Prevention of aspirin-induced faecal blood loss by prostaglandin E₂*

M M COHEN, GLADYS CHEUNG, AND D M LYSTER.

From the Departments of Surgery and Pathology, University of British Columbia, Vancouver, BC, Canada

SUMMARY Prostaglandins have been shown in animal laboratory studies to be capable of protecting the gastrointestinal tract against injury by exogenous agents. This study was conducted to determine if prostaglandin E₂ (PGE₂), which is native to the human gastric mucosa, could influence the increase in faecal blood loss associated with the ingestion of aspirin (ASA). A randomised double-blind study was performed on 27 healthy men. Faecal blood loss was measured by the ⁵¹Cr labelled red cell technique. ASA (600 mg four times daily) caused a significant increase in faecal blood loss. PGE₂ (1 mg four times daily) had no effect on faecal blood loss when administered alone. When given in addition to ASA it resulted in a faecal blood loss not significantly different from control. No significant alteration in intestinal transit occurred. It is concluded that PGE₂ protects man from the gastrointestinal injury associated with ASA.

Prostaglandin E₂ (PGE₂) occurs naturally in human gastric mucosa,¹ but its physiological role is not clear. Inhibition of acid secretion² and stimulation of mucus production^{3,4} and nonparietal cell secretion⁵⁻⁷ have been documented. In animal experiments a 'cytoprotective' property has been described⁸ by which gastric erosions induced by a variety of exogenous agents can be prevented by prostaglandin analogues incapable of acid inhibition or by doses of PGE₂ below the threshold for acid inhibition.⁹ There is evidence in man that aspirin-induced changes in gastric transmucosal potential difference can be prevented by PGE₂.¹⁰

In clinical practice, aspirin may cause significant gastrointestinal bleeding¹¹ probably due to injury to the gastric mucosa.¹² Most individuals on large doses of aspirin lose an extra few millilitres of blood each day in their faeces.^{13,14}

This study was conducted to determine if oral PGE₂ could prevent this increased faecal blood loss associated with the ingestion of aspirin.

Methods

Thirty-one healthy adult males (aged 19-40 years) were admitted to the trial. All gave informed written

consent and the study was approved by the ethics committee of the University of British Columbia. Initial screening comprised a full medical history, physical examination, and standard laboratory studies. Subjects were excluded who were known to be taking steroids within the past six months, or aspirin or any other analgesics within the previous two weeks. Individuals with a history of ulcer disease or haemorrhoids were also excluded.

The subjects were randomly allocated to one of four groups using a table of random numbers (Table 1). All drugs were administered before meals and at bedtime. PGE₂ was started 24 hours before the ASA and individual doses were taken 30 minutes before ASA. PGE₂ was supplied as 1.0 mg in 5 ml 10% ethanol, which was diluted by the subject in 100 ml tap water. ASA was supplied as non-buffered 300 mg tablets. Drug administration

Table 1 *Treatment programme, transit time (days) and day 7 serum salicylate (mg/dl) in the four groups of volunteers*

Group	No.	Day of study				Mean Transit time (±SD)	Mean salicylate level (±SD)
		4	5	6	7		
I	6	P ₁	P ₁ P ₂	P ₁ P ₂	P ₁ P ₂	3.2±1.2	2.5±1.3
II	9	P ₁	P ₁ A	P ₁ A	P ₁ A	3.0±0.7	10.0±2.8
III	7	PG	PGA	PGA	PGA	3.3±1.0	10.8±3.1
IV	5	PG	PGP ₂	PGP ₂	PGP ₂	3.4±0.9	3.2±0.56

P₁: prostaglandin placebo. A: aspirin 600 mg × 4 daily. P₂: aspirin placebo. PG: PGE₂ 1 mg × 4 daily.

*This work was reported at the Fourth International Prostaglandin Conference, Washington, DC, May 1979.

†Address for correspondence: Dr M M Cohen, Department of Surgery, Mount Sinai Hospital, 600 University Avenue, Toronto, Ont., M5G 1x5 Canada.

Received for publication 7 January 1980

started on the morning of the fourth day of stool collection. The placebo tablets were plain white scored tablets of lactose.

INTESTINAL TRANSIT

Intestinal transit was measured during the period of drug administration using a modification of the technique described by Hinton *et al.*¹⁵ Twenty radio-opaque plastic pellets (22 mg) were swallowed with the first dose of PGE₂ or placebo. Another 20 pellets were taken with the first dose of ASA or placebo. Each 24 hour stool collection was radiographed before being homogenised until all the pellets could be accounted for. Intestinal transit was determined in days as the time taken for 36 pellets to pass.

FAECAL BLOOD LOSS

Faecal blood loss was measured by the ⁵¹Cr labelled red blood cell technique.¹⁶ Approximately 20 ml of blood was withdrawn and 15 ml transferred to a vial containing ACD solution for labelling. The remaining 5 ml was used to measure radioactivity present in the subject's blood ('residual sample'). At least two minutes later exactly 10 ml of tagged blood was reinjected intravenously. Starting at midnight of the following day all stools passed were collected in plastic bags for 10 days, each 24 hour collection being separately labelled. Blood samples were taken on each weekday. Blood radioactivity was counted on 3 ml samples, using a Picker Autowell sample counter. The entire daily collection of stool was homogenised, weighed, and a sample counted in a Tubor sample counter. In order to relate the stool counts to the whole blood counts, which were performed on different instruments, standards were prepared which allowed for the variable geometry. A 'conversion factor' was calculated from the counts of each standard, and this was applied to all stool counts. All stool samples and the appropriate standards were counted on the day they were obtained.

CALCULATIONS

Background was subtracted from all samples and they were decayed to a common day postinjection. Each stool sample was corrected for decay, daily difference in instrument efficiency as determined by the standard preparation and for the difference in the two counting instruments using the 'conversion factor'. Total activity of the entire 24 hour stool collection was calculated. All blood samples were counted and a line of best fit calculated to provide total activity per ml of blood for each study day. The amount of blood (ml) in each 24 hour stool collection was obtained by dividing the 24 hour stool activity by the activity per ml of blood obtained from the subject the previous day.

ANALYSIS OF DATA

A one-way analysis of variance¹⁷ was computed for each study day. The pooled error variances so derived and Dunnett's *t* statistic were used to compare all active drug treatment means to the placebo control mean for each study day. A one-sided $\alpha=0.05$ level test was used. As the sample size varied among the treatment groups, Dunnett's *t* statistic was calculated using the harmonic mean of the sample sizes.

Serum salicylate was measured at the time of initial screening and on the last day of ASA administration the blood being drawn just before the second last dose of ASA. The method of assay was by Trinder's method which gives a low background value for serum which contains no salicylate. The initial laboratory studies were repeated at the conclusion of the study.

Results

Four of the original subjects were not included in the analysis of the data: two lost their drug supply and two had baseline faecal blood content greater than 5 ml/day. The only side-effect recorded

Table 2 Mean faecal blood loss ($\pm$ SD)

Treatment group		Day of study									
		1	2	3	4	5	6	7	8	9	10
I	Placebo	0.63 ± 0.41	0.72 ± 0.53	0.71 ± 0.55	1.19 ± 1.06	1.30 ± 1.50	0.97 ± 0.72	0.77 ± 0.35	0.64 ± 0.14	0.92 ± 0.74	0.54 ± 0.43
II	Aspirin + placebo	0.63 ± 0.27	0.56 ± 0.45	0.47 ± 0.37	0.93 ± 1.42	1.17 ± 1.70	2.12 ± 1.47	2.57 ± 1.70	3.71* ± 2.77	2.61* ± 1.11	1.94* ± 1.05
III	PGE ₂ + aspirin	1.44 ± 1.42	1.04 ± 1.05	1.32 ± 1.94	0.75 ± 0.64	0.52 ± 0.20	0.85 ± 0.74	1.45 ± 0.94	1.65† ± 0.80	1.58† ± 1.33	1.22 ± 0.62
IV	PGE ₂ + placebo	0.82 ± 0.45	0.44 ± 0.35	0.46 ± 0.45	0.38 ± 0.06	0.66 ± 0.46	0.32 ± 0.22	1.44 ± 0.98	1.72 ± 2.13	1.00 ± 1.17	0.53 ± 0.14

*Significantly different from placebo ($\alpha=0.05$ level one-sided test).

†Significantly different from aspirin plus placebo ($\alpha=0.05$ level one-sided test)

during the study was transient mild diarrhoea, lasting one day only, in two of the subjects who received PGE₂.

There was no difference in intestinal transit between the four groups (Table 1), and the time taken for 90% of the radiopaque pellets to pass was comparable with that found by Hinton *et al.* in normal subjects.¹⁵ The mean serum salicylate increased in the group taking aspirin and placebo and in the group taking aspirin and PGE₂ to values which were not significantly different from each other but were significantly higher ($P < 0.01$) than those in the double placebo group or the PGE₂ and placebo group (Table 1).

Faecal blood loss in ml/day in the four treatment groups is given in Table 2 and shown in the Figure. Subjects treated with aspirin and placebo showed a significant ($P < 0.05$) increase in blood loss compared with placebo-treated subjects. The highest blood

Figure Faecal blood loss during each day of the study in the four treatment groups. Asterisks indicate losses significantly ($P < 0.05$) greater than control.

content in the stool occurred one day after aspirin administration was stopped—that is, it coincided with the second day of aspirin treatment after intestinal transit time was allowed for. Mean blood loss in the groups who received aspirin plus PGE₂ or PGE₂ plus placebo was not significantly different from that in the double placebo group. Mean blood loss in the group receiving aspirin plus PGE₂ was significantly less ($P < 0.05$) than in the group receiving aspirin plus placebo.

No changes were detected in the laboratory values at the completion of the study.

Discussion

The results of this study confirmed our preliminary data¹⁸ and indicate that oral PGE₂ will protect the human stomach against the damage normally caused by standard therapeutic doses of aspirin. Preliminary data also show that the damage done by indomethacin in arthritic patients can be similarly prevented.¹⁹

The concept that prostaglandins, which are synthesised by the gastric mucosa, play a role in protecting the gastric mucosa against injury is attractive. A 'cytoprotective' action of the prostaglandins has been known for some years, but it has only recently been recognised that this action is unrelated to inhibition of acid secretion. Thus the precise mechanism of this protective action remains unclear. Prostaglandins have several additional actions on the gastric mucosa all of which are potentially beneficial. Mucus secretion is stimulated,^{3,4} bicarbonate secretion is stimulated,^{5-7,19} and blood flow is increased.²¹⁻²³ Although the prostaglandins have no effect on the normal gastric mucosal barrier,^{24,25} there is evidence that prostaglandin can prevent¹⁰ and reverse²⁶ damage to the gastric mucosal barrier. The results of this investigation do not permit deductions about the mechanism of action of prostaglandins but clearly confirm that the protection afforded is unrelated to acid inhibition as oral PGE₂ has no effect on human gastric acid secretion.²⁷

Aspirin, indomethacin, and the other more recently introduced non-steroid anti-inflammatory analgesics all inhibit prostaglandin synthesis and all cause damage to the gastric mucosa. The proof that these agents damage the mucosa as the direct result of the inhibition of prostaglandin synthetase is still lacking. The indirect evidence is accumulating and our results add human data to that obtained from animal studies.

Aspirin is the most widely used of all drugs and in the United States alone it is estimated that

20 to 30 tons are consumed daily.²⁸ The majority of individuals who take therapeutic doses of aspirin lose an extra few millilitres of blood daily in their stool,^{13,14} and in a few who are sensitive there can be frank clinical bleeding.^{11,29} Ingelfinger³⁰ has wryly calculated that the more than 20 thousand million tablets of aspirin consumed in the United States annually cause 10 million litres of blood to go down the toilet per year.

This trial suggests a potential clinical application of the prostaglandins in the prevention of this blood loss and possibly also the gastric upsets associated with the use of aspirin. The dose of PGE₂ used in this study (4 mg/day) caused mild transient diarrhoea in two subjects and is certainly too large to be administered to women who might be pregnant. If a lower dose of PGE₂ could be shown to be equally effective in preventing mucosal damage this would open up the possibility of a combination of aspirin and PGE₂ as perhaps the ideal analgesic. It remains to be shown, however, that this combined therapy is as effective as an anti-inflammatory analgesic.

We are grateful to the Upjohn Co., Kalamazoo, Michigan, USA for providing the drugs used in this trial and for financial support. This work was also supported by the Canadian MRC Grant No. MA5316. We are indebted to Gayle Henderson for typing the manuscript.

References

- ¹Bennett A, Murray JG, Wyllie JH. Occurrence of prostaglandin E₂ in human stomach and a study of its effect on human isolated gastric muscle. *Br J Pharmacol* 1968; **32**: 339-49.
- ²Newman A, Prado J, Philippakos D, Misiewicz JJ. The intravenous effect of infusions of prostaglandin E₂ and F_{2α} on human gastric function. *Gut* 1975; **16**: 272-6.
- ³Bolton JP, Palmer D, Cohen MM. Effect of the E₂ prostaglandins on gastric mucus production in rats. *Surg Forum* 1976; **27**: 402-3.
- ⁴Tao P, Scruggs W, Wilson DE. The effects of a prostaglandin endoperoxide analogue on canine gastric acid and mucus secretion. *Dig Dis Sci* 1979; **24**: 449-54.
- ⁵Bolton JP, Palmer D, Cohen MM. Stimulation of mucus and non-parietal secretion by the E₂ prostaglandins. *Am J Dig Dis* 1978; **23**: 359-64.
- ⁶Bolton JP, Cohen MM. Stimulation of non-parietal cell secretion in canine Heidenhain pouches by 16,16-dimethyl prostaglandin E₂. *Digestion* 1978; **17**: 291-9.
- ⁷Garner A, Heylings JR. Stimulation of alkaline secretion in amphibian-isolated gastric mucosa by 16,16-dimethyl PGE₂ and PGF_{2α}. *Gastroenterology* 1979; **76**: 497-503.
- ⁸Robert A, Schultz JR, Nezamis JE, Lancaster C. Gastric antisecretory and antiulcer properties of PGE₂, 15-methyl PGE₂, and 16,16-dimethyl PGE₂. Intravenous, oral and intrajejunal administration. *Gastroenterology* 1976; **70**: 359-70.
- ⁹Robert A. Antisecretory, antiulcer, cytoprotective and diarrhoeogenic properties of prostaglandins. *Adv Prostaglandin Thromboxane Res* 1976; **2**: 507-20.
- ¹⁰Cohen MM, Pollett JM. Prostaglandin E₂ prevents aspirin and indomethacin damage to human gastric mucosa. *Surg Forum* 1976; **27**: 400-1.
- ¹¹Jick H, Porter J. Drug-induced gastrointestinal bleeding. *Lancet* 1978; **2**: 87-9.
- ¹²Loebl DH, Craig RM, Culic DD, Ridolfo AS, Falk J, Schmid FR. Gastrointestinal blood loss: effect of aspirin, fenoprofen, and acetaminophen in rheumatoid arthritis as determined by sequential gastroscopy and radio-active fecal markers. *JAMA* 1977; **237**: 976-81.
- ¹³Grossman MI, Matsumoto KK, Lichter RJ. Fecal blood loss produced by oral and intravenous administration of various salicylates. *Gastroenterology* 1961; **40**: 383-8.
- ¹⁴Croft DN, Wood PHN. Gastric mucosa and susceptibility to occult gastrointestinal bleeding caused by aspirin. *Br Med J* 1967; **1**: 137-41.
- ¹⁵Hinton JM, Lennard-Jones JE, Young AC. A new method for studying gut transit times using radio-opaque markers. *Gut* 1969; **10**: 842-7.
- ¹⁶Holt PR. Measurement of gastrointestinal blood loss in subjects taking aspirin. *J Lab Clin Med* 1960; **56**: 717-26.
- ¹⁷Winer BJ. *Statistical principles in experimental design*. New York: McGraw-Hill 1962: 46-102.
- ¹⁸Cohen MM. Mucosal cytoprotection by prostaglandin E₂. (Letter.) *Lancet* 1978; **2**: 1253-4.
- ¹⁹Johansson C, Kollberg B, Nordemar R, Bergström S. Mucosal cytoprotection by prostaglandin E₂. (Letter.) *Lancet* 1979; **1**: 317.
- ²⁰Kauffman GL Jr, Grossman MI. Gastric alkaline secretion: Effect of topical and intravenous 16-16 dimethyl prostaglandin E₂ (Abstract). *Gastroenterology* 1979; **76**: 1165.
- ²¹Main IHM, Whittle BJR. The effects of E and A prostaglandins on gastric mucosal blood flow and acid secretion in the rat. *Br J Pharmacol* 1973; **49**: 428-36.
- ²²Gerkens JF, Flexner C, Oates JA, Shand DG. Prostaglandin and histamine involvement in the gastric vasodilator action of pentagastrin. *J Pharmacol Exp Ther* 1977; **201**: 421-426.
- ²³Boughton-Smith NK, Vane JR, Whittle BJR. Effects of prostacyclin (PGI₂), PGI₁, and 6-oxo-PGF_{1α} on the rat gastric mucosa. *Br J Pharmacol* 1978; **62**: 413p.
- ²⁴Bolton JP, Cohen MM. The effect of 16,16-dimethyl prostaglandin E₂ on the gastric mucosal barrier. *Gut* 1979; **20**: 513-7.
- ²⁵Bolton JP, Cohen MM. Permeability effects of the E₂ prostaglandins on canine gastric mucosa. *Can J Physiol Pharmacol* 1979; **57**: 1082-7.
- ²⁶Bolton JP, Cohen MM. The effect of prostaglandin E₂, 15-methyl prostaglandin E₂ and metiamide on established canine gastric mucosal barrier damage. *Surgery* 1979; **85**: 333-338.
- ²⁷Karim SMM, Carter DC, Bhana D, Ganesan PA.

Effect of orally administered prostaglandin E_2 and its 15-methyl analogues on gastric secretion. *Br Med J* 1973; **1**: 143–6.

²⁸Abrishami MA, Thomas J. Aspirin intolerance—a review. *Ann Allergy* 1977; **39**: 28–37.

²⁹Levy M. Aspirin use in patients with major upper gastrointestinal bleeding and peptic-ulcer disease. *N Engl J Med* 1974; **290**: 1158–62.

³⁰Ingelfinger FJ. The side effects of aspirin (Editorial). *N Engl J Med* 1974; **290**: 1196–7.