincter.

Key words: PGs – Indomethacin – Smooth muscle – Ileocecal junction

Introduction

There are several lines of evidence which indicate that endogenous prostaglandins (PGs) may play a role in regulating the motility of gastrointestinal smooth muscles. These include, i) prostaglandin E series (PGEs) are produced and released from muscle or mucosal layers of gastro-intestinal tract (Bennett et al. 1967; Jouvenaz et al. 1970; Ferreira et al. 1972; Botting and Salzmann 1974; Sanders and Ross 1978; Sanders 1978), ii) exogenous PGs in relatively low concentrations and inhibitors of PG synthesis modify gastrointestinal activities (Bennett et al. 1968; Shehadeh et al. 1969; Türker and Onur 1971; Ferreira et al. 1972; Willis et al. 1974; Dajani et al. 1975), and iii) changes in the motility of intestine induced by inhibitors of PG synthesis correlate with changes in the concentration of endogenous PGs (Sanders and Ross 1978).

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In the preceding manuscript (Kubota 1982), two distinct cell populations such as the cells with or without spontaneous spike activity were observed in smooth muscle cells of the guinea-pig ileocecal junction, and individual preparations were characterized by cell populations showing spontaneous spike activity, whereas other preparations showed few. In an attempt to clarify whether or not these two distinct cell populations correlate with the endogenous PGs, we compared the effects of exogenous PGs and indomethacin on the electrical and mechanical properties of the smooth muscle cells of guinea-pig ileocecal junction, and determined by radioimmunoassay the PGE and PGF levels in the muscle tissue from both control and indomethacin-treated guinea-pigs.

Methods

The apparatus and experimental procedures used for microelectrode or tension recording were the same as those described in the preceding paper manuscript (Kubota 1982).

A modified Krebs' solution (hereafter referred to as Krebs') of the following composition was used (mM) Na^+ 137.4, K^+ 5.9, Mg^{2+} 1.2, Ca^{2+} 2.5, Cl^- 134.0, HCO_3^- 15.5, $H_2PO_4^-$ 1.2 and glucose 11.5 equilibrated with 97% O_2 and 3% CO_2. To observe the effects of indomethacin on the electrical and mechanical properties of the smooth muscle cells of the ileocecal junction, in vitro, indomethacin was dissolved with an equimolar amount of sodium bicarbonate, and diluted to the final concentration with Krebs' solution. Prostaglandin 1 mg/ml was prepared in 99% ethanol and kept in a freezer. Dilutions with Krebs' solution were made immediately before the experiments. The following drugs were used; indomethacin, prostaglandin E_1, prostaglandin E_2 and prostaglandin $F_{2\alpha}$.

PGE and PGF contents in the muscle tissue of the guinea-pig ileocecal junction were estimated in both control and indomethacin-treated animals; seven guinea-pigs, weighing 200–300 g, were given indomethacin suspended in 0.5% sodium carboxymethyl cellulose subcutaneously in doses of 15 mg/kg body weight per day for 3 days, and control animals were given injections of an equal volume of the vehicle only. The guinea-pigs were stunned and bled, the ileocecal junction and circular muscle layers were separated from the mucosal layer and connective tissue, and immediately frozen with liquid nitrogen. The dissection took about 3–4 min to complete.

The frozen samples (8–12 mg) were suspended into ice-cold methanol (3 ml) and homogenized. Chloroform (6 ml) was then added and the homogenate was allowed to equili-

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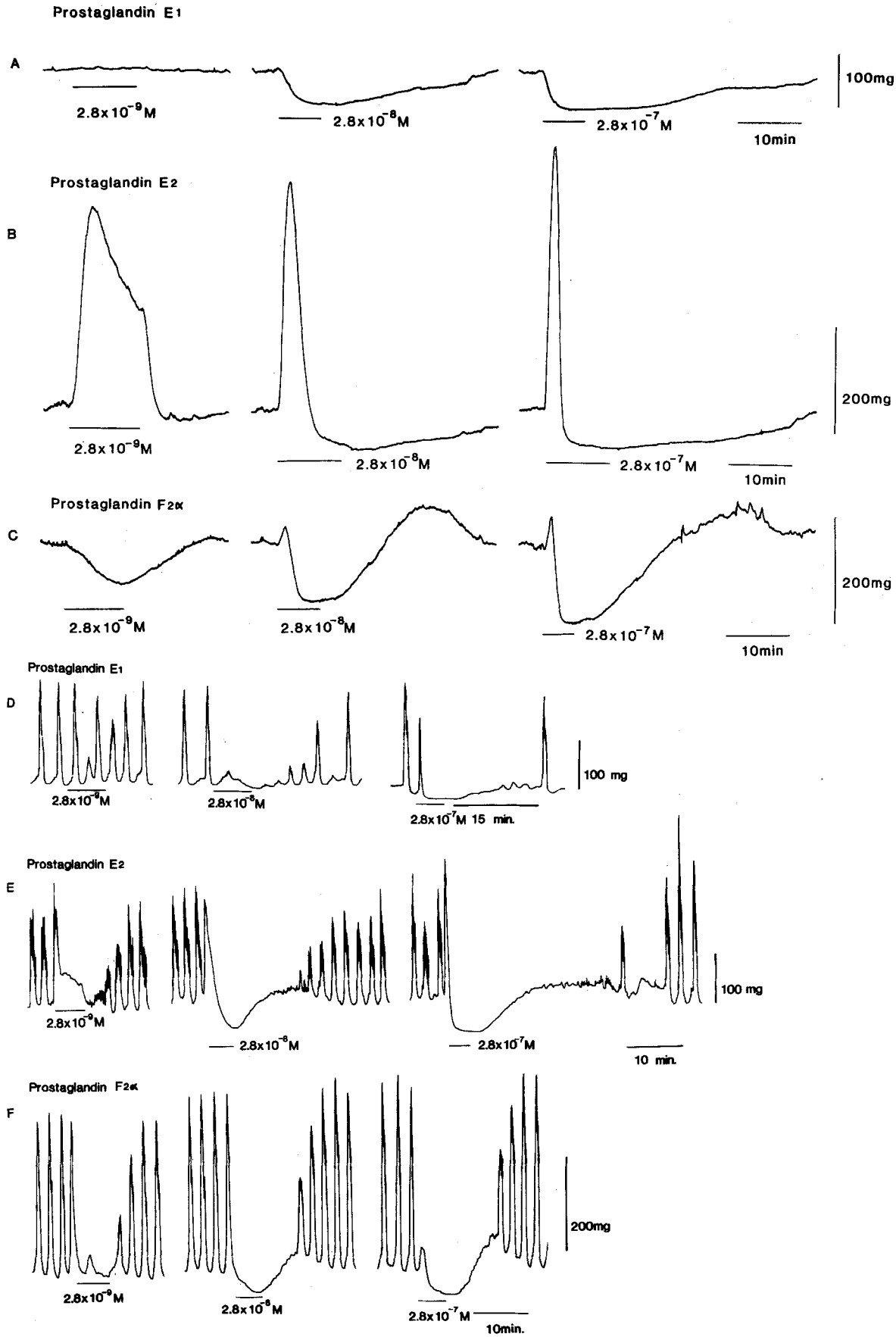

Fig. 1A—F. Effects of PGs (PGE_1, PGE_2 and $PGF_{2\alpha}$) on the mechanical activities of smooth muscle preparations of the guinea-pig ileocecal junction. In **A**—**C** the tissue was mechanically quiescent and in **D**—**F** there was spontaneous mechanical activity. *Bars* indicate the duration of exposure to PGs in various concentrations ($2.8 \times 10^{-9} - 2.8 \times 10^{-7}$ M). (**A**) PGE_1, (**B**) PGE_2, (**C**) $PGF_{2\alpha}$, (**D**) PGE_1, (**E**) PGE_2 and (**F**) $PGF_{2\alpha}$

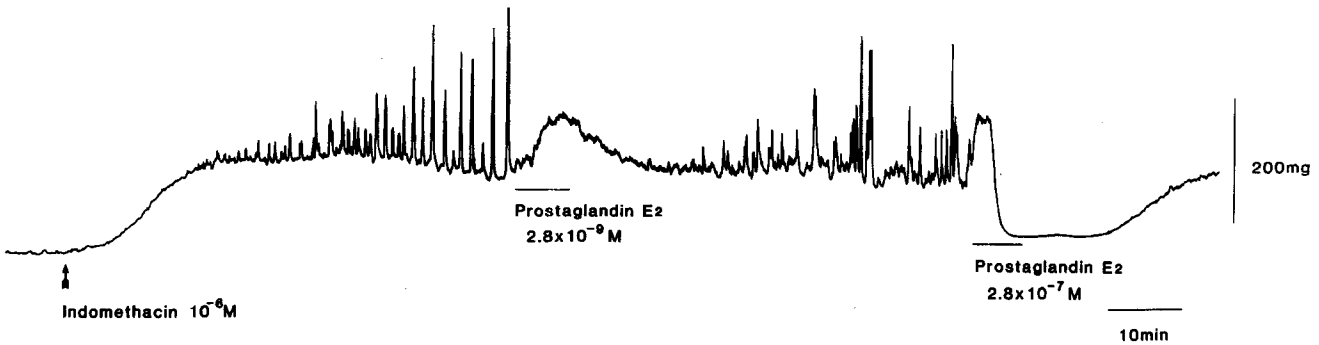

Fig. 2. Effects of indomethacin (10^{-6} M) on the mechanical properties of smooth musce preparations which had no spontaneous mechanical activity, and effects of PGE_2 (2.8×10^{-9} M or 2.8×10^{-7} M) on the contractions induced by indomethacin

brate at room temperature for 30 min after which it was filtered and crude lipid extracts were obtained from the filtrate (Folch et al. 1957). The eluted solution was evaporated under a stream of nitrogen gas, and the residue suspended in saline and adjusted to pH 3.0 with 1 N HCl. After re-extraction of PGs with ethylacetate, the solvent was evaporated. Dried extracts were redissolved with solvent I (benzene:ethylacetate = 60:40 (V/V)], and chromatographed on columns of silicic acid (Jaffe and Behrman 1974). The fractions containing PGs were eluted with solvent II [benzene:ethylacetate:methanol = 60:40:20 (V/V)], and after evaporation of the eluted solution, these fractions were dissolved in phosphate-buffer solution (pH 7.54) containing gelatin, then radioimmuno-assayed for PGE_2 and $PGF_{2\alpha}$. The recovery of PGs was examined at each step of the extraction using [^{3}H] PGE_2 or $PGF_{2\alpha}$ (New England Nuclear Corp., Boston, USA), and showed the final recovery of PGE_2 or $PGF_{2\alpha}$ to be consistently in the range 60 – 70 %. Antibodies to PGE_2 or $PGF_{2\alpha}$ obtained from rabbits were gifts from the Research Institute of Ono Pharmaceutical Co., LTD (Osaka, Japan). The antiserums against PGE_2 or $PGF_{2\alpha}$ were capable of binding 50 % of 10,000 cpm of [^{3}H]PGE_2 or [^{3}H]$PGF_{2\alpha}$ at a final dilution of 1:750 or 1:12,000 respectively. The antibodies against PGE_2 or $PGF_{2\alpha}$ showed cross reactivity to PGE_1 (53.5 % of the value for PGE_2) or to $PGF_{1\alpha}$ (41.7 % of the value for $PGF_{2\alpha}$) respectively, thus making it possible to express prostaglandin activity as the total of PGE and PGF. Radioimmunoassay for PGE or PGF was performed according to the method described by Jaffe and Behrman (1974). The range of the minimum detection of PGE_2 or $PGF_{2\alpha}$ was 30 – 40 pg, and standard curves using freshly prepared PGE_2 or $PGF_{2\alpha}$ were obtained for each assay. Concentrations of PGE or PGF were corrected for recovery from extraction and chromatography and normalized as nanograms PGE or PGF per gram wet weight of tissue sample.

Results

Effects of PGs on the Mechanical Properties of the Guina-Pig Ileocecal Junction

Figure 1A – C shows the effects of PGs (PGE_1, PGE_2 and $PGF_{2\alpha}$) on the mechanical properties of preparations of the guinea-pig ileocecal junction which had no spontaneous phasic activity. PGE_1 ($< 2.8 \times 10^{-9}$ M) produced no phasic activity, although in higher concentrations ($> 2.8 \times 10^{-8}$ M) this agent induced a long-lasting relaxation. In contrast to the effects of PGE_1, PGE_2 ($> 2.8 \times 10^{-9}$ M) evoked phasic contraction of the muscle tissue, and in higher concentrations ($> 2.8 \times 10^{-8}$ M) evoked biphasic mechanical responses, i. e. an initial phasic contraction followed by a long-lasting relaxation.

$PGF_{2\alpha}$ ($> 2.8 \times 10^{-8}$ M) also evoked biphasic responses of the muscle tissue, however the amplitude of the initial contraction was much smaller compared than that evoked by PGE_2. Lower concentrations of $PGF_{2\alpha}$ ($< 2.8 \times 10^{-9}$ M) produced a gradual decrease in muscle tone.

Approximately 10 % of the examined muscle preparations showed spontaneous phasic activity of constant amplitude and frequency (0.2 – 0.5 per min). All of the PGs we examined completely suppressed this spontaneous phasic activity (Fig. 1D – E). The subsequent effects of each PG was then broadly similar to that observed in preparations without spontaneous mechanical activity, i. e. PGE_1 reduced the muscle tone and PGE_2 or $PGF_{2\alpha}$ evoked biphasic changes in the muscle tone.

Effects of Indomethacin on the Mechanical Properties of the Smooth Muscle Cells

Since the majority of the examined muscle preparations were mechanically quiescent and in view of our observation that any spontaneous mechanical activity in the smooth muscle strips was abolished by low concentrations of exogenous PGs, it was of interest to observe the effects of indomethacin on muscle preparations which had no spontaneous phasic activity.

Figure 2 shows an example of such an experiment. On treatment with indomethacin (10^{-6} M) there was a delay of several minutes and then tone gradually increased to reach a plateau with superimposed rhythmic contractions. The amplitude of these "spontaneous" contractions gradually increased whilst the indomethacin remained in the organ bath and a maximum amplitude was reached about 1 h after the drug was first introduced. The frequency of the spontaneous phasic contractions ranged from 0.1 to 0.4 per min.

Figure 2 also shows the effects of PGE_2 on the mechanical activities induced by the presence of indomethacin. Exogenous PGE_2 (2.8×10^{-9} M) further increased the muscle tone, and abolished the phasic mechanical activity, and in higher concentrations (2.8×10^{-7} M), PGE_2 evoked contraction followed by a long-lasting relaxation of the muscle tissue, just as in the normal Krebs' solution.

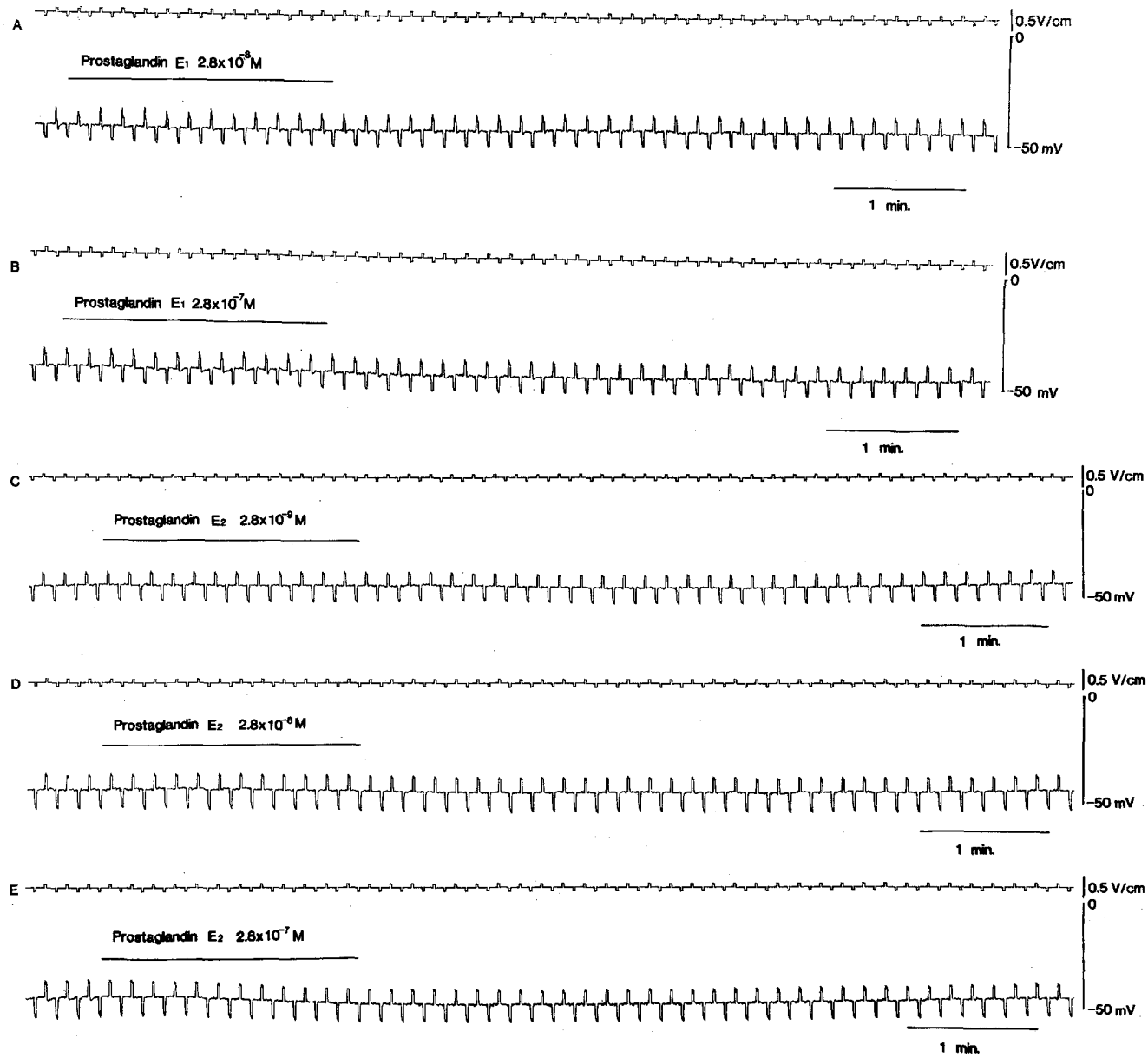

Fig. 3A—E. Effects of PGs (PGE_1 or PGE_2) on the resting membrane potential of smooth muscle of the guinea-pig ileocecal junction, and on electrotonic potentials and local responses evoked by inward and outward current pulses (1 s in duration). (**A** and **B**) Effects of 2.8×10^{-8} M or 2.8×10^{-7} M of PGE_1, (**C—E**) effects of PGE_2 2.8×10^{-9} M, 2.8×10^{-8} M and 2.8×10^{-7} M respectively

Effects of PGs on the Membrane Properties of the Smooth Muscle Cells

To observe the effects of PGs on the membrane properties, microelectrode recordings were employed.

As reported previously, the majority of smooth muscle cells of the ileocecal junctions were electrically quiescent, but muscle cells in small populations of preparations showed spontaneous spike discharges. We were, therefore, able to observe the effects of PGs in two distinct groups of the smooth muscle cells.

Figures 3 and 4 show the effects of PGs on the resting membrane potential, electrotonic potentials and local responses evoked in electrically quiescent smooth muscle cells by extracellularly applied inward and outward current pulses.

PGE_1 (up to 10^{-7} M) or PGE_2 ($2.8 \times 10^{-9} - 10^{-8}$ M) had no effect on the membrane potential or on the membrane resistance, as far as could be judged from the amplitude of electrotonic potentials. The membrane potential, for example, was -43.9 ± 3.4 mV ($\pm$SD, $n = 20$) in Krebs' solution, and -44.0 ± 2.8 mV ($\pm$SD, $n = 19$), -45.1 ± 2.4 mV ($\pm$SD, $n = 18$), and -45.4 ± 3.0 mV ($\pm$SD, $n = 19$) in 2.8×10^{-9} M, 2.8×10^{-8} M PGE_2 and 2.8×10^{-7} M of PGE_1 respectively. In these concentrations the PGE series reduced the amplitude of the local responses evoked by extracellularly applied outward current pulses, without affecting the amplitude of electrotonic potentials evoked by constant inward current pulses (1 s in duration). A higher concentration of PGE_2 (2.8×10^{-7} M) hyperpolarized the membrane from -43.5 ± 3.0 mV ($\pm$SD, $n = 20$) to -47.1 ± 3.0 mV ($\pm$SD, $n = 20$), and simultaneously reduced the amplitude of the electrotonic potentials to about 90% of its control value.

$PGF_{2\alpha}$, in low concentrations consistently hyperpolarized the membrane from -43.5 ± 2.5 mV to -45.9 ± 1.9 mV

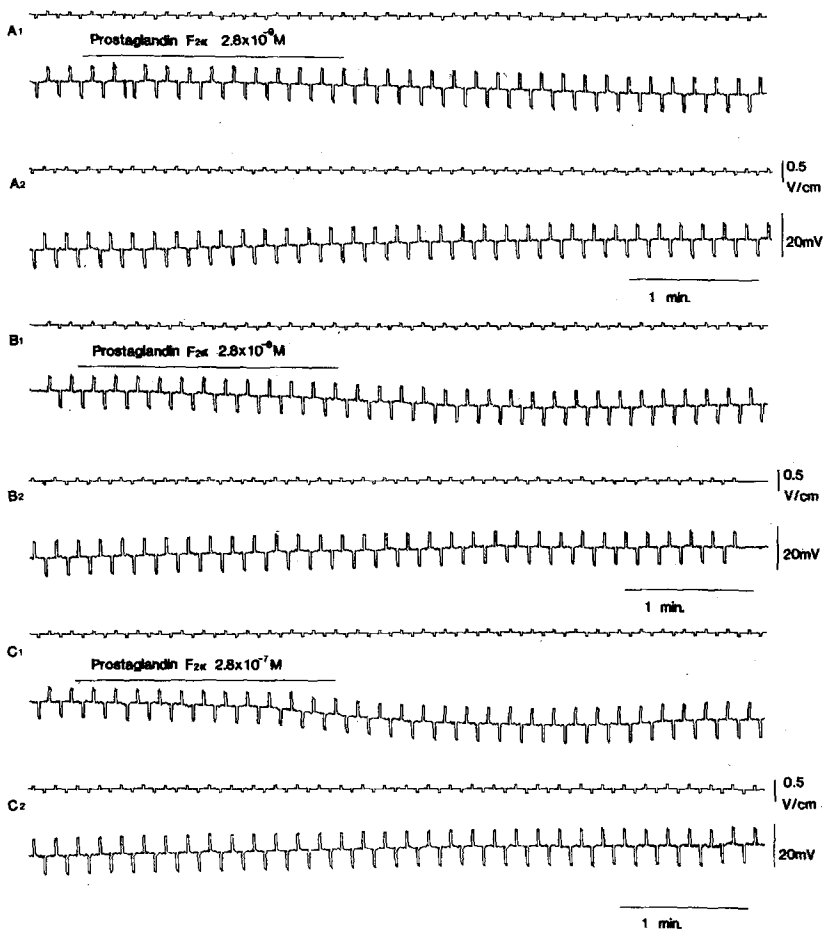

Fig. 4
Effects of $PGF_{2\alpha}$ (2.8×10^{-9} M – 2.8×10^{-7} M) on the resting membrane potential, and on electrotonic potentials and local responses evoked by inward and outward current pulses in smooth muscle of the guinea-pig ileocecal junction. A_1 and A_2, B_1 and B_2 and C_1 and C_2 are continuous recordings. *Bars* in A_1, B_1 and C_1 indicate the duration of exposure to 2.8×10^{-9} M, 2.8×10^{-8} M and 2.8×10^{-7} M of $PGF_{2\alpha}$ respectively

($\pm$SD, $n = 8$) with 2.8×10^{-9} M $PGF_{2\alpha}$, and to -51.4 ± 2.3 mV ($\pm$SD, $n = 8$) or to -54.6 ± 4.2 mV ($\pm$SD, $n = 9$) with 2.8×10^{-8} M or 2.8×10^{-7} M $PGF_{2\alpha}$. During these hyperpolarizations, there was no detectable change in the amplitude of electrotonic potentials (Fig. 4).

Figure 5A shows the effects of PGE_2 (2.8×10^{-9} M) on spontaneous changes in membrane potential of the muscle cells which mainly consisted bursts of spike discharges. PGE_2 reduced the frequency of spikes in each burst and the amplitude of individual spikes. After a few minutes of treatment, spontaneous spike activity was completely suppressed.

Smooth muscle cells which showed spontaneous spike activity usually responded in an all or none fashion to extracellularly applied outward current pulses (Fig. 5B). To observe the effects of PGs on the threshold potential for generation of action potentials, on the amplitude and on the rate of rise of evoked action potentials, PGE_1 was used, since this agent had little effect on the resting membrane potential. As shown in Fig. 5C, 2.8×10^{-8} M PGE_1 increased the threshold membrane depolarization required to trigger an action potential. In the presence of 2.8×10^{-7} M of PGE_1, an all or none action potential could not be evoked by extracellularly applied outward current pulses, although increasing the intensity of the outward current pulses slightly enhanced the amplitude of the abortive action potential (Fig. 5D). By contrast the amplitude of electrotonic potentials evoked by inward current pulses was not affected by the treatment with PGE_1 (2.8×10^{-7} M).

Effects of Indomethacin on the Electrically Quiescent Smooth Muscle Cells

Having established that low concentrations of exogenous PGs suppressed spontaneous spike discharges and increased the threshold depolarization for generation of action potentials, we set out to investigate the role that the endogenous prostaglandins might play in controlling membrane properties by studying effect of indomethacin on electrically quiescent muscle cells.

As shown in Fig. 6A and B, after about 30 – 60 min after the treatment with indomethacin (10^{-6} M), "spontaneous" action potentials with after-hyperpolarization were observed. The amplitude of the action potentials were in the range 10 to 50 mV, and the frequency was between 10 and 20 per min. These "spontaneous" discharges could be classified into two types, (i) regular burst of short trains of spike alternating with silent periods and (ii) continuous regular spike discharges.

These "spontaneous" action potentials were completely suppressed when PGE_2 (2.8×10^{-9} M) applied exogenously.

Figure 6C – F, shows the effect of indomethacin on the local responses evoked by the outward current pulses in electrically quiescent smooth muscle cells. In Krebs' solution, an increase in the intensity of the outward current pulses slightly enhanced the amplitude and rate of rise of the local response (Fig. 6 C_{1-3}). The maximum values for the amplitude and rate of rise of the local response were 8 mV and 0.2 V/s, respectively. In the presence of indomethacin, the amplitude and rate of rise of the local response gradually

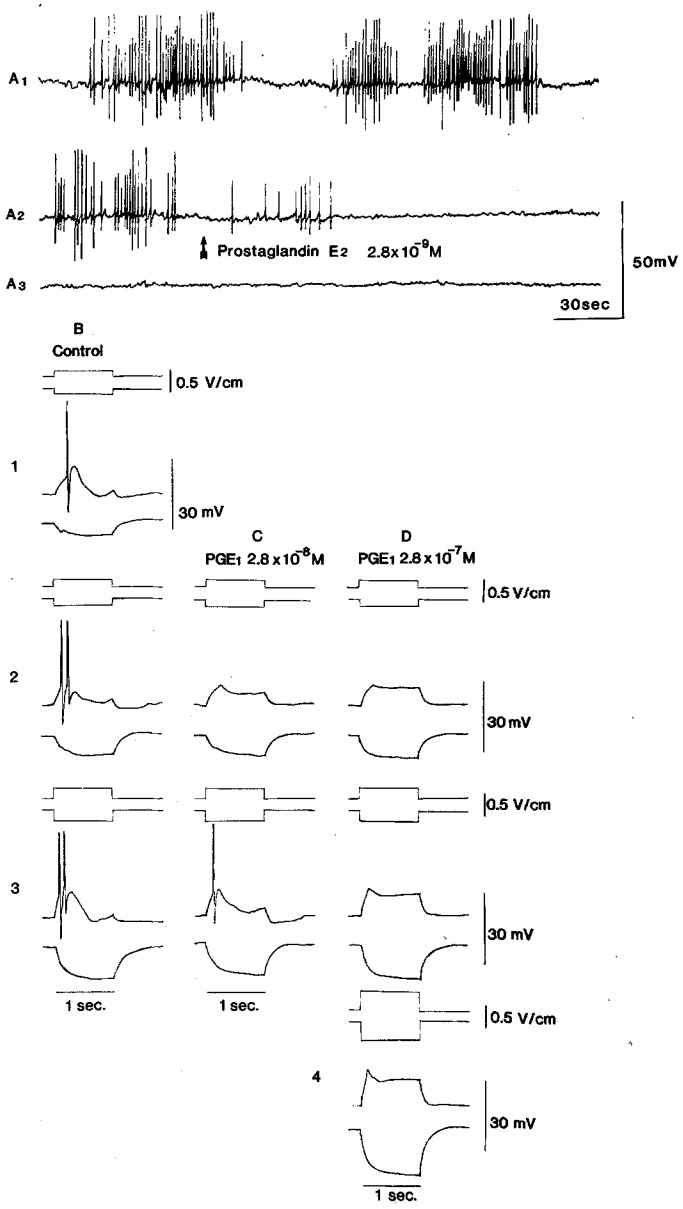

Fig. 5. (**A**) Effects of PGE_2 (2.8×10^{-9} M) on spontaneous spike discharges of the smooth muscle cells of guinea-pig ileocecal junction A_1, A_2 and A_3 are continuous recordings. (**B—D**) Effects of PGE_1 on the threshold for generation of action potentials and on the amplitude of the action potentials evoked by extracellularly-applied outward current pulses. (**B**) Control, (**C**) PGE_1 2.8×10^{-8} M and (**D**) PGE_1 2.8×10^{-7} M. Intensities of outward and inward current pulses were increased in a step wise manner from 1 to 4 in **B—D**

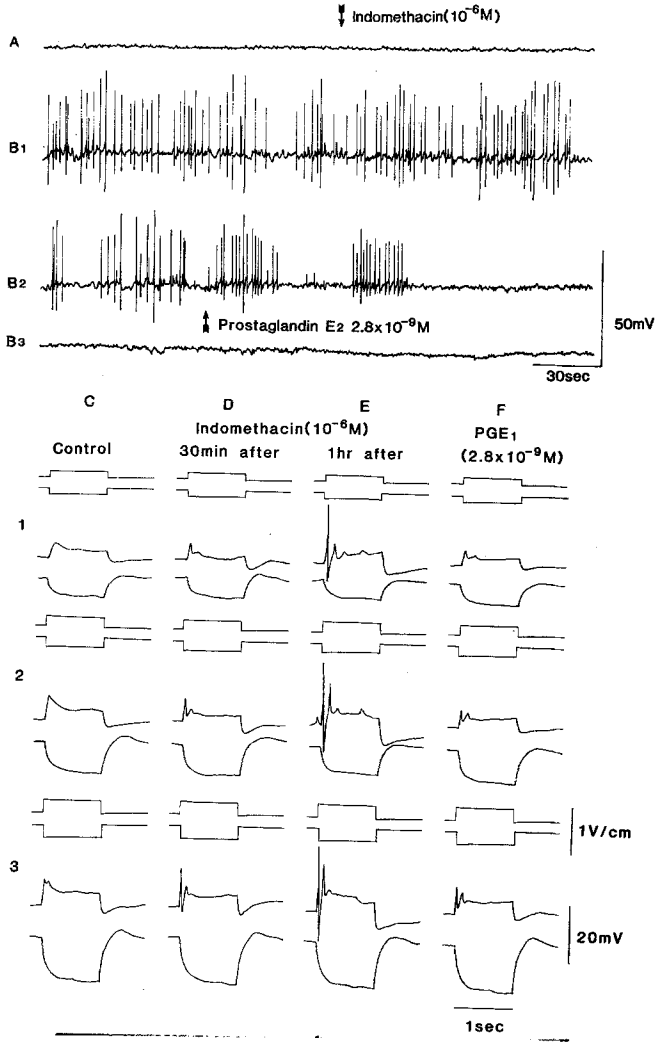

Fig. 6. Effects of indomethacin (10^{-6} M) on the membrane properties of the smooth muscle cell (**A—B**) and effects of indomethacin (10^{-6} M) or PGE_1 (2.8×10^{-9} M) on local responses, action potentials and electrotonic potentials evoked by the outward and inward current pulses (**C—F**). (**A**) Spontaneous spike activities were not observed in this muscle cell, and at the arrow indomethacin (10^{-6} M) was applied. (B_1) about 30 min after the application of indomethacin, spike activity occurred. (B_2 and B_3) Indomethacin-induced spike activities were suppressed by PGE_2 (2.8×10^{-9} M). (A_1 and B_1) and (B_2 and B_3) are recordings from the same cells. (**C**) Control; (**D** and **E**) effects of indomethacin; (**F**) in the presence of indomethacin, effects of PGE_1 on the action potential were observed. Intensities of outward and inward current pulses were increased in a step-wise manner from 1 to 3 in **C—F**

increased with time, and about 1 h after the treatment began, "all or none" action potentials were produced when outward current pulses were applied. The amplitude and the maximum rate of rise of the action potential was 25 mV and 4.0 V/s, respectively. In the presence of indomethacin, exogenous PGE_1 (2.8×10^{-9} M) suppressed the generation of action potentials (Fig. 6F), and only abortive action potentials were evoked.

Every attempt was made to keep the microelectrode in the same cell throughout these experiments, and Table 1 summarizes the results obtained from successful experiments on 2 muscle cells with and 7 without spontaneous spike discharges. Indomethacin increased the maximum rate of rise of the evoked potential from 0.5 ± 0.5 V/s ($\pm$SD, $n = 7$) to 2.6 ± 1.3 V/s ($\pm$SD, $n = 7$) in electrically quiescent cells, but did not affect this parameter in electrically active smooth muscle cells.

PGE and PGF Concentration in Muscle Tissue of Ileocecal Junction Taken from Control and Indomethacin-Treated Guinea-Pigs

As indomethacin reduced the threshold depolarization required for generation of action potential and evoked spontaneous action potentials in vitro experiments and as these effects of indomethacin were reversed in the presence of low

Table 1. Effects of indomethacin on the max. rate of rise (V/s) of action potentials evoked by the constant outward current pulses. In the smooth muscle cells listed as 1–7, "all or none" action potentials were not observed in the control Krebs' solution, whereas in cells, 8 and 9, "all or none" action potential was elicited by the extracellularly applied outward current pulses. The lower two rows contain measurements made on cells in which the microelectrode had been left in situ for at least 1 h after the application of indomethacin. Data were obtained from 9 preparations

Cell	1	2	3	4	5	6	7	8	9
Control	0.1	0.4	0.9	1.7	0.3	0.2	0.2	7.6	6.8
IND									
15 min after	0.2	2.4	0.9	2.6	1.1	1.1	0.9	7.6	6.8
60 min after	1.0	4.2	2.6	3.3	1.3	4.0	1.5	7.6	6.8

IND.: Indomethacin (10^{-5} M)

concentrations of PGs, we were particularly interested to analyse the PG content and electrical or mechanical properties of tissue taken from guinea-pigs which had been given indomethacin before experiment. The three muscle preparations taken from indomethacin treated animals all showed spontaneous contractions (0.1–0.4 c/min), whereas such activity was observed in only 10% of the muscle strips taken from the 10 control animals. Furthermore, in the spontaneously active preparations, regular bursts of short trains of spike alternating with silent periods were recorded in almost all the cells examined.

The concentrations of PGE in the tissues taken from control animals ranged from 80–128 ng/g wet wt. tissue with a mean value of 93 ± 20 ($\pm$SD, $n = 5$) ng/g wet wt. tissue. The range of PGE content in the tissues taken from indomethacin-treated animals was 43–73 ng/g wet wt. tissue with a mean value was 55 ± 13 ($\pm$SD, $n = 4$) ng/g wet wt. tissue. Thus indomethacin reduced the contents of PGE to about 60% of the control value.

The concentration of PGF in the control tissue ranged from 23–58 ng/g wet wt. tissue [the mean value was 32 ± 15 ($\pm$SD, $n = 5$)], but PGF was not detectable in tissues taken from any of the 4 indomethacin treated animals.

Discussion

The present results can be summarized as follows; i) PGE_1 relaxed the muscle tissue of the guinea-pig ileocecal junction, whereas both PGE_2 and $PGF_{2\alpha}$ evoked phasic contraction followed by relaxation; ii) all PGs tested suppressed both the spontaneous spike discharges and the mechanical activity of the smooth muscle cells, and raised the threshold for the generation of action potentials; iii) in electrically and mechanically quiescent muscle cells, indomethacin reduced the threshold for action potentials, and evoked spontaneous spike discharges and contractions; iv) spontaneous electrical and mechanical activity evoked by treatment with indomethacin was suppressed by low concentrations of PGs.

It has been reported that PGE_1 and PGE_2 contract the intestinal longitudinal muscle but relax the circular muscle, while $PGF_{2\alpha}$ contracts both types of smooth muscle cells in vitro (Bennett and Fleshler 1970). In the present experiment the actions of PGs on the contractile properties of the guinea-pig ileocecal junction were rather complex, i.e. PGE_1 simply relaxed the muscle while both PGE_2 and $PGF_{2\alpha}$ evoked biphasic changes in muscle tone. We also found that all the PGs tested suppressed spontaneous spike discharges. However while both $PGF_{2\alpha}$ (10^{-9} M) and PGE_2 (10^{-8} M) hyperpolarized the membrane, only PGE_2 produced significant changes in membrane resistance, whereas PGE_1 had no significant effect on the membrane potential. These results clearly show that the biphasic mechanical responses of the smooth muscle cells to PGs do not correlate with the effects of the PGs on the membrane potential of the smooth muscle cells. PGs, therefore, apparently have two distinct actions, one on the muscle tone and the other on the threshold potential at which an action potential is generated.

Kuriyama and Suzuki (1975) reported that PGE_1 suppressed the membrane activity evoked by outward current pulses in the circular muscle cells of the guinea-pig ileum. Our present results show that both the PGE series and $PGF_{2\alpha}$ raise the threshold depolarization for action potential and thereby suppress spontaneous action potentials in the guinea-pig ileocecal junction. The precise mechanisms involved in the inhibitory actions of PGs on electrical excitability of the smooth muscle membrane remain unknown. However, it has been reported that PGs interact with extracellular Ca^{2+} at activated nerve terminals, thereby reducing the amount of transmitter released by adrenergic or cholinergic nerve fibres (Ito and Tajima 1979, 1981a; Kuriyama and Makita 1982). Such an interaction between PGs and extracellular Ca^{2+} may explain the effects of PGs on the threshold for evoked action potentials and on spontaneously generated action potentials of the smooth muscle cells which we have observed in the present experiments.

In order to establish a physiological role for endogenous PGs, it is generally considered important to show the effects of a prostaglandin synthesis inhibitor on a physiological response. In the dog trachea, the inhibitory action of endogenous PGs on the amplitude of e.j.p. evoked by excitation of cholinergic nerve fibres was abolished by indomethacin, in both in vitro and in vivo experiments (Ito and Tajima 1981a, b), indicating that endogenous PGs play a physiological role in regulating the transmitter release from cholinergic nerve terminals.

In the present experiments, indomethacin whether administered in vivo or in vitro induced spontaneous spike activity in the electrically quiescent smooth muscle cells, and when administered in vivo indomethacin reduced the PGE and PGF contents in the muscle tissue to 50 and 0% of the control value respectively. Moreover the application of exogenous PGs suppressed not only spontaneous spike discharges observed in electrically and mechanically active smooth muscle cells but also electrical activity uncovered in the presence of indomethacin. These observations suggest that PGs have inhibitory actions on the electrical excitability of the smooth muscle cells, and that indomethacin evokes spontaneous membrane activity of the smooth muscle cells by an inhibitory action on prostaglandin biosynthesis rather than by a direct action on the smooth muscle cells. It is already known that changes in prostaglandin synthesis in the cat ileum in vivo correlate with changes in motility of the ileum; indomethacin and eicosatetraenoic acid increase motility, and at the same time they reduce the intestinal concentration of prostaglandins (Sanders 1978; Sanders and Ross 1978).

The present observations of the effects of PGs and indomethacin on the contractile properties and electrical properties of the smooth muscle cells of guinea-pig ileocecal

junction therefore lend further support to the hypothesis that endogenous PGs play an important physiological role in regulating the motility of the intestine and possibly the ileocecal sphincter.

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