American Journal of Obstetrics and Gynecology (2006) **194**, 1384–90

ELSEVIER

American Journal of Obstetrics & Gynecology

www.ajog.org

Effect of magnesium sulfate on contractile force and intracellular calcium concentration in pregnant human myometrium

Victor P. Fomin, PhD,[a,b] Shawn G. Gibbs, PhD,[c,d] Ram Vanam, BS,[e] Akira Morimiya, BS,[f] William W. Hurd, MD[c,*]

Department of Obstetrics and Gynecology, Indiana University School of Medicine, Indianapolis, IN[a]; Department of Biological Sciences, University of Delaware, Newark, DE[b]; Department of Obstetrics and Gynecology, Wright State University Boonshoff School of Medicine, Dayton, OH[c]; School of Public Health, University of Texas at Houston, El Paso, TX[d]; Departments of Biochemistry[e] and Pathology,[f] Indiana University School of Medicine, Indianapolis, IN

Received for publication July 25, 2005; revised October 28, 2005; accepted November 28, 2005

KEY WORDS
Magnesium sulfate
Oxytocin
Calcium
Myometrium
Uterus
Premature labor
Pregnancy

Objective: This study was undertaken to evaluate the effects of magnesium sulfate ($MgSO_4$) on contractile force and increases in free intracellular calcium concentration ($[Ca^{2+}]_i$) in human myometrial strips from pregnant women.

Study design: Simultaneous measurements of isometric tension and $[Ca^{2+}]_i$ were measured in myometrial strips obtained at the time of cesarean delivery from pregnant nonlaboring women at term with the use of a fluorescence spectrometer equipped with a displacement force transducer. Changes in $[Ca^{2+}]_i$ were measured with fura-2, a Ca^{2+}-sensitive fluorescent probe. Myometrial strips were exposed to $MgSO_4$ (5 or 10 mmol/L) for 5, 10, 20, and 30 minutes and observed for spontaneous contractions or stimulated with either oxytocin (OT; 0.1 μmol/L) or potassium chloride (KCl; 90 mmol/L).

Results: $MgSO_4$ reduced spontaneous, OT, and KCl-evoked contractions and increases in $[Ca^{2+}]_i$ in a time and concentration-dependent manner. After 20 minutes exposure to 5 mmol/L $MgSO_4$, the OT-elicited changes in contractile response and $[Ca^{2+}]_i$ were significantly decreased. $MgSO_4$ did not change [Ca2+]i/force relationship of the responses to OT or KCl, or during spontaneous activity.

Conclusion: At a pharmacologic concentration (5 mmol/L), $MgSO_4$ inhibits contractile response and [Ca2+]i in pregnant human myometrial strips by a pattern that is consistent with both extra- and intracellular mechanisms. At a suprapharmacologic concentration (10 mmol/L), the more immediate effect of $MgSO_4$ is consistent with an extracellular mechanism. $MgSO_4$ does not appear to interfere at the level of the calcium-calmodulin interface, since the [Ca2+]i/force relationship was not changed.

© 2006 Mosby, Inc. All rights reserved.

Supported by the NIH/NICHD grant R01 HD36692, and by United Cerebral Palsy Foundation grant R-733-04.

* Reprint requests: William W. Hurd, MD, 128 East Apple St, CHE 3800, Dayton, OH 45409-2793.
E-mail: william.hurd@wright.edu

0002-9378/$ - see front matter © 2006 Mosby, Inc. All rights reserved.
doi:10.1016/j.ajog.2005.11.045

Magnesium sulfate ($MgSO_4$) remains one of the most widely used drugs to inhibit premature labor, primarily because of its initial effectiveness, lack of tachyphylaxis, and minimal side effects.[1] The ability of $MgSO_4$ to inhibit uterine contractility both in vivo and in vitro has been appreciated for over 40 years.[2,3] Therapeutically, serum concentrations of $MgSO_4$ required for tocolysis are typically 4 to 6 mEq/L (2-5 mmol/L), which exceeds its physiologic serum concentration by several fold.[4] The magnesium cation (Mg^{2+}) is believed to be responsible for the tocolytic effect of $MgSO_4$ because, in vitro, magnesium chloride ($MgCl_2$) has similar effects. However, the cellular mechanisms by which $MgSO_4$ inhibits uterine contractility are only partially understood.

The mechanisms and time required for pharmacologic concentrations of $MgSO_4$ to inhibit myometrial contractility remains in question. In rat myometrium, $MgSO_4$ inhibits both oxytocin (OT)-mediated force and calcium influx within minutes by an effect counteracted by a calcium channel antagonist, suggesting that Mg^{2+} blocks Ca^{2+} channels by an extracellular mechanism.[5] However, in the absence of extracellular Ca^{2+}, $MgSO_4$ inhibits intracellular Ca^{2+} release in response to OT, suggesting intracellular effects as well. In nonpregnant human myometrium, 12 to 14 mmol/L $MgSO_4$ inhibits spontaneous contractions and Ca^{2+} uptake within minutes, again suggesting an extracellular mechanism.[6] However, in primary cultures of pregnant human myometrial cells, it takes more than 20 minutes for 10 mmol/L $MgSO_4$ to inhibit OT-mediated calcium influx, suggesting an intracellular mechanism, because it takes more than 20 minutes for free intracellular magnesium ($[Mg^{2+}]_i$) to increase to more than 150% of baseline.[7]

We designed this study to determine whether the immediate and delayed effects of $MgSO_4$ on myometrial contraction are concentration dependent. We also wanted to determine if $MgSO_4$ exposure altered the $[Ca^{2+}]_i$/force relationship, because such a change would suggest that $[Mg^{2+}]_i$ interferes with the ability of increased $[Ca^{2+}]_i$ to elicit a contraction.

We thus evaluated the time course required for exposure to $MgSO_4$ to decrease myometrial contractile activity and free intracellular calcium concentration ($[Ca^{2+}]_i$) in pregnant human myometrial strips during spontaneous contractions and after stimulation with OT or KCl. We evaluated 2 concentrations of $MgSO_4$: 5 mmol/L (designated pharmacologic), and 10 mmol/L (designated suprapharmacologic). We evaluated both maximal response and the initial rising phase of the response for both $[Ca^{2+}]_i$ and contractile force because this rising phase has been shown to at least partially reflect calcium influx. A delay in the effects of $MgSO_4$ would imply an intracellular effect, because it takes at least 20 minutes for $[Mg^{2+}]_i$ to maximally increase. Finally, we evaluated the $[Ca^{2+}]_i$/force relationship to determine whether $[Mg^{2+}]_i$ interferes with the ability of calcium to elicit contractile force.

Material and methods

Tissue acquisition and preparation

Pregnant human uterine tissue was obtained after written consent from nonlaboring women at term (38-40 weeks) undergoing cesarean sections using a protocol approved by the Indiana University, Miami Valley Hospital, and Wright State University Committees on Human Use in Research. Women who had any major complications of pregnancy (including hypertension, diabetes, and premature labor), or who had been treated with any medications other than prenatal vitamins before delivery, were excluded. At the time of cesarean section, a full thickness strip of uterine tissue was taken from the upper margin of a lower transverse incision. The tissue was transported on ice in a sterile Hanks balanced salt solution (HBSS) to the laboratory.

The uterine tissue was placed into physiologic salt solution (PSS) of the following composition: NaCl (120.5 mmol/L), KCl (4.8 mmol/L), $MgSO_4$ (1.2 mmol/L), NaH_2PO_4 (1.2 mmol/L), $NaHCO_3$ (20.4 mmol/L), $CaCl_2$ (1.6 mmol/L), D-glucose (10 mmol/L), and pyruvate (1 mmol/L). The solution was aerated with 95% oxygen (O_2) and 5% carbon dioxide (CO_2), pH 7.4 at 37°C. Myometrial tissue was cleaned of endometrium and serosa and cut into 10 × 1 × 1 mm strips (50-75 mg, wet weight). The strips were dissected (10 × 1 × 1 mm), stretched (1.2× slack length) in a small dark chamber, and incubated with PSS supplemented with fura-2/acetoxymethyl ester (fura-2/AM; 20 μmol/L), dissolved in 2.5% dimethyl sulfoxide, 0.05% Pluronic 127, and neostigmine (200 μmol/L) for 4 hours at room temperature.[8] After the incubation, the tissue strips were rinsed with fresh PSS for 45 minutes to remove extracellular fura-2 and byproducts of fura-2/AM de-esterification.

Simultaneous contractile force and $[Ca^{2+}]_i$ measurement techniques

Fura-2 fluorescence and muscle isometric tension were simultaneously measured by using a C-44 Ratio Fluorescence Spectrometer equipped with a FSG-01 displacement force transducer (Photon Technology International, Lawrenceville, NJ). After fura-2 loading, each myometrial strip was transferred to a 1-cm acryl cuvette filled with 2.5-mL PSS and mounted isometrically to a tension transducer inside the spectrometer sample compartment. The PSS solution was equilibrated at a constant rate with 95% O_2 and 5% CO_2, adjusted so that the strip was perfused at room temperature. The strips were stretched to 2 g tension for 1 hour, followed by the decrease of

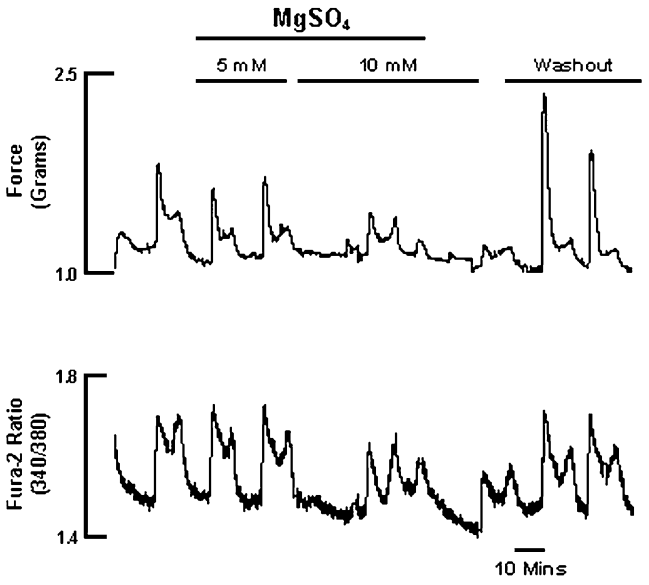

Figure 1 The effect of $MgSO_4$ on contractile force and $[Ca^{2+}]_i$ in response to oxytocin (OT; 0.1 μmol/L). The simultaneously recorded tracings are from a single representative myometrial strip obtained from a pregnant woman. The *solid bars* indicate the period in which $MgSO_4$ was applied at the indicated concentrations.

tension down to 0.8 g. Force was quantified by measuring the area under the curve in unit time.

Simultaneously, the strips were alternatively illuminated with 340 ± 5 nm and 380 ± 5 nm by using Delta Ram excitation monochromator (Photon Technologies International). The 340/380 fluorescence ratio (R340/380) and force data were recorded with the use of a desktop computer at a 1-second acquisition rate using Felix software (Photon Technology International).

Kinetic analysis of contractile and $[Ca^{2+}]_i$ responses was performed by examining the peak amplitudes of the contractile and $[Ca^{2+}]_i$ responses and the rates of initial (rising) phase of the responses. The $[Ca^{2+}]_i$/force relationship of the responses was analyzed by measuring the difference between the peak amplitude and the basal level of both force and $[Ca^{2+}]_i$, normalizing the difference to unity and plotting the normalized data points in $[Ca^{2+}]_i$ versus force coordinates.[9]

Experimental approach

Strips were exposed to $MgSO_4$ (5 or 10 mmol/L) for 5, 10, 20, or 30 minutes and then observed for spontaneous contractions or stimulated with either the receptor agonist OT (0.1 μmol/L) or KCl (90 mmol/L), which stimulates calcium influx by membrane depolarization. Control strips were not incubated with $MgSO_4$. The strips from 5 patients of similar gestational age (38-40 weeks) were used for each experiment.

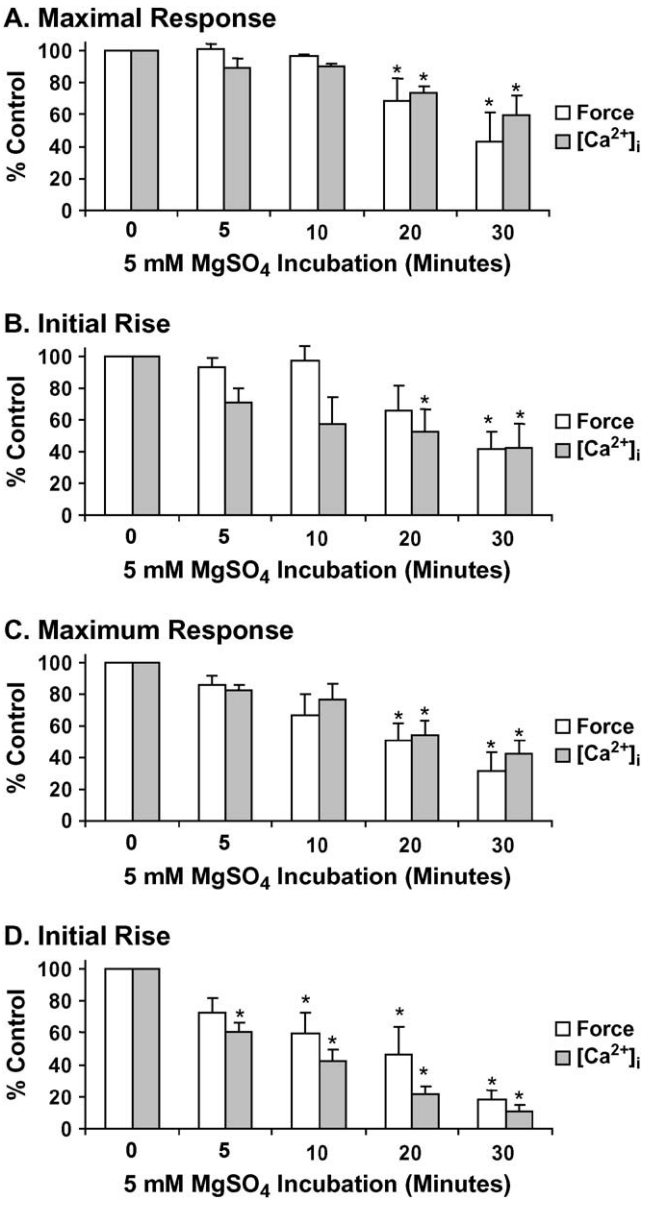

Figure 2 The effect of $MgSO_4$ on contractile force and $[Ca^{2+}]_i$ in response to oxytocin (OT; 0.1 μmol/L) in pregnant human myometrial strips. Individual myometrial strips were preincubated with 5 mmol/L (**A** and **B**) or 10 mmol/L (**C** and **D**) $MgSO_4$ for 5 to 30 min and stimulated with 0.1 μmol/L OT. The histograms represent the mean ± SEM responses in myometrial strips obtained from 5 women. *Asterisk* indicates $P < .05$ compared with control strips not treated with $MgSO_4$.

Statistical analysis

Contractile force and $[Ca^{2+}]_i$ are presented as mean ± SEM. Treatment groups were compared by 1-factor analysis of variance (ANOVA), followed by Fisher Protected Least Significant Difference test by using StatView software (Abacus Concepts, Inc, Berkeley, CA). *P* values less than .05 were considered significant.

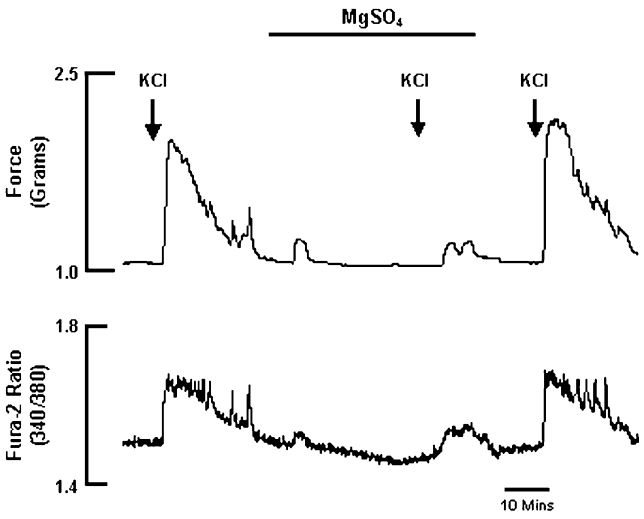

Figure 3 The effect of $MgSO_4$ on contractile force and $[Ca^{2+}]_i$ in response to potassium chloride (KCl; 60 mmol/L). The simultaneously recorded tracings are from a single representative myometrial strip obtained from a pregnant woman. The time of KCl addition is indicated by *arrows* and the time and duration of the $MgSO_4$ exposure is indicated by the *bar*. The time of KCl addition is indicated by *arrows* and the *solid bar* indicates the period in which $MgSO_4$ (5 mmol/L) was applied.

Materials

Fura-2/acetoxymethyl ester and Pluronic 127 were purchased from Molecular Probes, Inc, Eugene, OR. Oxytocin, KCl, neostigmine, and dimethyl sulfoxide were purchased from Sigma Chemical Co, St. Louis, MO.

Results

OT (0.1 μmol/L) increased both contractile force and $[Ca^{2+}]_i$ in human myometrial strips (Figure 1). Exposure to 5 mmol/L $MgSO_4$ for 20 minutes or more reduced the maximal response to OT in terms of both contractile force and $[Ca^{2+}]_i$ (Figures 1 and 2). The initial rise of these responses was similarly reduced by 5 mmol/L $MgSO_4$, except that the reduction in initial rise of contractile force did not reach significance until 30 minutes of $MgSO_4$ exposure. At 10 mmol/L concentration, $MgSO_4$ reduced the initial rise of $[Ca^{2+}]_i$ after 5 minutes or more exposure. After decreasing the Mg^{2+} in the media back to physiologic levels (1.2 mmol/L), the OT-responses were restored within 10 minutes (Figure 1).

Depolarization with KCl (60 mmol/L) stimulated both contractions and associated increases in $[Ca^{2+}]_i$ in myometrial strips (Figure 3). Exposure to 5 mmol/L $MgSO_4$ for 10 minutes or more reduced the KCl-evoked maximal response and initial rise in terms of both contractile force and $[Ca^{2+}]_i$ (Figures 3 and 4). The

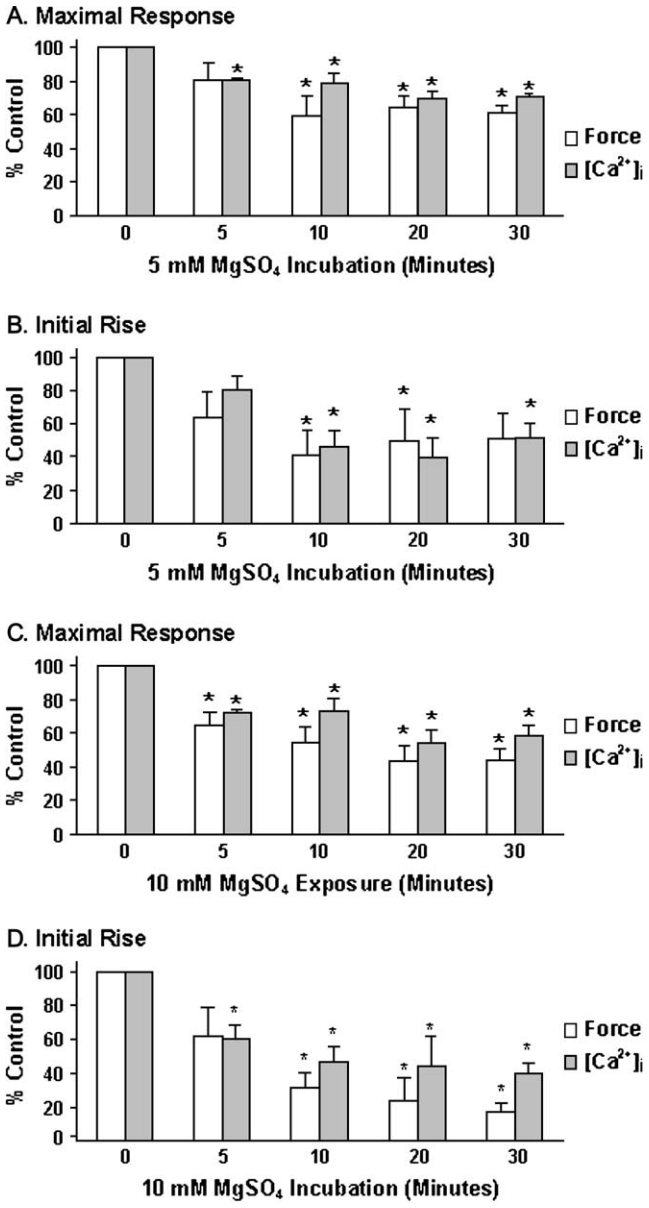

Figure 4 The effect of $MgSO_4$ on contractile force and $[Ca^{2+}]_i$ in response to KCl in pregnant human myometrial strips. Individual myometrial strips were preincubated with 5 mmol/L (**A** and **B**) or 10 mmol/L (**C** and **D**) $MgSO_4$ for 5 to 30 min and stimulated with 60 mmol/L KCl. The histograms represent the mean ± SEM responses of myometrial strips obtained from 5 women. *Asterisk* indicates $P < .05$ compared with control strips not treated with $MgSO_4$.

reduction in maximal $[Ca^{2+}]_i$ was significant after 5 minutes of $MgSO_4$ exposure. Exposure to 10 mmol/L $MgSO_4$ for 5 minutes or more reduced the KCl-evoked maximal response and initial rise in terms of both contractile force and $[Ca^{2+}]_i$, except for initial rise of force was not significantly reduced until 10 minutes. Again, after decreasing media Mg^{2+} back to physiologic levels, the KCl-responses were restored within 10 minutes (Figure 1).

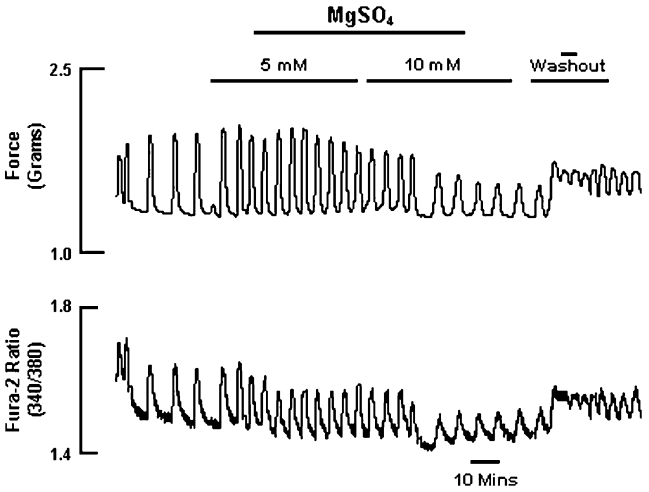

Figure 5 The effect of $MgSO_4$ on spontaneous contractile force and $[Ca^{2+}]_i$ oscillations in pregnant human myometrial strips. The simultaneously recorded tracings are from a single representative strip. The *solid bars* indicate the period in which $MgSO_4$ was applied at the indicated concentrations.

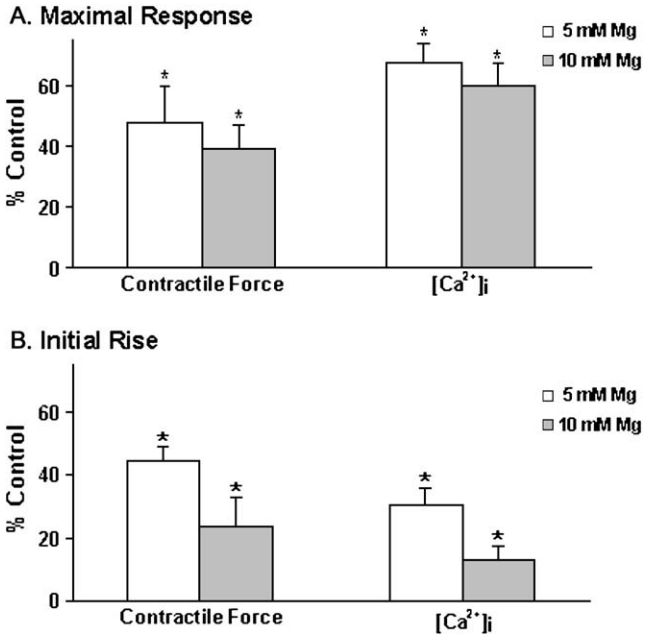

Figure 6 The effect of $MgSO_4$ on spontaneous contractile force and $[Ca^{2+}]_i$ oscillations in pregnant human myometrial strips. The effect of 5 mmol/L and 10 mmol/L $MgSO_4$ was determined for (**A**) maximal response and (**B**) initial rise of contractile force and $[Ca^{2+}]_i$ during spontaneous contractions. The histograms represent the means ± SEM responses of myometrial strips obtained from 5 women. *Asterisk* indicates $P < .05$ compared with control strips not treated with $MgSO_4$.

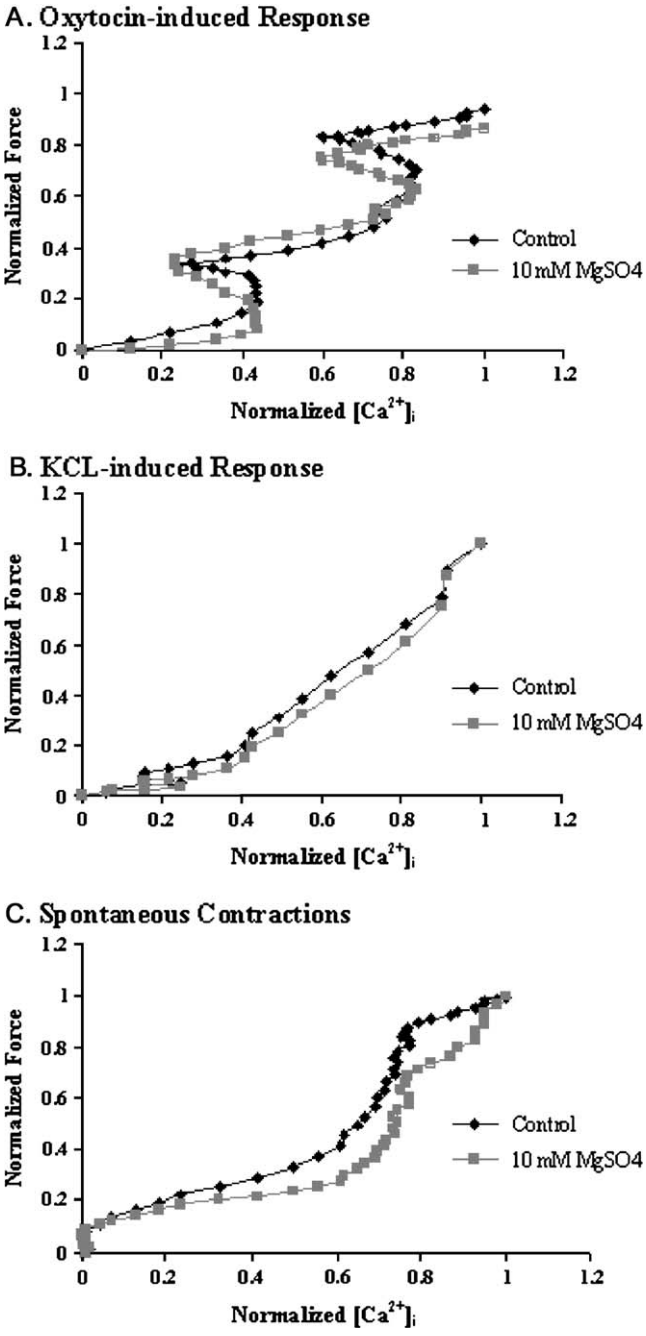

Figure 7 Effect of 10 mmol/L $MgSO_4$ exposure on $[Ca^{2+}]_i$-force relationship in pregnant human myometrial strips during (**A**) OT-induced contractions, (**B**) KCl-induced contractions, or (**C**) spontaneous contractions. For OT- and KCl-induced contractions the strips were exposed for 30 min, and for the spontaneous contractions the exposure time was 60 min. The rising phase of the contractile and $[Ca^{2+}]_i$ responses was normalized to the peak amplitude. Data are from individual strips obtained from 5 pregnant women.

Spontaneous contractions and $[Ca^{2+}]_i$ oscillations were inhibited by exposure to both 5 mmol/L and 10 mmol/L $MgSO_4$ in pregnant human myometrium (Figures 5 and 6). $MgSO_4$ gradually decreased the amplitude, frequency, and rising phases. The effect of 5 mmol/L $MgSO_4$ appeared to be gradual, whereas the effect of 10 mmol/L was more immediate. However, a time course for these effects could not be determined because of the variable frequency of spontaneous

contractions in human strips, which varied from 1 to 20 times per hour (Figure 5). To combine data from several strips, it was necessary to quantify contractions over 1 hour, making it impossible to measure the effects of preincubation periods of 5 to 30 minutes. These spontaneous contractions and changes in $[Ca^{2+}]_i$ returned to normal within 10 minutes after decreasing media Mg^{2+} back to physiologic levels.

Exposure to 10 mmol/L $MgSO_4$ for 30 minutes did not alter the $[Ca^{2+}]_i$/force relationship during spontaneous contractions and $[Ca^{2+}]_i$ oscillations or during responses evolved by OT or KCl (Figure 7).

Comment

We examined the effect of $MgSO_4$ on both OT and KCl-induced contractions because these substances evoke myometrial contractions through different mechanisms. OT stimulates contractions by at least 2 receptor-mediated mechanisms: (1) a second messenger system involving phospholipase C (PLC), which results in release of calcium from intracellular stores; and (2) the opening of calcium channels with the resultant calcium influx.[10] In contrast, depolarizing concentrations of KCl result in myometrial contractions primarily by Ca^{2+} influx via voltage-sensitive L-type Ca^{2+} channels.[11]

In the current study, $MgSO_4$ at both pharmacologic (5 mmol/L) and suprapharmacologic (10 mmol/L) concentrations inhibited the OT-mediated changes in $[Ca^{2+}]_i$ and contractile activity. However, the effects of the pharmacologic concentration did not reach significance until 20 minutes of $MgSO_4$ exposure (Figures 1 and 2). This is evidence that at least part of the effect of $MgSO_4$ is intracellular, because an extracellular effect would be expected to manifest immediately. The finding that $MgSO_4$ inhibits both initial rising phases of both the $[Ca^{2+}]_i$ and contractile responses suggests that $MgSO_4$ decreases both calcium influx and intracellular calcium release. The ability of the suprapharmacologic concentration of $MgSO_4$ to inhibit the OT-induced initial rise of $[Ca^{2+}]_i$ within 5 minutes is further evidence of an extracellular effect of $MgSO_4$ on calcium influx.

We also found that $MgSO_4$ inhibited KCl-induced increases in $[Ca^{2+}]_i$, after only 5 minutes incubation (Figures 3 and 4). The immediacy of this effect is additional evidence that a portion of the effect of $MgSO_4$ on myometrial contractility is extracellular, and apparently includes a blocking effect on L-type Ca^{2+} channels. The somewhat delayed effect of pharmacologic concentrations of $MgSO_4$ (5 mmol/L) suggests that at this lower concentration, an intracellular effect on calcium channels is operational.

Both concentrations of $MgSO_4$ also inhibited spontaneous contractions and the rise in $[Ca^{2+}]_i$ in terms of initial rise and maximal response (Figures 5 and 6). Inhibition in the presence of 5 mmol/L $MgSO_4$ appeared to be gradual, whereas in the presence of 10 mmol/L inhibition appeared to be more immediate, consistent with what we observed in the OT- and KCl-evoked responses.

It was unanticipated that the OT response would return after only a 10-minute washout period when we replaced the elevated Mg^{2+} concentrations in the contraction media (5-10 mmol/L) with a physiologic concentration (1.2 mmol/L). Our assumption is that the mechanisms that stringently regulate $[Mg^{2+}]_i$ (ie, membrane Na^+-Mg^{2+} exchangers and intracellular buffers) quickly return the intracellular concentration to the physiologic range once the extracellular Mg^{2+} concentration was returned to the physiologic range.

Our findings are consistent with the hypothesis that $MgSO_4$ inhibits uterine contractions by both extracellular and intracellular mechanisms. By using a ^{45}Ca method, it was demonstrated that supraphamacologic concentrations of extracellular $MgSO_4$ (12-14 mmol/L) block calcium channels.[6] Subsequently, it was shown that the maximal suppressive effect of $MgSO_4$ of OT-induced Ca^{2+} influx across the cell membrane did not occur until after 20 minutes of $MgSO_4$ exposure, which was shown to reflect the time it takes for $[Mg^{2+}]_i$ to maximally rise.[7] It was hypothesized that Mg^{2+} blocks calcium channels by an intracellular mechanism. This is consistent with the more recent finding in cardiac cells that increased $[Mg^{2+}]_i$ blocks L-type Ca^{2+} channels.[12]

Our laboratory has previously demonstrated other intracellular effects of Mg^{2+} that could inhibit myometrial contractility. In human myometrial cells, $MgSO_4$ exposure for more than 20 minutes inhibits IP_3 production and activates protein kinase C, either of which could inhibit contractility.[13,14] Another possibility is that increased $[Mg^{2+}]_i$ interferes with the binding of Ca^{2+} to calmodulin, a key component of the contractile cascade. However, the finding from the current study that $MgSO_4$ exposure does not result in dissociation of $[Ca2+]_i$/force relationship suggests that this latter possibility is unlikely (Figure 7).

The multiphasic response of the $[Ca^{2+}]_i$/force relationship to OT (Figure 7, *A*) deserves some discussion. This pattern was seen in the majority of cells stimulated with OT in our experiments. The response differs from those seen after KCl stimulation or during spontaneous contractions. This appears to be a reflection of the complex nature of the OT-mediated contractile response. Whenever $[Ca^{2+}]_i$ increases faster than contractile force, the curve is deflected towards the *x*-axis. We assume that the first deflection is related to calcium influx through activation of calcium membrane channels, and the second deflection is related to a secondary release of Ca^{2+} from intracellular stores. Whenever the contractile force increases faster than $[Ca^{2+}]_i$, the curve is deflected toward the *y*-axis. We assume that this is related to the Ca^{2+}-independent aspect of the myometrial contractile response.

Understanding the mechanisms by which $MgSO_4$ inhibits uterine contractility may lead to the development of more effective tocolytics in the future. Tocolytics used today, including $MgSO_4$, beta adrenergic agonists, and prostaglandin synthase inhibitors, immediately decrease or stop uterine contractility in the majority of patients presenting in premature labor. Unfortunately, the effectiveness of these drugs in preventing preterm birth is limited, in part because of their narrow therapeutic ranges and harmful side effects. Drugs that specifically target $MgSO_4$-sensitive elements of the myometrial contractile cascade (ie, phospholipase C and protein kinase C) could be investigated for possible use as tocolytics of the future.

We conclude that $MgSO_4$ inhibits myometrial contractility by a combination of extracellular and intracellular mechanisms; and that $MgSO_4$ exposure does not interfere with the binding of Ca^{2+} to calmodulin. This information is important in our developing knowledge of the mechanisms by which $MgSO_4$ inhibits uterine contractility, and may help guide the development of more effective tocolytics in the future.

References

1. Gordon MC, Iams JD. Magnesium sulfate. Clin Obstet Gynecol 1995;38:706-12.
2. Hall DG, McGaughey HS, Corey EL, Thornton WN. The effects of magnesium therapy in the duration of labor. Am J Obstet Gynecol 1959;78:27-32.
3. Kumar D, Zourlas PA, Barnes AC. In vitro and in vivo effects of magnesium sulfate on human uterine contractility. Am J Obstet Gynecol 1963;86:1036-40.
4. Cunningham FG, MacDonald PC, Gant NF, Leveno KJ, Gilstrap LC III. Preterm and postterm pregnancy and fetal growth retardation. In: Williams obstetrics. 19th ed. Norwalk (CT): Appleton & Lange; 1993. p. 853-89.
5. Phillippe M. Cellular mechanisms underlying magnesium sulfate inhibition of phasic myometrial contractions. Biochem Biophys Res Communs 1998;252:502-7.
6. Popper LD, Batra SC, Akerlund M. The effect of magnesium on calcium uptake and contractility in the human myometrium. Gynecol Obstet Invest 1989;28:78-81.
7. Mizuki J, Tasaka K, Masumoto N, Kasahara K, Miyake A, Tanizawa O. Magnesium sulfate inhibits oxytocin-induced calcium mobilization in human puerperal myometrial cells: possible involvement of intracellular free magnesium concentration. Am J Obstet Gynecol 1993;169:134-9.
8. Word RA, Tang D, Kamm KE. Activation properties of myosin light chain kinase during contraction/relaxation cycles of tonic and phasic smooth muscles. J Biol Chem 1994;269:21596-602.
9. Burdyga TV, Wray S. The relationship between the action potential, intracellular calcium and force in intact phasic, guinea-pig uretic smooth muscle. J Physiol 1999;520:867-83.
10. Zhuge R, Li S, Chen TH, Hsu WH. Oxytocin induced a biphasic increase in the intracellular Ca^{2+} concentration of porcine myometrial cells: participation of a pertussis toxin-insensitive G-protein, inositol 1,4,5-trisphosphate-sensitive Ca^{2+} pool, and Ca^{2+} channels. Mol Reprod Dev 1995;41:20-8.
11. Trujillo MM, Ausina P, Savineau JP, Marthan R, Strippoli G, Advenier C, et al. Cellular mechanisms involved in iso-osmotic high K+ solutions-induced contraction of the estrogen-primed rat myometrium. Life Sci 2000;66:2441-53.
12. Yamaoka K, Kameyama M. Regulation of L-type Ca^{2+} channels in the heart: overview of recent advances. Mol Cell Biochem 2003; 253:3-13.
13. Hurd WW, Natarajan V, Fisher JR, Singh DM, Gibbs SG, Fomin VP. Magnesium sulfate inhibits the oxytocin-induced production of inositol 1,4,5-trisphosphate in cultured human myometrial cells. Am J Obstet Gynecol 2002;187:419-24.
14. Bermeo ME, Fomin VP, Ventolini G, Gibbs SG, McKenna DS, Hurd WW. Magnesium sulfate induces translocation of protein kinase C isoenzymes alpha and delta in myometrial cells from pregnant women. Am J Obstet Gynecol 2004;190:522-7.