A simple method for measuring thickness of the mucus gel layer adherent to rat, frog and human gastric mucosa: influence of feeding, prostaglandin, N-acetylcysteine and other agents

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(Received 5 November 1981; accepted 3 March 1982)

Summary

1. A technique has been developed for measuring thickness of the gastric surface mucus gel layer. Mucosal sections (1-6 mm) were cut from frog and rat stomach and human antrum, mounted transversely and viewed by an inverse microscope (x 200 magnification) under dark field illumination or phase contrast. The mucus layer was readily distinguishable and its dimensions could be recorded by means of an eyepiece graticule.

2. Mean mucus gel thickness in rat, frog (Rana temporaria and Rana pipiens) and human was 73, 76, 55 and 192 μm respectively. However, there was variation in the average thickness of the gel layer between individual mucosae from the same species (up to twofold). Mucus thickness between adjacent regions of the same mucosal section also varied markedly (up to tenfold).

3. Topical administration of 16,16-dimethylprostaglandin E₂ by oral intubation caused a significant increase in thickness in both rat and frog at doses of 5 μg/ml and 0.5 μg/ml respectively. Feeding and exposure of the mucosa to N-acetylcysteine (10–20%, w/v) produced variable effects, whereas pepsin (1 mg/ml) caused a marked reduction in thickness of the surface gel layer in both rat and frog.

4. The technique provides a rapid and simple method for determining gastrointestinal mucus thickness in relation to mucosal morphology. It is ideally suited for studying the control of mucus secretion and effect of drugs.

Key words: N-acetylcysteine, 16,16-dimethylprostaglandin E₂, feeding, gastric mucus, gel thickness, pepsin, shear.

Introduction

Gastrointestinal mucus is secreted to form a gel which adheres to the mucosal surface. Surface gel is considered to protect the mucosal cells against shear forces during digestion and, in the stomach, it acts as a barrier to large molecules such as luminal pepsin [1]. Recent studies on HCO₃⁻ transport by gastric and duodenal epithelial cells [2] have re-awakened interest in the role of mucus gel as a mixing barrier in which surface neutralization of luminal acid can occur [3]. The protective properties of such a mucus-bicarbonate barrier would depend not only on gel structure but may also depend on the amount or thickness of the layer covering the mucosal surface. Structural studies have demonstrated that the native mucus glycoprotein is broken down by pepsin into lower molecular weight components with consequent solubilization of the surface gel and release of glycoprotein subunits into the gastric lumen [4, 5]. Thus the thickness of the surface gel represents a dynamic balance between the rate of mucus secretion and its erosion by peptic digestion and shear [1, 3].

A variety of methods have been used in an attempt to measure mucus production in vivo: these include incorporation of radioactive glycoprotein precursors, radioimmunoassay and determination of mucus glycoprotein by chemical
methods (see Allen [1] for review). However, application of these techniques to soluble mucus, e.g. luminal aspirates, makes it difficult to interpret results in terms of the amount of gel present on the mucosal surface. Two approaches have specifically investigated the surface mucous gel, namely an Alcian blue binding method [6] and the use of a slit lamp and pachymeter [7]. In this paper we describe a method for measuring surface mucous gel thickness which has the following virtues: it is rapid, many measurements can be made on the same sample of mucosa, and the detailed topology of the gel surface can be visualized. The thickness of mucus gel adherent to the surface of rat and amphibian gastric and human antral mucosae has been measured and the effects of various agents have been investigated.

Methods

Preparation of tissues

Male Wistar rats (180–220 g) and two species of frog (Rana temporaria and Rana pipiens) were used. Human antral mucosa was obtained during surgical resection of two stomachs, one for gastric carcinoma and the other for pyloric stenosis. Except when the effects of feeding were being determined, animals were starved for 48 h before being killed. Rats were killed by cervical dislocation and frogs by sectioning the spinal cord and pithing. The stomach was removed immediately and opened along the lesser curvature. Where the animals had been fed, food particles were removed by gentle washing in NaCl solution (150 mmol/l). Outer muscle layers were separated by blunt dissection (frog and human) or blistering (rat). The mucosa, luminal surface up, was mounted on a Millipore filter and sections were cut with a pair of parallel sharp razor blades separated by a distance of 1·6 mm. Except when being sectioned, tissues were kept immersed in NaCl solution (150 mmol/l) (rat and human) or frog Ringer (amphibia). Sections were mounted transversely between two cork strips (1 cm apart, to support the section) on a Perspex base (Fig. 1). Positioning of sections was checked stereoscopically (× 3·5 magnification) to ensure they were not mounted obliquely. Throughout the above procedures, extreme care was taken not to distort the mucosa by stretching or compression. The whole procedure took less than 15 min from death of the animal till mounting of the section.

The effects in vitro of hyperosmolarity (NaCl, 2 mol/l), N-acetylcysteine (200 mg/ml; 20%, w/v) and pepsin (1 mg/ml in citrate buffer, pH 2·2 [4]) were determined before sectioning. The flat mucosa (luminal surface up) was exposed to 10 ml of solution stirred mechanically and left in contact with the tissue at room temperature (≈ 20°C) or at 37°C for periods of 5–60 min. Stirring was by means of an overhead propeller rotating at 50 Hz (Serval Omnimixer). To determine the effect of feeding, rats were fed normally before being killed and frogs fed with house flies during the preceding 48 h period. The influence of 16,16-dimethylprostaglandin E₂ (dmPGE₂, 0·5 μg/ml in the frog and 2–10 μg/ml in the rat) and N-acetylcysteine (100 and 200 mg/ml in all species) were determined after administration via an orogastric tube in a volume of 0·5 ml instilled 2 h before sacrifice.

Measurement of thickness

Surface mucus thickness was measured by means of an inverse microscope (× 200 magnification) focused on the tissue to reveal three distinct regions under light and dark field illumination or phase contrast. These were interpreted as solution, mucus gel layer and mucosa (Figs. 1, 2). Distance between the solution–mucus interface and mucus–epithelial surface interface was recorded by using an eyepiece graticule. In general, mean mucus thickness of each stomach was computed from a minimum of 18 measurements (six readings for each of three to six sections). Measurements of gel thickness were taken at intervals of approximately 500 μm along the section. After thickness had been recorded, the bathing solution was replaced with NaCl solution (2 mol/l), absolute ethanol or N-acetylcysteine (20%, w/v), and readings were taken at times between 1 min and 2 h after addition of these agents. Student’s t-test for unpaired variables was used for all statistical evaluations (P < 0·05).

Validation of measurement technique

The following experiments were performed to validate the technique.

1. Measurement of mucus thickness from both sides of the section: after the first series of readings, the section was turned over longitudinally through 180° and remounted to record thickness from the other side (Fig. 3).

2. Successive measurements of surface gel thickness were recorded from the same section up to 60 min after preparation.

3. Mucus thickness was determined in sections of different thickness (0·8, 1·6 and 2·4 mm) obtained from the same mucosal piece.
FIG. 1. Diagram to show the preparation method for the cutting and positioning of mucosal sections. Full details are given in the Methods section.

FIG. 2. Appearance of a rat gastric mucosal section viewed under dark field in an inverse microscope (×70 magnification). Three distinct phases comprising solution, mucus gel layer and mucosa can be clearly seen. Note also the marked variation in thickness of the surface gel layer between different regions of the section.

(4) Three adjacent sections of equal thickness (1.6 mm) were cut simultaneously from a piece of mucosa by using four parallel razor blades fixed together.

(5) Mucus thickness was measured in sections cut from frog and rat mucosae with their muscle layers intact. These sections were compared with similar sections from mucosae that had their muscle layers removed by blunt dissection (frog) or blistering (rat).

(6) Three sections were cut from the same mucosa, one section with two razor blades placed at 90° to the mucosa surface. The other two sections were cut with the razor blades at an oblique angle (about ±10°) to the mucosa either side of vertical.

**Experimental solutions**

For routine preparation of mucosae and measurement of thickness, NaCl solution (150 mmol/l) was used for the rat and an unbuffered frog Ringer solution for the amphibia. For experiments in vivo the following solutions were administered: NaCl (150 mmol/l); 16,16-dimethylprostaglandin E₂ (0.5–10 μg/ml in phosphate buffer (150 mmol/l), pH 7.4; gift from Dr J. Pike, Upjohn Co., Kalamazoo, U.S.A.] N-acetylcysteine (10%, w/v); NaCl (150 mmol/l). For experiments in vitro the following solutions were used: NaCl (150 mmol/l); NaCl (150 mmol/l)/N-acetylcysteine (20%, w/v); phosphate buffer (200 mmol/l), pH 7.4; N-
FIG. 3. Mucus gel thickness measured at intervals of approximately 500 µm across a section of mucosa from a starved frog (Rana temporaria). Note (i) the wide variation between individual points on the same section and (ii) that measurements from opposite sides of the same section show good agreement; after the first series of readings (direction A → B), the section was turned over longitudinally through 180° and remounted to record thickness from the other side (direction B ← A). See also Table 2.

Results

General observations and validation of technique

In all preparations, the surface gel layer was optically distinct from the dense mucosa and clear bathing solution (Fig. 2). Indian ink added to the bathing solution was observed to settle at the mucus gel–solution interface and did not penetrate the gel itself. In unfixed sections, periodic acid/Schiff reagent (PAS) stained the mucosa intensely. The surface gel layer stained a light pink and the solution was clear and the submucosa unstained. The thickness of the mucus layer was unchanged by staining with PAS. When the mucosa was sectioned, fixed in ethanol and stained with haematoxylin and eosin, acetylcysteine (20%, w/v); citrate buffer (200 mmol/l), pH 2.2/N-acetylcysteine (20%, w/v); pepsin [1 mg/ml in citrate buffer (200 mmol/l), pH 2.2] [4].

acetylcysteine (20%, w/v); citrate buffer (200 mmol/l), pH 2.2/N-acetylcysteine (20%, w/v); pepsin [1 mg/ml in citrate buffer (200 mmol/l), pH 2.2] [4].
the mucosa showed normal morphological appearance and only occasional patches of surface mucus gel were observed.

There was no significant difference in mucus gel thickness measured at various times after sectioning at room temperature (frog and rat) or 37°C (rat) (Table 1). The quantitative variation in thickness between different sites from the same section is illustrated in Fig. 2 for rat and shown for frog in Fig. 3. Comparison of thickness measurements made from opposite sides of the same mucosal section for both individual read-
ings (Fig. 3) and overall means (Table 2) do, however, show good agreement. This confirmed that the section had been mounted transversely and that the mucus gel was not viewed at an oblique angle. Sections were cut with the razor blade always placed 90° to the mucosal surface as judged by eye. To ensure that the placing of the razor blades at 90° to the mucosa within the limits of observation was not critical, three sections were cut from the same piece of rat mucosa with the razor blades placed at 90° and at opposite imposed, oblique angles (about ±10°) to its surface. No difference was found between the three sections with means (±SEM) of 98 ± 5 (−10° from normal), 102 ± 6 (normal) and 88 ± 4 (+10° normal), n = 7 sections. No significant difference was found between mucus thickness measured in sections of different width cut from the same mucosal piece or between three adjacent sections cut simultaneously (Table 3). This showed that cutting did not distort the mucus gel.

By stripping the muscle layers before sectioning much better adhesion to the Millipore filter was obtained. Sections taken from frog and rat stomachs without the muscle layer having first been removed had a mean (±SEM) mucus gel thickness of 77 ± 5 µm (15 sections) and 94 ± 5 µm (six sections) respectively. These results show that the removal of the muscle layers does not significantly affect the gel thickness measurements in the frog. In the rat the value is somewhat higher than the value found in stripped sections (73 µm, Table 4) but in this species the technique was very difficult with the muscle layers intact. Nevertheless the results are within the variation found between individual rat stomachs.

### Table 1. Gastric mucus gel thickness measured at various times after sectioning

<table>
<thead>
<tr>
<th>Species</th>
<th>Temperature (°C)</th>
<th>n</th>
<th>Mean thickness (µm)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Time (min)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Rat</td>
<td>20</td>
<td>7</td>
<td>71 ± 5</td>
</tr>
<tr>
<td>Rat</td>
<td>37</td>
<td>9</td>
<td>86 ± 6</td>
</tr>
<tr>
<td>Frog</td>
<td>20</td>
<td>7</td>
<td>87 ± 7</td>
</tr>
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<table>
<thead>
<tr>
<th>Species</th>
<th>Temperature (°C)</th>
<th>n</th>
<th>Mean thickness (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frog</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Renal</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Rana</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>temporary</td>
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<td></td>
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<tr>
<td>Rana</td>
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<tr>
<td>piperiens</td>
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</tbody>
</table>

### Table 2. Comparison of mucus gel thickness measured from either side of gastric mucosal sections

<table>
<thead>
<tr>
<th>Species</th>
<th>Temperature (°C)</th>
<th>n</th>
<th>Mean thickness (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rat</td>
<td>20</td>
<td>7</td>
<td>71 ± 5</td>
</tr>
<tr>
<td>Rat</td>
<td>37</td>
<td>9</td>
<td>86 ± 6</td>
</tr>
<tr>
<td>Frog</td>
<td>20</td>
<td>7</td>
<td>87 ± 7</td>
</tr>
</tbody>
</table>

### Table 3. Comparison of mucus gel thickness from (a) sections of different width and (b) three adjacent sections cut simultaneously from the same piece of mucosa

<table>
<thead>
<tr>
<th>Method</th>
<th>Section width (mm)</th>
<th>Mean thickness (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(a) Sections of different width from one mucosa</td>
<td>0-8</td>
<td>63 ± 4</td>
</tr>
<tr>
<td>(b) Sections cut adjacent and simultaneously from one mucosa</td>
<td>1-6 (outer)</td>
<td>50 ± 4</td>
</tr>
</tbody>
</table>

**Gastric mucus gel thickness**
Table 4. Mucus gel thickness in different species

Mean mucus gel thickness (µm) is given together with the percentage distribution of individual measurements taken. At least six measurements per section and three sections per stomach were taken (i.e. minimum of 18 measurements per stomach). 16,16-Dimethylprostaglandin E₂ (dmPGE₂) was administered by orogastric tube in phosphate buffer (150 mmol/l); 0.5 µg/ml (Rana pipiens) and 10 µg/ml (rat) 2 h before animals were killed. Overall mean values were calculated from individual sections.

<table>
<thead>
<tr>
<th>Species</th>
<th>No. of stomachs</th>
<th>No. of measurements</th>
<th>Mean mucus gel thickness (µm)</th>
<th>Distribution of measurements of mucus gel thickness (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>5-50 µm</td>
</tr>
<tr>
<td>Rat</td>
<td>6</td>
<td>247</td>
<td>73 ± 5</td>
<td>57</td>
</tr>
<tr>
<td>Rat (after 10 µg of dmPGE₂)</td>
<td>4</td>
<td>155</td>
<td>139 ± 7</td>
<td>19</td>
</tr>
<tr>
<td>Frog (Rana temporaria)</td>
<td>6</td>
<td>203</td>
<td>75 ± 5</td>
<td>50</td>
</tr>
<tr>
<td>Frog (Rana pipiens)</td>
<td>7</td>
<td>167</td>
<td>55 ± 5</td>
<td>69</td>
</tr>
<tr>
<td>Frog (Rana pipiens) (after 0.5 µg of dmPGE₂)</td>
<td>4</td>
<td>89</td>
<td>71 ± 4</td>
<td>54</td>
</tr>
<tr>
<td>Human</td>
<td>2</td>
<td>103</td>
<td>192 ± 7</td>
<td>11</td>
</tr>
</tbody>
</table>

Fig. 4. Effects of oral administration of 16,16-dimethylprostaglandin E₂ (dmPGE₂) and N-acetylcysteine (N-Ac) on mucus gel thickness in rat and frog (Rana pipiens). Control animals received 0.5 ml of NaCl solution (150 mmol/l) by oral intubation. There was a significant increase in gel thickness measured 2 h after administration of dmPGE₂ in rat (5 and 10 µg/ml) and frog (0.5 µg/ml); N-acetylcysteine (10%, w/v) decreased thickness of the mucus layer only in the frog (*P < 0.05). The increase with 2 µg/ml of dmPGE₂ in the rat was not significant.

Effect of feeding

Mean thickness of the surface mucus gel in rats feeding normally up until the time of being killed (48 ± 5 µm) was 34% less compared with starved animals (73 ± 5 µm). However, feeding Rana pipiens increased mean mucus thickness by 73%, from 55 ± 5 µm after starving to 95 ± 8 µm. These mean values (±SEM) were a minimum of six readings per section and three sections per stomach from six animals. Although opposite in effect, both changes were statistically significant (P < 0.05). When opened, the stomachs of fed rats were observed to be completely full whereas frog stomachs contained considerably less material.

Effect of topical administration of prostaglandin and N-acetylcysteine in vivo

In starved rats and frogs receiving 0.5 ml of NaCl solution (150 mmol/l) (control), mucus gel thickness was not statistically different from measurement in animals which were not dosed with vehicle. In the rat, dm-PGE₂ (2-10 µg/ml) caused a dose-dependent increase in thickness which was significant at concentrations of 5 and 10 µg/ml (Fig. 4). In the frog, a concentration of 0.5 µg/ml of this prostaglandin was sufficient to produce a significant increase in gel thickness. The distribution of individual measurements at the highest doses of dm-PGE₂ (10 µg/ml (rat) and 0.5 µg/ml (frog), Table 1] shows that mucus thickness increases overall and not just in the thick or thin areas. N-Acetylcysteine (10%, w/v) did not influence mucus thickness in rats measured 2 h after administration via an orogastric tube, although a small, but statistically significant, decrease in thickness occurred in frogs subjected to the same treatment (Fig. 4).

Studies in vitro

Mucus gel thickness in both rat and frog was not altered when the luminal side of the mucosa
(before sectioning) was exposed to a solution of NaCl (150 mmol/l) stirred mechanically at room temperature for 60 min. A similar lack of effect was obtained when the mucosa was bathed in hyperosmolar NaCl solution (2 mol/l) and the solution stirred as above. When the mucosa from both species was exposed at room temperature to N-acetylcysteine (20%, w/v)/NaCl solution (150 mmol/l) and the same shear force applied, the depth and appearance of the mucus gel did not alter up to 30 min, but after further exposure it became granular in appearance and lost adherence to the mucosal surface. The effect of temperature and pH in this system was investigated by exposing isolated mucosa at 37°C for 30 min to the following solutions, with stirring: (1) N-acetylcysteine (20%, w/v)/citrate buffer (200 mmol/l), pH 2-2; (2) N-acetylcysteine (20%, w/v)/phosphate buffer (200 mmol/l), pH 7-4. Under these conditions there was no decrease in thickness or change in appearance of the mucus layer. Without stirring, no change was seen in the mucus gel layer which had been exposed to N-acetylcysteine (20%, w/v)/NaCl solution (150 mmol/l) for 60 min. After just 10 min exposure to pepsin [1 mg/ml in citrate buffer (200 mmol/l), pH 2-2] with stirring, the mucus layer became granular in appearance with some reduction in thickness and within 20 min almost all the surface gel had been removed. When sections were exposed to absolute ethanol, the surface mucus gel layer was observed to disappear within 5 min.

Discussion

Evidence that the mucus gel layer could be observed directly by microscopy under the present experimental conditions is indicated as follows: (i) three distinct layers were always visible (mucosa, gel and solution) under both light and dark field illumination or phase contrast; (ii) the mucus layer had a distinct optical appearance; (iii) in contrast to the bathing solution, the gel was not penetrated by Indian ink particles; (iv) the mucus gel stained lightly and the underlying mucosa stained intensely with PAS (positive for mucus glycoproteins). The depth of gel was independent of section thickness over the range 0.8-2.4 mm, reproducible for three adjacent sections obtained from the same mucosal sample and reproducible when estimated from either side of the section. Furthermore, mucus thickness did not vary with time up to 60 min after preparation of sections. These results indicate that flow properties of the gel do not pose a problem since the mucus layer adherent to the epithelial surface remains sufficiently immobile to enable reproducible measurements of its thickness to be obtained. The wide variations in the thickness of the gel observed across sections of both untreated and prostaglandin stimulated mucosa is also consistent with its comparative lack of flow.

Two observations of major interest emerge from the present study. Firstly, the gel layer is relatively shallow, with a mean thickness of less than 100 μm in the rat and frog and about 200 μm for human. It should be noted that the value for human mucus is for abnormal mucosa and may not be a reflection of the true thickness in the healthy subject. Secondly, variations in gel thickness are considerable, differing by tenfold on a single sample of mucosa. In one or two extreme cases, minimum thickness was 5 μm and maximum values were 400 μm for a single mucosa. These results differ from measurements reported by Bickel & Kauffman [7] using a slit lamp and pachymeter, who reported mean values of 166 and 652 μm for rat and human gastric gel thickness respectively. One factor which may contribute to this discrepancy relates to the fact that rats used in the study by Bickel & Kauffman [7] were pylorus-ligated 1-2 h before measurements were taken. In addition to preventing mucus flow through the pylorus, vagal activation may stimulate mucus secretion via a cholinergic mechanism [8, 9]. In estimating gel thickness by an image-splitting technique the refractive index of mucus is assumed to be the same as that of water [7]. Solute and cellular material would alter refractive index and could therefore affect the quantitative assessment of mucus thickness by this technique. Furthermore the image-splitting technique does not appear to detect the wide variations in gel thickness over a single mucosa that we have observed. As well as enabling direct visualization of the mucus gel layer, a distinct practical advantage of the method reported here is that a large number of measurements can be made rapidly at different sites and on many tissue samples.

Prostaglandins have been shown to increase amounts of luminal mucus glycoprotein by a number of workers [6, 10, 11]. Direct evidence for an increase in surface gel thickness after topical dmPGE₂ in the absence of increased luminal glycoprotein was obtained by using image-splitting optics in the rat [7]. Our results with the same prostaglandin analogue confirm these observations although the magnitude of the increase (74% compared with 140% at 10 μg/ml in rats) was less in the present study. However,
pyloric ligation would itself result in administered compounds remaining in contact with the mucosa for a longer period of time compared with those in our study. Using an Alcian blue assay, Bolton et al. [6] reported no increase in mucus gel after topical E-type prostaglandin in rats although they did report an increase in luminal glycoprotein. We observed a viscous solution in the rat gastric lumen after dmPGE₂ administration. There is now considerable evidence that prostaglandins cause an increase in mucus output but how this is reflected in terms of increased mucus glycoprotein in the lumen or mucus gel on the surface probably depends on the conditions under which experiments are conducted. The cytoprotective action of prostaglandins is well documented [12] and their stimulation of mucus secretion may play a part in this mechanism.

After feeding, there was a decrease in mucus thickness (34%) in rats and an increase in thickness (72%) in frogs when compared with mucus thickness in starved animals. A possible explanation for these opposite effects is that although feeding may stimulate mucus output in both cases, in the rat the large amount of food present in the stomach at the time of killing would impose a shear force which would erode the gel and thereby decrease its depth.

Rapid removal of surface mucus by pepsin is consistent with the documented effect of proteolysis in solubilizing the gel [4, 5]. In contrast, exposure of the surface gel to N-acetylcysteine (20%, w/v) in vitro was considerably less dramatic and the gel was removed only after 60 min in the presence of a continuous shear force. No detectable loss of surface mucus gel occurred when the rat mucosal surface was exposed, with continuous stirring, to N-acetylcysteine at either pH 2-2 or pH 7-4 and at 37°C. After instillation of N-acetylcysteine (10%, w/v) in vivo there was no reduction in the mucus gel thickness adherent to rat gastric mucosa and there was only a small, but significant, decrease in thickness in the frog. Detailed structural studies demonstrate that exposure to sulphhydryl reagents for 20 h or more causes complete breakdown of gastric mucus glycoproteins and solubilization of the gel [1]. However, exposure of native pig gastric mucus gel to 20% N-acetylcysteine for 30–60 min did not appreciably reduce the volume of gel measured after centrifugation (unpublished observations). A note of caution to arise from these findings is that effects observed at the mucosal surface on short-term exposure to N-acetylcysteine or other sulphhydryl reagents should not necessarily be attributed to breakdown of the mucus gel structure. The rapid disappearance of the surface mucus gel with absolute ethanol is compatible with denaturation of mucus by this agent and explains the absence of observable mucus gel in sections prepared by standard procedures of histological fixation.

The values for mucus gel thickness in our experiments are less than any of the previously assumed or measured dimensions. The greater the thickness of the gel then the better will be the protection it affords the mucosal surface against shear and proteolysis. However, while there is a finite thickness of mucus gel on the surface, its properties as a mixing barrier, as opposed to a diffusion barrier, need not necessarily be impaired. Under these conditions surface neutralization as postulated [3] would occur close to the mucosal surface and HCO₃⁻ would still be prevented from mixing with bulk acid in the lumen. In this respect it should be emphasized that recent evidence shows gastric HCO₃⁻ transport to be an anion-exchange process and should not strictly be considered as a secretion with a defined volume [2]. The results reported here also suggest that the pH gradient demonstrated across the mucus gel [13] may occur over smaller distances than previously supposed. Although at some points the mucus gel layer was thin it nevertheless appeared to form a continuous cover over the mucosal surface.

Acknowledgments

We thank Dr R. McBurney for introducing us to the technique, Dr D. Halley and Mr E. Waggett for histological processing and Dr O. Flint for preparation of the photomicrographs. Miss S. Kerss is supported by the Science Research Council.

References


