

Pharmacokinetics of a novel oral slow-release form of misoprostol

C.Fiala^{1,4,5}, A.Aronsson¹, F.Granath², O.Stephansson², H.W.Seyberth³, B.Watzer³
and K.Gemzell-Danielsson¹

¹Department of Woman and Child Health, Division for Obstetrics and Gynaecology, ²Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, S-171 76 Stockholm, Sweden, ³Department of Pediatrics, Philipps University Marburg, Deutschhausstr, Germany and ⁴Gynmed Ambulatorium, 1150 Vienna, Austria

⁵To whom correspondence should be addressed. E-mail: christian.fiala@aon.at

BACKGROUND: The pharmacokinetics of a novel slow-release (SR) misoprostol was studied and compared to conventional misoprostol. **METHODS:** Thirty-one women, pregnant between 8 and 12 weeks, requesting surgical abortion were randomly allocated to receive orally 400 µg conventional misoprostol, 400 µg SR misoprostol or 800 µg SR misoprostol. Venous blood samples were taken at 0, 30, 60, 120, 240 and 360 min after the administration of misoprostol. Misoprostol acid (MPA) was determined in serum samples using liquid chromatography/tandem mass spectrometry. **RESULTS:** Serum peak concentration (C_{max}) was highest for conventional oral misoprostol. The time to peak concentration (T_{max}) was similar for all groups. The area under the curve up to 360 min was similar for conventional and for 800 µg SR misoprostol and significantly greater for these groups compared to 400 µg SR misoprostol ($P = 0.013$). **CONCLUSION:** The new SR form of misoprostol demonstrated lower peak levels but longer-lasting elevation in plasma levels compared to conventional oral misoprostol. The AUC for 800 µg SR misoprostol was similar to that of 400 µg of conventional oral misoprostol. SR misoprostol may offer an alternative to repeated administration of oral misoprostol or to vaginal administration.

Key words: induced abortion/misoprostol/oral/pharmacokinetics/slow release

Introduction

Misoprostol is a prostaglandin (PG) E₁ analogue. It was introduced for treatment of gastric ulcer under non-steroidal anti-inflammatory drugs (NSAID) and is approved for this indication in >85 countries under the brand name of Cytotec®. Beyond this it is used worldwide for a variety of indications in obstetrics and gynaecology. This use, however, is off-label in most cases because the current patent holder has never applied for approval for any indication in obstetrics and gynaecology (Weeks *et al.*, 2005, www.misoprostol.org). However, even the British National Formula recommends its off-label use for some indications. Misoprostol is available in tablets containing 100 or 200 µg and approved for oral use.

A drawback of the use of misoprostol in many situations in obstetrics and gynaecology is the very short terminal half-life of 20–40 min (Product information). Several studies have therefore been done in order to find a way to prolong the clinical effect. Repeated oral doses are recommended in second trimester abortion and have also been studied in early first trimester. Oral administration of misoprostol and vaginal administration of gemeprost seem to be equally effective in terminating early pregnancy if the duration of amenorrhoea is <50 days. Thereafter clinical data indicate that oral misoprostol is less effective (El-Refaey, 1995; von Hertzen *et al.*, 2003).

However, if misoprostol (tablets for oral use) is administered vaginally the efficacy is increased and side-effects decreased. A possible reason for the more pronounced effect of vaginal misoprostol could be a slower uptake and metabolism and a more prolonged elevated plasma concentration compared to the oral route resulting in higher bioavailability (Zieman *et al.*, 1997; Gemzell-Danielsson *et al.*, 1999; Tang *et al.*, 2002). The prolonged exposure of the myometrium to misoprostol after vaginal administration results in the development of uterine contractions, which is not the case after oral administration (Gemzell-Danielsson *et al.*, 1999). A drawback of vaginal administration is the great variation in plasma levels between individual women (Zieman *et al.*, 1997; Gemzell-Danielsson *et al.*, 1999; Tang *et al.*, 2002). Furthermore, although vaginal administration has been shown to be more effective and gives fewer side-effects, oral administration is preferred by women (Ho *et al.*, 1997).

An alternative to vaginal administration would be to administer misoprostol in a slow-release form that would also allow a prolonged effect on the myometrium. Preliminary pharmacokinetic studies *in vitro* (Chen *et al.*, 2000) indicate that this regimen may result in a longer duration of elevated concentrations of the active misoprostol free acid. It was shown that, after 4 h in water solution with a pH of 1.2, ~40% of the SR misoprostol

had dissolved *in vitro*. In contrast, the uptake of oral misoprostol is rapid with a peak plasma level after ~30 min and thereafter declining to zero (Zieman *et al.*, 1997; Gemzell-Danielsson *et al.*, 1999; Tang *et al.*, 2002).

The objective of this study was to determine the pharmacokinetics of a slow-release form (SR) of misoprostol (Chen *et al.*, 2000) administered orally in two different doses, 400 and 800 µg, compared with 400 µg conventional misoprostol also administered orally. *In vitro* data indicate that the amount of misoprostol released from a 400 as well as from a 800 µg SR misoprostol tablet during 4 h following oral administration would be less than following oral administration of 400 mg conventional misoprostol. But the plasma levels would be elevated for a longer period of time for the SR compound.

Materials and methods

Healthy pregnant women requesting surgical termination of pregnancy at a gestation of 8–12 weeks were recruited for the study, which was carried out during the period February to August 2004 in the Department of Obstetrics and Gynecology at the Karolinska University Hospital, Sweden. Inclusion criteria were healthy women ≥18 years, who were pregnant with an apparent normal intrauterine pregnancy, scheduled for surgical abortion and who agreed to participate. Exclusion criteria were a history of allergy to misoprostol, any signs of local infections, major medical problems or not being able to understand the information provided. (Figure 1).

Duration of pregnancy was calculated from the last menstrual period and confirmed by gynaecological examination and vaginal ultrasound. The women gave their written informed consent and the study was approved by the Ethics Committee at the Karolinska Hospital/Institutet.

The patients were asked to fast overnight and were admitted to the hospital on the morning of the operation. The women were randomized into the three treatment groups according to a computer-generated randomization schedule. The allocations were put in opaque sealed envelopes. All patients received the study drugs from the same research nurse. The rest of the staff and the researchers were unaware of the treatment allocations. The women were randomized into three groups to receive orally 400 µg of conventional misoprostol (two

tablets) or SR misoprostol either 400 µg (one tablet) or 800 µg (two tablets). An 18-gauge i.v. catheter suitable for repeated blood sampling was inserted into the antecubital vein at the beginning of the study. The tablets were taken orally with water.

Venous blood samples (10 ml) were drawn over the next 6 h at 0, 30, 60, 120, 240 and 360 min after the administration of misoprostol. The blood samples were immediately centrifuged in a cooled centrifuge and frozen at –20°C in liquid nitrogen. Surgical termination of the pregnancy with vacuum aspiration was performed after about 4 h from treatment following the routine protocol. The samples were then stored below –20°C and sent to the Department of Paediatrics, Philipps University Marburg for analysis. Based on a former published gas chromatography–mass spectrometry (GC-MS/MS) method, misoprostol acid (MPA) was determined in plasma samples using a modified liquid chromatography/tandem mass spectrometry (LC-MS/MS) assay (Watzel *et al.*, 2005). After addition of 15(S)-15-methyl prostaglandin E₂ (15-methyl-PGE₂) as internal standard, MPA was extracted from plasma samples using a monolithic reversed phase cartridge. The prostanoids were eluted with diisopropyl ether. The dried and reconstituted sample was determined by LC-MS/MS using the [M–H][–] ions as precursor in the negative ion electrospray ionization (ESI) mode. The product ions used for quantification were [M–2H₂O–C₆H₁₀–H][–] (MPA) and [M–2H₂O–CO₂–C₇H₁₂–H][–] (15-methyl-PGE₂) respectively. The detection limit of the assay was 1 pg/sample.

The primary objectives of the study were the determination of the area under the curve of serum concentrations of MPA against time up to 360 (AUC₃₆₀) min, peak concentration of misoprostol level (C_{max}) and the time to attain the peak concentration (T_{max}). The area under the curve was calculated by dividing the area under the curve into trapezium segments according to the time intervals of blood sampling. The area of each segment was computed according to the following formula for calculation of trapezium area: 0.5 [C_x + C_{x–1}] [time interval], x = 1 to 12. The AUC₃₆₀ was calculated by summing the trapezium segments. The difference in the AUC₃₆₀ was used for calculation of the sample size.

A previous study has shown that the AUC₃₆₀ was 433 ± 183 pg·h/ml for vaginal misoprostol (Tang *et al.*, 2002). Therefore, 10 subjects in each group gave 80% power in detecting a difference of 243 pg·h/ml in AUC₃₆₀ at 5% level of significance. Kruskal–Wallis tests were applied to test the overall difference between the three groups. If the overall difference was significant Wilcoxon two-sample tests were used to test pair-wise differences. P < 0.05 (two-tailed) was considered statistically significant. P-values were not corrected for multiple comparisons; however, all stated differences remained significant (P < 0.05) after applying Bonferroni–Holm correction (Holm, 1979). Statistical analyses were performed by PROC NPARIWAY (exact tests) in SAS software (SAS Institute Inc., 1997).

Side-effects such as nausea, vomiting, diarrhoea and abdominal pain were recorded. The need for any analgesic up to the time of surgery was noted. The pain score was assessed post-administration of misoprostol. Pain levels were measured by a 100 mm linear visual analogue scale (VAS; 0 = no pain, to 100 = most severe pain). The subject was given the linear VAS by a research nurse (who was blinded to the study group assignment) and asked to indicate the pain level.

Uterine contractions were recorded in all patients. An analysis of these data has recently been published (Fiala *et al.*, 2005).

Results

A total of 377 women requested surgical termination of pregnancy during the study period. Among them 31 women met the inclusion criteria and agreed to participate in the study. The women were randomized, treated and completed 6 h blood

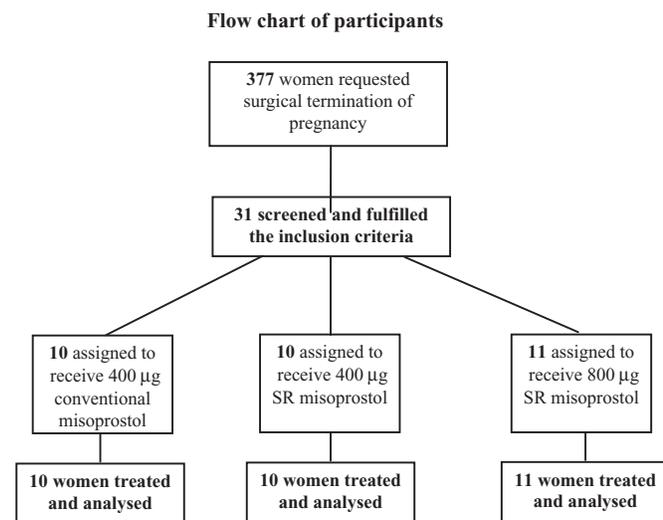


Figure 1. Flow chart.

sampling. The clinical characteristics of the patients are shown in Table I. Women in the three groups did not differ significantly in age, parity, gestational age, height, weight or body mass index (BMI). All women were at 8–12 weeks of gestation and mean parity was 0.8, 1.1 or 1.2 respectively (range 0–3).

The mean serum concentrations of MPA after administration of 400 µg oral misoprostol, 400 µg SR misoprostol or 800 µg SR misoprostol are shown in Figure 2. A number of pharmacokinetic parameters were studied, namely the peak concentration (C_{max}), time to the peak concentration (T_{max}) and AUC_{360} (Table II). Treatment with conventional orally administered misoprostol resulted in the highest serum peak concentration. The difference was statistically significantly higher compared to each of the other groups. Treatment with 800 µg SR misoprostol resulted in significantly higher serum concentrations than 400 µg SR misoprostol. The highest dose of SR misoprostol, 800 µg, gave a significantly higher serum peak concentration compared with the lower SR dose of 400 µg. The time to peak concentration was similar in the three groups.

Plasma levels of MPA were significantly higher following SR 800 µg misoprostol compared to 400 µg conventional misoprostol at 3 h ($P < 0.01$) and 4 and 6 h ($P < 0.001$).

At 6 h plasma levels of MPA following 400 µg SR misoprostol was significantly higher than levels following 400 µg conventional misoprostol ($P < 0.01$).

Table I. Patient characteristics

	Misoprostol		
	400 µg	400 µg SR	800 µg SR
Age (years)	26 (18–38)	29.5 (18–40)	32 (19–39)
Parity	0.8 (0–2)	1.2 (0–3)	1.1 (0–3)
Gestational length (days of amenorrhoea)	61.5 (48–80)	60 (44–83)	58 (45–75)
Body mass index	24.5 (19.8–31.5)	22.1 (19.7–34)	23.6 (20.2–31.2)

Values are median (range).
SR = slow release.

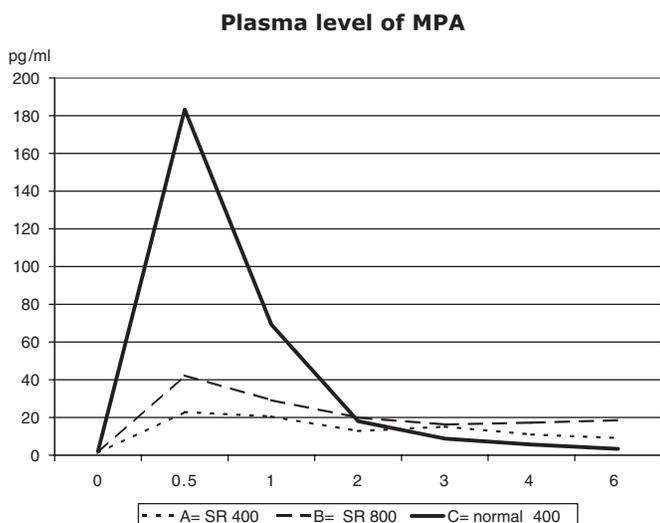


Figure 2. Mean plasma concentrations of misoprostol acid over time.

Table II. Pharmacokinetic parameters of the three groups of misoprostol

	400 µg SR (n = 10)	800 µg SR (n = 11)	400 µg oral (n = 10)	P^a
Peak concentration of misoprostol acid (C_{max}) (pg/ml)				
Mean ± SD	27.2 ± 14.5	43.5 ± 17.7	186.2 ± 118.8	< 0.0001 ^b
CV	53.4	40.7	63.8	
Time to attain peak concentration (T_{max}) (min)				
Mean ± SD	54.0 ± 46.5	81.8 ± 111.5	36.0 ± 12.6	NS ^c
CV	86.0	136.0	35.0	
Area under the curve to 360 min (AUC_{360}) (pgh/ml)				
Mean ± SD	80.7 ± 59.8	124.0 ± 35.0	182.8 ± 108.0	0.008 ^d
CV	28.6	17.8	12.6	

^aKruskal–Wallis test.

^b C_{max} 800 µg slow release (SR) was significantly higher than 400 µg SR ($P = 0.03$), C_{max} for the oral conventional group was significantly higher than 400 µg SR ($P = 0.0002$) and C_{max} 800 µg SR was significantly higher than for the oral conventional group ($P = 0.001$), by Wilcoxon two-sample test.

^c T_{max} for the three groups did not differ significantly ($P = 0.62$).

^d AUC_{360} for 800 µg was significantly greater than 400 µg SR ($P = 0.013$) and conventional 400 µg was significantly greater than 400 µg SR ($P = 0.012$) groups, by Wilcoxon two-sample test.

CV = coefficient of variation (%); NS = not significant.

The AUC_{360} in the 800 µg SR and conventional misoprostol groups were significantly greater than those in the 400 µg SR group, but no significant difference was found between the two former groups.

The individual variability of these pharmacokinetic parameters was denoted by the coefficient of variation (CV) (Table II).

The only side-effects reported were abdominal pain, nausea and vomiting. Seven women reported pain, two in the 400 µg conventional misoprostol group, two in the 400 µg SR misoprostol group and three in the 800 µg SR misoprostol group. Two of these women, one per SR misoprostol group, received pain treatment with paracetamol and codein (T. Citodon). The pain was regarded as mild or moderate. One woman per group reported nausea and two vomited, both following 800 µg SR misoprostol. Vomiting occurred 1.5–2.5 h following administration. These recordings were not excluded from analysis.

Discussion

This is the first report on the *in vivo* pharmacokinetics of a novel SR form of orally administered misoprostol. Recently a slow-release (SR) or sustained-release form of misoprostol was synthesized and the chemical properties described (Chen *et al.*, 2000). Pharmacokinetic studies *in vitro* (Chen *et al.*, 2000) indicate that the dissolution of this SR misoprostol is prolonged and after 4 h at a low pH only ~40% had been released from the tablets. Our study confirms that 800 µg of SR misoprostol results in a longer duration of elevated plasma concentration of the active misoprostol free acid (MPA) compared to plasma levels of MPA seen after 400 µg of conventional oral misoprostol. To date, there is no slow release or sustained release dosage form of misoprostol available on the market. Only very few previous studies have investigated the effect of SR prostaglandin analogues in induced abortion. A slow-release vaginal prostaglandin analogue was studied in second trimester termination of pregnancy (Bygdeman *et al.*, 1984) and later another vaginal prostaglandin that was released

during a 24 h period was studied for termination of early abortion up to 56 days gestation (Cameron and Baird, 1986). Both regimens were used without pre-treatment with mifepristone.

An oral slow-release preparation of misoprostol, if effective during a sufficiently long time period, is likely to be more acceptable to women (Ho *et al.*, 1997). Previously our group and others have studied and compared the pharmacokinetics of vaginal and oral administration of misoprostol. Following administration of oral misoprostol the increase in plasma levels of MPA occurred earlier and higher maximal levels were reached (Zieman *et al.*, 1997; Gemzell-Danielsson *et al.*, 1999; Tang *et al.*, 2002). After administration of vaginal misoprostol, MPA rose more slowly but remained elevated for a longer period, resulting in a higher bioavailability measured as the AUC of MPA. The newly described sublingual route resulted in higher peak plasma levels and a greater bioavailability of MPA than both oral and vaginal misoprostol. However, the serum levels of MPA remained elevated for a longer period following vaginal administration.

A longer-lasting elevation in plasma levels may lead to a prolonged stimulation of the myometrium by misoprostol. Previously we have shown that similar doses of vaginal misoprostol give a longer-lasting effect on the development of uterine contractions compared with oral misoprostol (Gemzell-Danielsson *et al.*, 1999). Sublingual misoprostol results in development of uterine contractions but the effect may be shorter-lasting than following the vaginal route (Aronsson *et al.*, 2004a).

Clinical studies have demonstrated that vaginally administered misoprostol is more effective than oral misoprostol for early and late medical abortion (El Refaey *et al.*, 1995; Ho *et al.*, 1997; von Hertzen *et al.*, 2003). Sublingual misoprostol seems to be slightly more effective than oral misoprostol and equally effective as vaginal misoprostol for cervical ripening but with more side-effects (Aronsson *et al.*, 2004b; Hamoda *et al.*, 2004; Tang *et al.*, 2004). In the second trimester, repeated doses of vaginal versus sublingual misoprostol resulted in a higher number of complete abortions in the vaginal group at 24 h whereas there was no difference at 48 h (Tang *et al.*, 2004).

A slower release and prolonged elevation of MPA plasma levels of misoprostol following treatment with mifepristone may result in a lower total dose of prostaglandin needed and a lower rate of side-effects compared to oral and sublingual administration, which have rapid and strong effects on uterine tone (Tang *et al.*, 2002; Aronsson *et al.*, 2004a). Prolonged plasma levels may reduce the need for repeated doses in later gestations and is also likely to be more acceptable to women than the vaginal route of administration. The SR misoprostol may also offer a way to reduce the interval between mifepristone and misoprostol or even make single dose administration possible.

The optimal dose of SR to be effective in medical abortion remains to be identified. In a previous study comparing the pharmacokinetics of oral and vaginal administered misoprostol, the same dose of vaginal misoprostol resulted in an AUC that was three times higher than that seen after oral administration (Zieman *et al.*, 1997). Thus it is possible that the dose of SR misoprostol should be increased to >800 µg, a dose that was

shown to result in a similar AUC up to 6 h after administration as 400 µg conventional oral misoprostol. Previous studies on conventional misoprostol have shown that the AUC₃₆₀ is greater for vaginal route compared to oral route. From the present study, it was shown that the AUC₃₆₀ of 800 µg of SR preparation was still inferior to oral preparation of conventional misoprostol although not statistically significant. Therefore, a higher dose of SR preparation may be needed to obtain a bioavailability similar to that of vaginal administration of conventional misoprostol. In the present study SR and conventional misoprostol both given orally were not compared in a dose to dose way for 800 µg. In further studies it would also be important to compare the optimal SR dose with the optimum vaginal and oral administration of conventional misoprostol. Further studies will also be needed to give information on the plasma levels of MPA beyond 6 h following administration of conventional as well as SR misoprostol.

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