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Effect of Progesterone on Calcium Activated Potassium Currents and Intracellular Calcium in Guinea Pig Colon Myocytes

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SUMMARY

Aims: *To study the effects of progesterone on contractile activity of smooth muscle strips and on ion currents and intracellular Ca^{2+} ($[Ca^{2+}]i$) intensity in single colonic myocytes in guinea pig proximal colons*. Methods: *Strips and single cells were dissected from female guinea pig proximal colon. Contraction of strips through an isotonic transducer was assessed and the responsible currents to progesterone were recorded with EPC-9 amplifier in nystatin perforated whole-cell configuration. Detection of $[Ca^{2+}]i$ fluorescence loading fura-2 acetoxymethylester (fura-2/AM) was measured with confocal microscope*. Results: *Progesterone significantly inhibited contraction of guinea pig colon strips in a dose-dependent pattern. Inhibitory concentration 50 (IC_{50}) of progesterone in longitudinal strips and circular strips was, respectively, 9.7 μM and 1.0 nM. Iberiotoxin (IbTX) partially blocked inhibition of progesterone in both oriented smooth muscle strips. Ca^{2+} activated K^+ (K_{Ca}) channel currents recorded by depolarizing pulse protocol were enhanced by progesterone to 138% ± 13% ($n = 9$, $p < 0.01$), and to 143% ± 12% ($n = 8$, $p < 0.01$) when perfused with 10 μM onapristone. Progesterone reduced L-Ca^{2+} currents to 67% ± 6% ($n = 7$, $p < 0.01$) and had no effect with 5 μM nicardipine in bath solution. $[Ca^{2+}]i$ fluorescence was reduced by progesterone to 75% ± 12% ($n = 8$, $p < 0.01$)*. Conclusions: *Progesterone decreases the contraction of colonic smooth muscles by enhancing K_{Ca} currents and reducing Ca^{2+} influx*.

Key words: Ca^{2+} activated K^+ (K_{Ca}) channel - Colonic smooth muscle - Contraction - Intracellular calcium - Progesterone

INTRODUCTION

Irritable bowel syndrome (IBS) is a common digestive functional disease. A higher prevalence of IBS occurs in women, especially in the phases of pre- and postmenopause (1, 2). Altered bowel function and IBS-like gastrointestinal complaints are extremely exacerbated at mensal phase in clinic (3, 4). Most studies have demonstrated that progesterone, not estrogen, may be the hormone responsible (5).

It is well known that progesterone plays an important role in relaxation in myometrium (6, 7). In the gastrointestinal tract, progesterone decreased the contractile activity of the murine jejunum in a dose-dependent manner, which is not inhibited by onapristone, a specific blocker of the progesterone receptor, indicating the involvement of progesterone in a nongenomic pathway (8). In isolated segments of guinea pig small intestine, progesterone produced a prompt and concentration-related increase of the peristaltic pressure threshold at which propulsive muscle contractions elicited. The inhibition of the peristaltic motor caused by progesterone at micromolar concentrations suggests a depressant action on intestinal muscle activity (9).

Several studies about the effects of progesterone on colon smooth muscle suggest that the female sex steroid hormones, including progesterone, can affect the myoelectric and mechanical activity of smooth muscle *in vitro* (10). By using the geometric center method of analysis, used earlier on adult male rats' colonic transit *in vivo* (11), pretreatment of ovariectomized rats with estrogen and progesterone was found to result in a significant decrease in the colonic transit, compared with that of the untreated ovariectomized rats. Gill *et al*. (12) reported that progesterone reduced the contractility in both circular and longitudinal strips and the contractile frequency of the longitudinally orientated strips in a dose-dependent manner, in human colon. Bielefeldt *et al*. (13) showed that progesterone caused relaxation of human colonic smooth muscles, by inhibiting Ca^{2+} entry from extracellular space but not Ca^{2+} release from intracellular stores. And this suppression, in rat portal vein, was demonstrated through the activation of the conductance of Ca^{2+} activated K^+ (K_{Ca}) channel (14). But the effects of progesterone on K_{Ca} channels and Ca^{2+} concentration change in single colonic myocytes remain unknown.