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Anti-ulcer Activity of a Slow-release Zinc Complex, Zinc Monoglycerolate (Glyzinc)

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Abstract—A slow-release zinc complex, zinc monoglycerolate (ZMG) was examined for its potential gastroprotective activity in various gastric ulcer models. These models comprised (a) oral or parenteral non-steroidal anti-inflammatory drugs (NSAIDs) given to rats whose gastrointestinal mucosa was pre-sensitized by prior development of arthritis, oleyl alcohol-induced inflammation and cold exposure, (b) oral ethanol (12·5–100%) with and without added 4% HCl, (c) intraperitoneal reserpine (5 mg kg^{-1}) in arthritic and normal rats and in normal mice, (d) oral NSAIDs given to mice in which acid and pepsin production was stimulated by co-administration of intraperitoneal bethanechol chloride (5 mg kg^{-1}) to enhance ulcer development, and (e) NSAIDs given to carrageenan-inflamed rats to determine effects of ZMG on paw inflammation. In these models, ZMG given orally was effective in preventing development of gastric lesions, except with propionic acid NSAIDs; the effective doses being apparently dependent on the severity of the mucosal injury. In many of the models ZMG was superior to zinc sulphate and other zinc salts or metal ion complexes investigated but was slightly more effective or equipotent compared with zinc acexamate. ZMG did not impair the anti-oedemic effects of NSAIDs. ZMG is thus an effective agent in preventing ulcer development in a wide range of model systems and may be more effective than zinc salts because of the controlled slow-release of zinc from the complex.

Zinc sulphate has been reported to enhance the healing of gastric ulcers in patients with proven gastric ulcer (Fraser et al 1972; Frommer 1975). Zinc salts and complexes with amino acids have been shown to have anti-ulcer activity in a variety of experimentally-induced ulcer models in laboratory animals (Cho et al 1977, 1977; Cho & Ogle 1977, 1978; Mann et al 1981; Cho & Pfeiffer 1982; Dupuy & Szabo 1986; Barbarino et al 1988). Zinc acexamate has also been used in the treatment of gastric and duodenal ulcers in man (Alcala-Santaella et al 1985; Navarro et al 1985) and in rats this compound protects against gastric ulcers induced by various drugs and necrotizing agents (Esplugues et al 1985; Escolar 1987; Pfeiffer et al 1987).

A novel slow-releasing complex developed recently, zinc monoglycerolate (ZMG, Glyzinc) has been shown to have anti-arthritic activity when applied topically or subcutaneously to rats in which arthritis was induced by the injection of mycobacterial adjuvant (Whitehouse et al 1986, 1990). Equivalent doses of soluble zinc salts were without effect (Whitehouse et al 1986). Studies with radiolabelled [^{65}Zn]ZMG showed that this complex slowly releases zinc during permeation through the skin or following subcutaneous administration to the arthritic rats (Whitehouse et al 1986). The slow release of zince from this complex could confer advantages as a potential anti-ulcer agent compared with conventional zinc compounds. It was thus decided in the present studies to examine the potential anti-ulcer effects of zinc monoglycerolate compared with other zin salts or complexes in various ulcer models in rats and mice.

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Materials and Methods

NSAID-induced ulcers in normal, arthritic and acutely-inflamed rats

Male Sprague-Dawley rats (200 ± 10 g, Charles River, Toronto, Canada) were individually housed and fasted for 24 h but with free access to water before the experiments were initiated.

For the arthritic group, the condition was induced in female Hooded, Dark Agouti and Sprague-Dawley rats (Waite Institute, University of Adelaide, Australia; Tucks, Raleigh, Essex, UK) by the subcutaneous injection into the tailbase of 0·5 mg mL^{-1} heat-killed and delipidated *Mycobacterium tuberculosis* (Weybridge strain) or *Mycobacterium butyricum* (Difco) suspended in 0·05 mL squalane (Sigma) or mineral oil (Sigma, Poole, Dorset) as previously described (Whitehouse et al 1974). The animals were used 14–18 days following induction of the disease at which stage the polyarthritis was manifest.

In other studies a long-lasting acute inflammation was induced in male and female Sprague-Dawley, Porton and Dark Agouti rats by injecting 0·1 mL oleyl alcohol subcutaneously into the tailbase 4 days before the experiments (Whitehouse & Rainsford 1987). In some experiments the animals were concurrently exposed to brief chilling (−18°C, 25 min) (Rainsford 1978). The exposure to such inflammatory insults and physical stress markedly sensitizes the gastric mucosa to the ulcerogenic effects of non-steroidal anti-inflammatory drugs (NSAIDs) and provides models for the effects of disease and stress exposure upon the gastrointestinal tract of chronically inflamed arthritic patients (Rainsford 1978, 1987a; Whitehouse & Rainsford 1987).

Where fasting was employed, the animals were placed in all-wire mesh cages in groups of 4–7 each and deprived of

food, but allowed free access to water, for 18–24 h before experimentation.

The NSAIDs were administered either orally as 1 mL fine aqueous suspensions prepared by homogenization immediately before use or intraperitoneally (1 mL) as sodium salts in 1·2 molar equivalents of sodium bicarbonate or sodium carbonate. The doses of NSAIDs employed were limited to those which are known from previous studies to give maximal responses.

Zinc complexes were prepared immediately before use as finely homogenized aqueous suspensions and administered orally or, in some experiments, in 0·9% NaCl (saline) parenterally. ZMG (Glyzinc) was obtained from Glyzinc Pharmaceuticals, Norwood (Adelaide, Australia). Zinc acexamate was prepared by dissolving basic zinc carbonate in equimolar aqueous acexamic acid (Aldrich, Poole, Dorset). These and other complexes (sulphate, oxide, chloride obtained from BDH, or Aldrich) were administered orally immediately before or 15 min before the NSAID. Details of the timing and group treatments will be found in the figure legends. In some experiments the protective effects of zinc compounds were compared with the standard anti-ulcer drugs, cimetidine (100 μmol kg^{-1} = 25 mg kg^{-1}, orally) and misoprostol (0·3 μmol kg^{-1} = 100 μg kg^{-1}, orally) against NSAID-induced mucosal injury in arthritic rats, some of which were exposed to brief chilling.

Two or 3 h after the NSAID or zinc compounds were administered, the animals were killed by cervical dislocation and the number (N) and severity (S, graded on a scale of 0 to 4+ based on increasing area of injury) of gastric lesions as well as the percentage of animals with gastric damage (%I) was recorded. The lesion index (LI) was computed as:

$$LI = N/n + S/n + \%I/10$$

where n = number of animals per group.

Ethanol-induced gastric ulcers in arthritic and normal rats

Adjuvant arthritis was induced in male or female Hooded and Dark Agouti rats (as above); animals were fasted before the experiments. The zinc complexes were administered orally or subcutaneously at 18 h, 30, 5 or 1 min before, or 30 min following oral administration of 1 mL 25% ethanol. In a second series of experiments the zinc complexes were given 15 or 30 min before the oral administration of 1 mL of 12·5%, or 20% aqueous, or 100% (absolute) ethanol. In a third study the ethanol was given in the same way but in 4% HCl. The animals were killed 1 or 2 h later and the number and severity of gastric lesions assessed for computing of the lesion index as above.

Reserpine-induced gastric ulcers in normal and arthritic rats and in mice

Previous reports (Ogle & Cho 1978; Pfeiffer et al 1980, 1987; Barbarino et al 1988) have described the anti-ulcer effects of zinc compounds in normal rats given reserpine, a drug which induces the release of endogenous amines. Using the same protocol as employed by Ogle & Cho (1978) ZMG and zinc sulphate were administered intraperitoneally 48 h before reserpine (5 mg kg^{-1}). In addition to employing the methods used by those authors, another group of arthritic as well as normal rats was also dosed orally with the zinc compounds for the same period. At this stage fasting was commenced and 18 h later the animals were killed and the gastric lesion index determined as above. Mice were treated similarly except that additional doses of the zinc complexes were given 60 h before termination. In one group of experiments, the rats were given reserpine for only 3 h to establish the effect of zinc pretreatment on lesion development after this short period of amine release.

The reserpine solutions were freshly prepared before use according to a method previously used in their commercial production (Ciba-Geigy, Horsham, UK, personal communication from Ms Georgina Fell). Thus, reserpine (Sigma, Poole, Dorset), 25 mg, was mixed with 20 mg ascorbic acid and 1 mg EDTA sodium in 2 g propylene glycol to which was added 0·33 mL 5% aqueous phosphoric acid. The mixture was stirred until dissolved (45–60 min) in a beaker which was protected from light by aluminium foil. Approximately 7 mL H_2O was then added, the mixture neutralized with 5% NH_4OH and made up to a final volume of 10 mL with water. The same mixture without the reserpine added was employed for intraperitoneal administration to control animals. The reserpine mixture produced similar gastric ulcers to those caused by solutions of the drug prepared according to the procedure of Pletscher et al (1955). The drug solutions prepared according to the latter authors were not, however, used in the main studies because they contain 25% ethanol and it was considered that this might influence the ulcerogenic effects of the reserpine.

NSAID-induced gastric ulcers in cholinomimetic-treated mice

Treatment of rodents with the cholinomimetic agent, bethanechol chloride (carbamyl-β-methyl choline chloride), markedly sensitizes their mucosa to the ulcerogenic effects of the NSAIDs presumably due to the stimulation of acid and pepsin production, both of which are important in gastric ulcerogenesis (Rainsford 1978, 1987a, b). Using this model (Rainsford 1987b), the effects of immediate or prior oral administration of ZMG (25–200 mg kg^{-1}) were assayed for potential gastro-protective activity against gastric lesions induced by aspirin (200 mg kg^{-1}, Monsanto, USA), and indomethacin (30 mg kg^{-1}, Merck, Sharpe and Dohme, UK) given to 24 h-fasted MF1 mice (Central Animal Services, University of Cambridge, UK). These drugs were prepared as fine suspensions as described above. The animals were killed by CO_2 asphyxiation, 2 h after drug administration. The stomachs were inflated with 4% formalin in buffered saline and the glandular mucosae mounted on glass microscope slides and cleared in glycerol for subsequent quantitative determination of the area and number of gastric lesions by visual image analysis (Rainsford 1987b).

Assay of anti-oedemic effects of NSAIDs given with ZMG

To determine if the gastro-protection afforded by ZMG compromised the normal anti-inflammatory effects of NSAIDs, the anti-oedemic effects of the combination of NSAID and ZMG were determined using the carrageenan paw oedema model; in the case of aspirin the model was adapted to include concomitant assay of gastro-irritancy as previously described (Rainsford & Whitehouse 1977). The compounds were given orally to fasted rats 30 min before subplantar injection of 50 μL 10 mg mL^{-1} carrageenan in

sterile non-pyrogenic saline in one paw with 50 μL saline given in the other paw as a control. Paw inflammation was determined 3 h after drug dosage. Immediately afterwards the animals in the aspirin group were killed, stomachs excised and the gastric lesion indices determined as described above.

Results

Gastro-protection by ZNG compared with other zinc complexes in arthritic and normal rats given aspirin or other NSAIDS

Fig. 1 shows that when orally administered, the zinc compounds were effective in preventing the gastric lesions induced by aspirin or indomethacin in normal (i.e. non-diseased) rats. In aspirin-treated rats, ZMG exerted the same anti-ulcer effects as $ZnSO_4$, except at the highest dose where ZMG appeared more effective. In indomethacin-treated animals $ZnSO_4$ appeared slightly more effective than ZMG. However, marked aggregation of mucus, opacification of the mucosa and discharge of the watery fluid was evident in aspirin, indomethacin and control animals given zinc sulphate, reflecting denaturation and disruption of the mucosal barrier; these effects were not observed with ZMG or zinc acexamate. Such destruction has been confirmed by observations from scanning electron microscopy of the mucose of animals given $ZnSO_4$ details of which have been submitted for publication elsewhere. Similar reduction in aspirin-induced lesions was also observed with zinc acexamate compared with ZMG (data not shown).

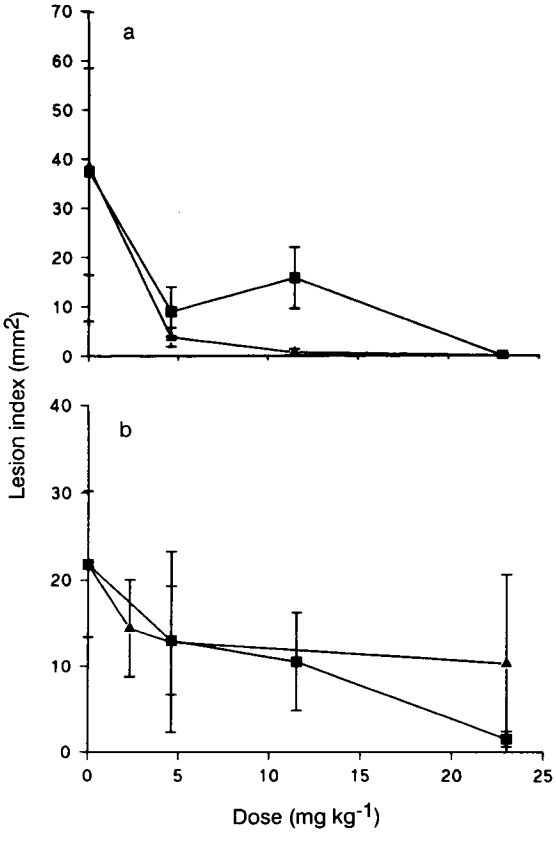

FIG. 1. Dose related protection against (a) aspirin (150 mg kg^{-1}, p.o.), and (b) indomethacin (30 mg kg^{-1}, orally) in 24 h fasted normal Sprague-Dawley rats. ■ ZMG, ▲ $ZnSO_4$; both given orally immediately before the NSAIDs. $ZnSO_4$ produced effusion of watery-like fluid and marked opacification of the mucosa of NSAID and control rats reflecting changes in the mucosal integrity of the mucosa. Values are means ± s.d. Statistically significant reduction (Mann-Whitney U-test, $P < 0.05$) in lesion index at > 5 mg kg^{-1} zinc (as $ZnSO_4$) and with 5 and 23 mg kg^{-1} zinc as ZMG, n = 5–6 per group.

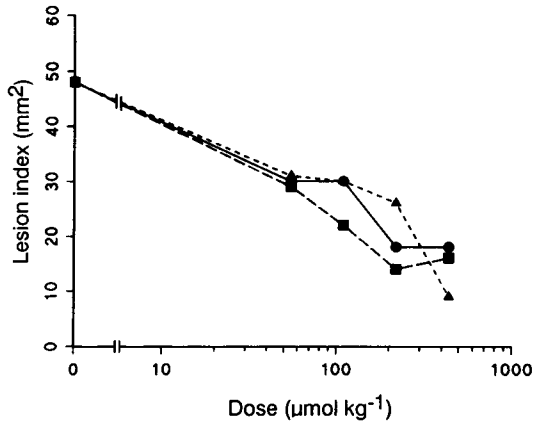

FIG. 2. Dose-related protection by zinc compounds against aspirin (32 mg kg^{-1}, oral)-induced lesions in arthritic female Dark Agouti rats. ● p.o. ZMG; ■ $ZnSO_4$; ▲ zinc acexamate. Each compound was given orally 5 min before aspirin. Misoprosotol (100 μg kg^{-1}), by comparison, reduced the ulceration from aspirin by 76% and cimetidine (25 mg kg^{-1}) by 61% under these conditions. Values are means ± s.d. Statistically significant reduction (Mann-Whitney, U-test, $P < 0.05$) in lesion indices occurred at doses of zinc compounds > 12 mg kg^{-1} (as zinc). n = 5–6 per group.

In arthritic rats, ZMG, $ZnSO_4$ and zinc acexamate were approximately equally effective in reducing the ulcerogenicity of aspirin in a dose-related fashion (Fig. 2). Similar effects were observed in arthritic animals given aspirin (120 mg kg^{-1}) orally and concurrently exposed to brief chilling; the gastroprotective effects of ZMG were comparable with $ZnSO_4$ both given at doses of 0·65 mmol kg^{-1} (control 59 ± 17 (mean ± s.d., number of lesions), ZMG 16 ± 14 and $ZnSO_4$ 12 ± 9; mean lesion index for control 32, and 12 for both ZMG and $ZnSO_4$). These results show that, in the most sensitive NSAID ulcer models (employing diseased and cold-exposed animals) ZMG still manifests appreciable anti-ulcer effects against aspirin-induced mucosal damage. Reduction in the ulcerogenicity of indomethacin by ZMG, $ZnSO_4$ and zinc acexamate was observed in arthritic rats exposed to brief chilling (Fig. 3). Likewise, inhibition of ulcerogenic effects of this and some other NSAIDs was observed in arthritic rats exposed to cold, the notable exception being the lack of effects observed with the propionic acids (Fig. 4a). The protective effects of ZMG against NSAID ulceration were comparable with those of cimetidine but not as marked as those of misoprostol (Figs 2, 3, 4b) when compared on a molar dosage basis.

The gastroprotective effects of ZMG and zinc acexamate were also evident in oleyl alcohol-inflamed rats as well as in normal and arthritic, fasted or replete rats exposed to brief chilling and given aspirin or non-propionic acid anti-inflammatory agents (data not shown). Neither ZMG nor $ZnSO_4$ affected the development of gastric lesions induced by

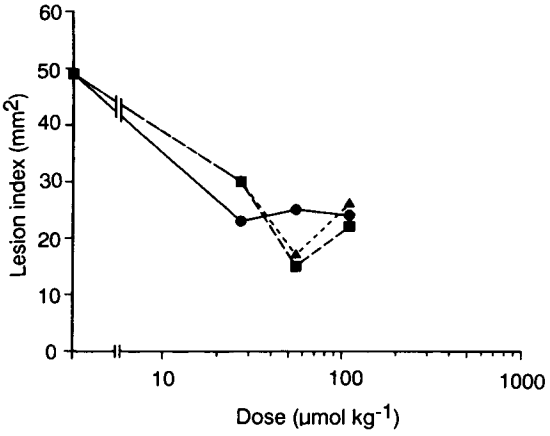

FIG. 3. Dose-response anti-ulcer effects of zinc compounds on indomethacin (3·6 mg kg^{-1}, p.o.) in male Dark Agouti arthritic rats exposed to −25°C for 20 min. ● ZMG; ■ $ZnSO_4$; ▲ zinc acexamate. By comparison indomethacin-treated rats given misoprostol (100 μg kg^{-1}, p.o.) showed no mucosal damage while those given cimetidine (25 mg kg^{-1}, p.o.) showed 91·4% reduction in ulceration under these conditions. Mean values plotted (n = 4–6 per group). All zinc compounds showed significant (Mann-Whitney U-test, $P < 0.05$) reductions in lesion index at doses of ⩾100 μmol kg^{-1}.

the intraperitoneal administration of aspirin, indomethacin or phenylbutazone in arthritic rats (data not shown).

Protection by ZMG and $ZnSO_4$ against gastric ulcers induced by ethanol with or without added HCl

The results in Fig. 5a and b show that the prior administration of ZMG or $ZnSO_4$, protected the gastric mucosa against 1 mL 20% aqueous ethanol in normal and arthritic rats, respectively. The protective effects were only manifest when these zinc compounds were given simultaneously with, or up to 30 min before the alcohol (data not shown). Longer periods of prior oral or subcutaneous administration of the zinc complexes, or giving them 30 min after the alcohol, failed to protect the gastric mucosa from ethanol injury (data not shown). The protective effects of ZMG were also evident in arthritic animals given 12·5–15% ethanol in 4% HCl but not with higher concentrations of ethanol in the acid (data not shown).

Protection by zinc compounds against reserpine-induced ulcers

Both ZMG given orally and $ZnSO_4$ given intraperitoneally inhibited the production of gastric ulcers in normal rats induced by 5 mg kg^{-1} intraperitoneal reserpine (Fig. 6). Moreover, ZMG (50 and 200 mg kg^{-1} orally) prevented the development of ulcers induced by the same dose of reserpine in arthritic rats, regardless of the time of administration of this ulcerogen or the severity of arthritic disease (Fig. 7). This zinc complex, given as two oral doses of 50 mg kg^{-1} each 60 and 48 h before the amine, also prevented the development of gastric ulcers in mice induced by 5 and 10 mg kg^{-1} reserpine (Fig. 7). In contrast to protection afforded by parenteral $ZnSO_4$, the administration of the monoglycerolate complex by this route at a dose of 50 mg kg^{-1} (i.e. equivalent to the dose of zinc in the $ZnSO_4$ of 21 mg kg^{-1} zinc), did not prevent the development of reserpine-induced ulcers in either rats or mice (data not shown).

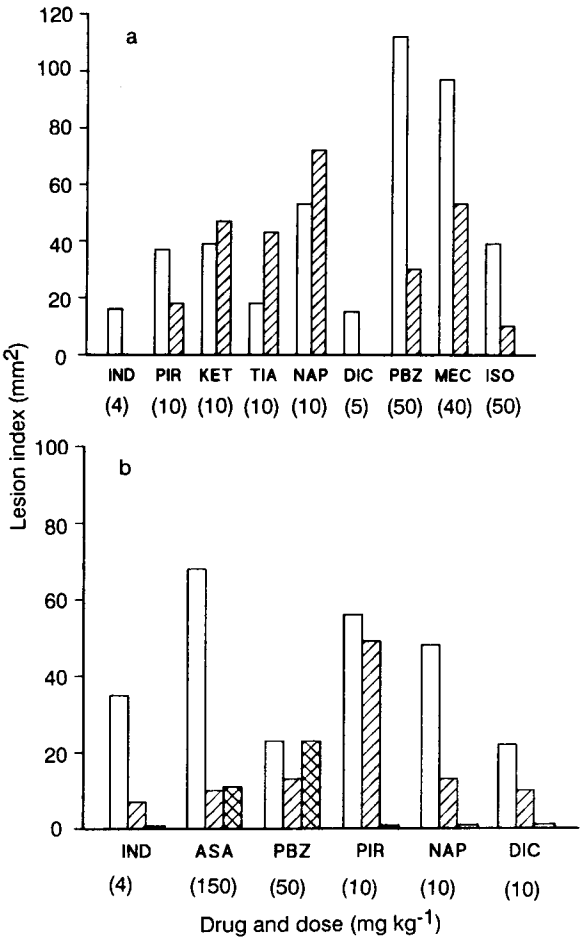

FIG. 4. Anti-ulcer effects of ZMG (100 mg kg^{-1}), cimetidine (25 mg kg^{-1}) and misoprostol (100 μg kg^{-1}) in Hooded rats exposed to −20°C for 25 min, and given various NSAIDs orally for 2 h. n = 5 per group. a. Effects: without ZMG (open), with ZMG (hatched). b. Comparison of ZMG (open), with cimetidine (hatched) and misoprostol (cross-hatched). IND = indomethacin, ASA = aspirin, PIR = piroxicam, KET = ketoprofen, TIA = tiaprofenic acid, NAP, naproxen, DIC = diclofenac sodium, PBZ, phenylbutazone, MEC- = meclofenamic acid, ISO = isoxicam. Some experiments with these NSAIDs were repeated in arthritic rats exposed to brief chilling ZMG consistently failed to inhibit the ulcerogenicity of propionic acid NSAIDs but always reduced the ulcerogenicity of other NSAIDs. Zinc and anti-ulcer compounds given 5 min before NSAIDs. n = 3–5 per group.

Protection by ZMG against gastric lesions induced by aspirin and indomethacin in cholinomimetic-treated mice

The results in Fig. 8 show that ZMG prevented the development of gastric haemorrhagic lesions induced by aspirin in cholinomimetic-treated mice in a dose-related fashion. The dose of the zinc complex required to achieve 50% reduction of lesions induced by 200 mg kg^{-1} aspirin in this model was 25 mg kg^{-1} while that against gastric lesions produced by 30 mg kg^{-1} indomethacin was approximately 20 mg kg^{-1}.

Effects of other metal ions

Other metal ions were tested in arthritic and chilled Hooded rats dosed orally with NSAID. The following complexes had no effect on NSAID-induced gastric injury: $BaSO_4$; $CaCl_2$,

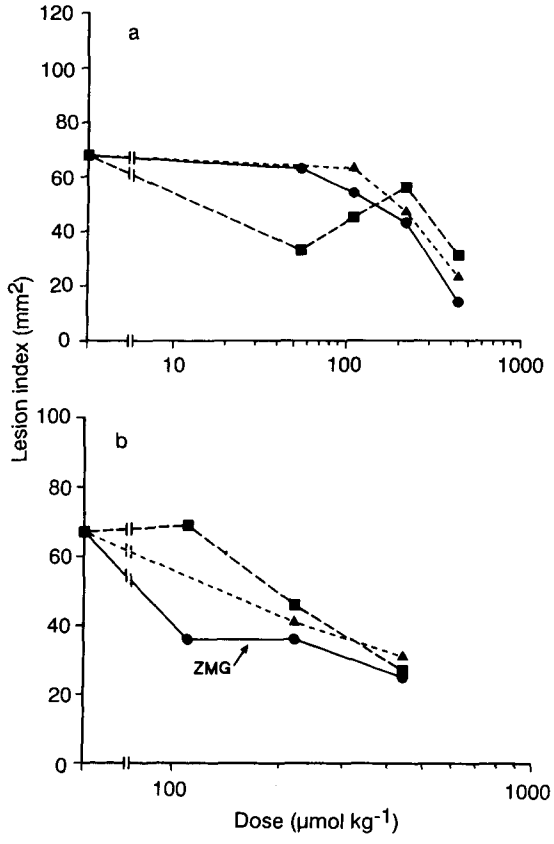

FIG. 5. Dose-response effects of zinc compounds against the gastric ulcerogenic effects of 20% aq. EtOH + 110 mM HCl in (a) normal and (b) arthritic male Hooded rats exposed to brief cold (−25°C, 20 min). ● ZMG; ■ $ZnSO_4$; ▲ zinc acexamate. Zinc compounds given orally 5 min before EtOH/HCl then killed 1 h later for determination of gastric mucosal damage. n = 3–5 per group. Values are mean. Statistically significant reduction in lesion indices was found with all zinc compounds.

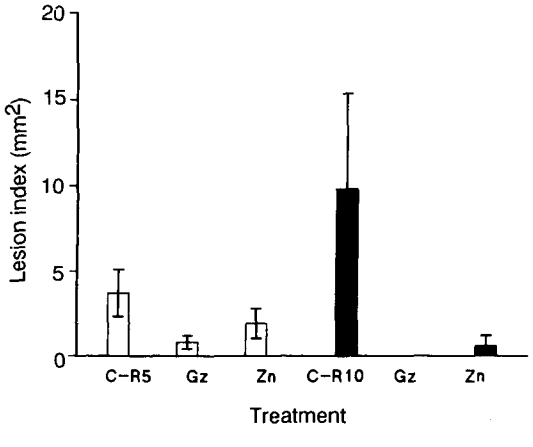

FIG. 6. Anti-ulcer effects of ZMG (Gz) and $ZnSO_4$ (Zn) 32 mg zinc kg^{-1} 48 h before and 5 and 10 mg kg^{-1} reserpine (C-R5, and C-R10, respectively). The rats were killed 18 h later during which time they were fasted but allowed free access to water. Area lesion index (mean ± s.d.). n = 5–6 per group. All zinc compounds showed a statistically significant reduction (Mann-Whitney U-test, $P < 0.05$) compared with reserpine alone.

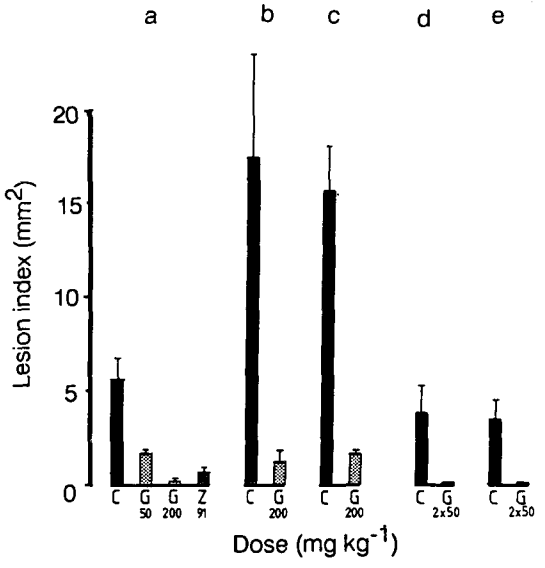

FIG. 7. Anti-ulcer effects of ZMG (G) and $ZnSO_4$ (Z) on the gastric ulcers induced by reserpine in normal and arthritic female (Hooded) rats and in normal MFI male mice. Experimental time denotes the time after reserpine treatment. All zinc complexes gave statistically significant (Mann-Whitney U-test, $P < 0.05$) reduction in area lesion indices. n = 5–6 per group. C = Control.

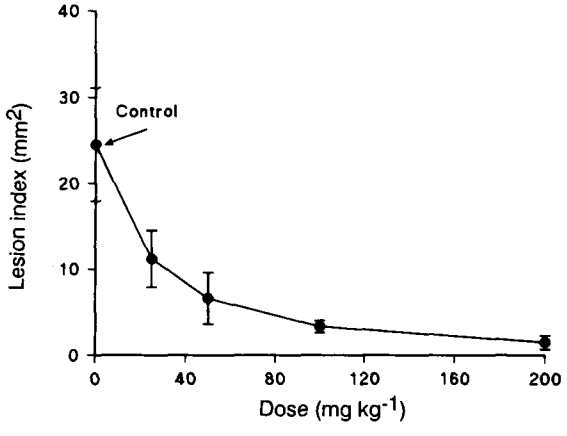

FIG. 8. Anti-ulcer effects of ZMG on aspirin-induced gastric mucosal lesions induced in male MFI mice (fasted 24 h) and given the cholinomimetic, acetyl-β-methylcholine chloride (= bethanechol chloride; 5 mg kg^{-1}, i.p.) for 2 h. The zinc complex was given orally 5 min before administration of aspirin (200 mg kg^{-1}). Area lesion index (mean ± s.d.) showed significant reduction at doses of ⩾ 25 mg kg^{-1} ZMG. (Mann-Whitney U-test, $P < 0.05$; n = 5 per group.)

$Co(NO_3)_2$, $FeSO_4$, $Fe_2(SO_4)_3$; $MgSO_4$ (data not shown). By contrast $CuSO_4$ and $CdSO_4$ effectively mimicked $ZnSO_4$ in reducing the development of lesions by some NSAIDs. Bismuth complexes also proved to have little or no gastroprotectant effects in arthritic dark Agouti rats (not subject to brief chilling).

Effects of concurrent administration of ZMG on the anti-oedemic actions of aspirin and other NSAIDs

The results in Table 1 show that the anti-oedemic effects of aspirin in the standard carrageenan paw assay were not

Table 1. Combined gastrotoxicity efficacy and assay of ZMG given with aspirin in normal Hooded rats given subplantar carrageenan.

Treatment	Mean swelling	Gastric lesion %	Gastric lesion index
None	2·67	—	04
Aspirin only	1·20	54	82
Aspirin + ZMG* prechill	1·10	59	34
Aspirin + ZMG* postchill	1·11	58	46
Aspirin + $ZnSO_4 \cdot 7\ H_2O$ (187 mg kg^{-1})	1·08	60	26

* ZMG = 150 mg kg^{-1} orally. Aspirin = 150 mg kg^{-1} orally. n = 3–4 per group.

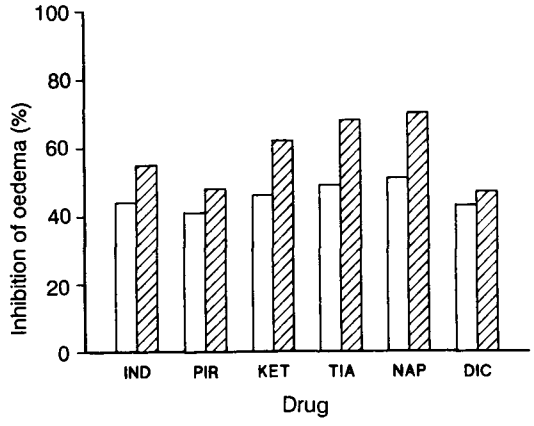

FIG. 9. Anti-inflammatory activity of NSAIDs given alone (open) or in combination with ZMG (100 mg kg^{-1}, p.o.) (hatched) in normal male Hooded rats. Paw swelling was induced by carrageenan injection (50 μL, 10 mg mL^{-1} in saline) in one paw and the difference measured by comparison with that in the saline-injected paw 3 h after drug dosage. The carrageenan was injected 30 min after dosing with the drug. Abbreviations and drug dosages as shown in Fig. 4.

compromised by co-administration of ZMG in doses at which anti-ulcer effects are observed in normal and cold-treated rats. $ZnSO_4$ was also without any untoward effects on the anti-inflammatory effects of aspirin. Likewise ZMG did not affect the anti-oedemic effects of other NSAIDs, and with propionic acids, this was actually slightly increased (Fig. 9).

Discussion

These studies confirm the property of zinc compounds to afford gastroprotection in a wide variety of ulcer models (Dupuy & Szabo 1986). Only copper and cadmium salts mimicked zinc in this respect but both enhanced mucosal oedema and mucus aggregation at higher doses (> 100 μmol kg^{-1}) and paradoxically with certain drugs they enhanced gastric injury at lower doses. Thus zinc complexes such as ZMG exhibit an element of specific mucosal protection and might provide an appropriate pharmaceutical delivery system for alleviating gastric toxicity, in general being more effective and less toxic systematically than other metal complexes.

The merit of ZMG is that it is insoluble in water but soluble in mild acidic solutions (e.g. 0·01–0·1 M HCl) such as are present in the stomach contents. It is virtually tasteless in contrast to the sharp metallic taste of soluble zinc salts and also is devoid of the marked astringency and irritancy to the gastric mucosa (observed by scanning electron microscopy—see Results) as well as to the oesophagus observed with $ZnSO_4$ and related salts. It also does not exhibit the mucous denaturing or potent astringent effects of $ZnSO_4$. It is rapidly solubilized in gastric juice < pH 3·5 and on the basis of physiochemical data (Whitehouse et al unpublished data) appears to act as a prodrug.

The results showed, overall, that ZMG exhibited almost uniform anti-ulcer effects in a variety of standard and highly sensitive ulcer models (Figs 1–8). It is evident that while ZMG did not produce total protection or prevention of ulceration this compound is effective without producing any visible gastric irritancy (e.g. as reflected by mucus aggregation and opacification of the mucosa seen with $ZnSO_4$). The protection afforded by ZMG is about comparable with that of cimeditine but not as potent as misoprostol against NSAID-induced ulceration (Figs 2, 3, 4b) when compared on a molar basis. ZMG has the advantage over misoprostol, however, in not producing any evident diarrhoea as seen with the latter drug (unpublished observation).

In the ethanol ulcer model, a precise relationship was observed between the timing of the administration of ZMG and its ulcer preventive effects. The results of studies of ZMG protection against NSAID-induced gastric lesions showed that this agent will only protect against mucosal damage induced by the orally-administered drugs and not that when the drugs are given by the parenteral route. This may be of importance in delineating the effects of NSAIDs on the local microcirculation in the mucosa from that produced by parenteral dosage, since microvascular damage is most evident when the NSAIDs are given orally (Rainsford & Willis 1982; Rainsford et al 1982, 1984, 1985).

The lack of protective effects of ZMG against the gastro-irritancy induced by a range of propionic acids, in contrast to the positive effects against other NSAIDs, was most puzzling (Fig. 4). There can be no effect on the metabolism of the inactive *R*(−) enantiomers to their active *S*(+) forms since the irritant effects of *S*(+)-naproxen were, like other propionates, unaffected by ZMG. It is possible that ZMG could have enhanced the absorption of the propionic acids as this may have accounted for their slightly enhanced acute anti-inflammatory effects when given with ZMG (Fig. 9). Whatever the mechanisms underlying these exceptional effects, the results highlight one limit to the range of protection afforded by this complex against NSAID-induced gastric damage.

The protective effects of ZMG against reserpine-induced ulcers (Figs 6, 7) could derive from the membrane-stabilizing properties of zinc ions (Pfeiffer et al 1980, 1987) especially those involving stabilization of mast cells (Cho & Ogle 1977; Ogle & Cho 1978; Barbarino et al 1988).

The lack of effects of ZMG given parenterally in the reserpine model contrasts with the effects of $ZnSO_4$ which Ogle & Cho (1978) showed had protective effects when given intraperitoneally, and which was confirmed here. This difference might be due to the limited bioavailability of zinc from the monoglycerolate complex in the neutral pH of the peritoneal cavity. The low pH in the stomach should favour

controlled dissociation of zinc from ZMG, not achieved under neutral pH conditions (Whitehouse et al unpublished data) such as evident at the injection site in the peritoneal cavity. The lack of irritancy of ZMG when given parenterally is in contrast to marked irritation by the zinc salts (Whitehouse et al 1990) and is further evidence for the limited or controlled release of zinc from the monoglycerolate complex.

The results show that ZMG has anti-ulcer activity against NSAID-induced muscosal damage only when given by the oral route. When this complex is given with aspirin or other NSAIDs there is no compromising of the anti-inflammatory activity of these drugs (Table 1, Fig. 9). Thus ZMG may have therapeutic value as a mucosal protective agent, i.e. in preventing gastric ulcers induced by non-propionic acid NSAIDs, and as well an agent for control of other ulcers.

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