Luminal distension as a possible consequence of experimental intestinal perfusion

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SUMMARY In an experimental jejunal perfusion study, distress in healthy subjects occurred during eight out of 16 perfusions in which intestinal secretion was provoked. Calculation demonstrates the volumetric consequences of inadequate recovery of secretory perfusates, and analysis of the perfusion studies shows that distress was significantly associated with poor recovery of the perfusate. These observations are pertinent to increasing interest in the phenomenon of intestinal fluid secretion.

The development of intestinal intubation techniques since the work of Abbott and Miller (1936) has allowed extensive study of intestinal absorption in man. Until recent years, such perfusion studies have usually involved the perfusion of physiological electrolytes or nutrients into the intestine (Fordtran and Ingelfinger, 1968), and hazards associated with such perfusions have been rarely reported. Even the use of balloons obstructing the lumen of the intestine seems to be a safe procedure except in the presence of intestinal disease (Schmid, Phillips, and Summerskill, 1968) although pancreatic duct obstruction by the balloon has occurred (J. R. Malagelada, personal communication). Recently, there has been interest in the perfusion of solutes which, at low concentrations, inhibit intestinal water absorption or provoke intestinal water secretion; such substances include laxatives, hydroxy fatty acids, and bile acids. In a recent study of the effect of glycine-conjugated bile acids on jejunal absorption in man (Wingate, Phillips, and Hofmann, 1973), it was briefly reported that perfusions of bile acids which induced net secretion were in some cases associated with unpleasant symptoms in healthy subjects. However, further instances of adverse effects from perfusions with bile acids (E. Krag and S. F. Phillips, personal communication), bisacodyl (P. Czybyski and K. Ewe, personal communication) and prostaglandins (C. Matuchansky, personal communication) have come to our attention, suggesting that this phenomenon is likely to occur in the presence of secretory perfusions.

The introduction of a physiological fluid (perfusate) into the intestinal lumen is implicit in perfusion studies, as is the subsequent sampling of the luminal contents. This sampling, commonly by siphonage, rarely (if ever) allows total recovery of the perfusate. With solutions which will be absorbed, incomplete recovery may be of no consequence, but this may not be true for studies in which net fluid secretion into the intestinal lumen is provoked, especially when the secretogenic agent is a solute in the perfusate. We are not aware of any previous quantitative analysis of the consequence of incomplete perfusate recovery. The clinical effects of the perfusions reported here were associated with poor recovery of secretogenic perfusates, and calculation supports the hypothesis that these phenomena were causally related.

Methods

Proximal jejunal segments of healthy adult volunteers who had given informed consent were perfused with glucose electrolyte solutions containing 0, 2.5, 5.0, or 10.0 mM/l concentrations of glycine-conjugated bile acids, by a double-lumen technique (Phillips and Summerskill, 1968). A detailed account of the solutions and methods employed has been given elsewhere (Wingate et al, 1973). Each subject received four consecutive 90-minute perfusions, each perfusion containing one of the bile acid concentrations in a sequence determined by a Latin square design. The bile acids employed were glycocholic acid (GC), glycochenodeoxycholic acid (GCDC), and glycodeoxycholic acid (GDC).

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NET WATER MOVEMENT
Distal sampling from the perfusion system was collected and the collections were divided into 10-minute aliquots. The perfusate contained polyethylene glycol (PEG) labelled with 14-carbon as a volume indicator and the change in isotope concentration between perfusate and aspirate was used to calculate the change in volume in the perfused segment (Wingate, Sandberg, and Phillips, 1972). The values taken for net water movement were for the last four 10-minute periods in each perfusion when it was assumed that steady state conditions had been achieved.

PERCENTAGE RECOVERY
The percentage recovery of the perfusate was obtained by expressing the volume recovered by distal sampling as a percentage of the volume perfused over the same 10-minute period. The mean percentage recovery for each perfusion was calculated for the entire 90-minute period of each perfusion.

Results
NET WATER MOVEMENT
The changes in net water movement induced by the different concentrations of glycine-conjugated bile acids are shown in table I. The trihydroxy bile acid GC had little effect on net water movement but both GDC and GCDC induced secretion or arrested absorption at 5 and 10 mM/l concentrations.

ADVERSE EFFECTS
Adverse effects were malaise, vomiting, non-specific abdominal pain, and diarrhoea. These occurred only during perfusion with 5 and 10 mM/l dihydroxy bile acids, and their distribution in these perfusions is shown in table II. In two of the perfusions, the adverse effects were severe enough to compel medication (pethidine intramuscularly) and the termination of the perfusion before the end of the 90-minute period. All adverse effects were rapidly relieved within a few minutes of replacing the bile-acid perfusate with a control (bile acid-free) solution.

Since adverse effects only occurred with dihydroxy bile acid concentrations which arrested absorption or provoked net secretion, it was assumed that subjects who experienced discomfort would show greater net secretion rates. In fact, within experimental groups, there was no correlation between the magnitude of net secretion and the occurrence of adverse effects. It was this latter finding that prompted scrutiny of the variation in recovery of perfusates.

PERCENTAGE RECOVERY OF PERFUSATES
Percentage recoveries for perfusions of dihydroxy bile acids in concentrations associated with adverse effects were calculated, and some significant differences were found between 'toxic' and 'non-toxic' perfusions. These are shown in table III. In this context, 'toxic' merely denotes the association of adverse effects with a perfusion.

A significant difference ($p < 0.02$) was found between recoveries of toxic and non-toxic perfusates. The data provide an explanation for the otherwise baffling phenomenon of toxicity with GCDC perfusates, when, as shown in table I, the effect of GCDC on absorption is less than that of GDC. Recovery of GCDC perfusates was, overall, significantly less than recovery of GDC perfusates. It is also clear that when toxicity was associated with 5 mM/l perfusates (in three perfusions) recovery was very poor.

### Table I  Net water movement during jejunal perfusion with different concentrations of glycine-conjugated bile acids

<table>
<thead>
<tr>
<th>Concentration of Bile Acid (mM)</th>
<th>GC (± SEM)</th>
<th>GCDC (± SEM)</th>
<th>GDC (± SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 (control)</td>
<td>2.1 ± 0.1</td>
<td>1.8 ± 0.3</td>
<td>2.0 ± 0.4</td>
</tr>
<tr>
<td>2.5</td>
<td>1.9 ± 0.4</td>
<td>1.2 ± 0.2</td>
<td>1.1 ± 0.2</td>
</tr>
<tr>
<td>5.0</td>
<td>2.0 ± 0.5</td>
<td>0.3 ± 0.2</td>
<td>-0.8 ± 0.2</td>
</tr>
<tr>
<td>10.0</td>
<td>1.7 ± 0.1</td>
<td>-1.3 ± 0.1</td>
<td>-2.3 ± 0.4</td>
</tr>
</tbody>
</table>

### Table II  Toxicity of glycine-conjugated bile acids

<table>
<thead>
<tr>
<th>Subject</th>
<th>Toxic Sign with 5-0 mM</th>
<th>Toxic Sign with 10-0 mM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glycochenodeoxycholic</td>
<td>Vomiting, pain</td>
<td>Vomiting, abdominal pain</td>
</tr>
<tr>
<td>Glycodeoxycholic</td>
<td>Vomiting, abdominal pain</td>
<td>Vomiting, abdominal pain</td>
</tr>
</tbody>
</table>

1 Water movement expressed as ml/min absorbed (+ ve) or secreted (- ve) in 25 cm segment of jejunum. Each value is mean ± SEM of perfusion in four subjects.

2 Required medication
The cause of poor recovery was obvious during the conduct of the experiment. Five and 10 mM/l dihydroxy bile acid perfusions provoked considerable secretion of mucus, and this mucus caused intermittent obstruction of the siphonage tube. Even continuous vigilance by the experimenters did not always solve the problem, since flow from the siphonage tube is intermittent even when the lumen of the tube is unobstructed.

**Bile Acid Absorption**

Bile acid absorption was minimal, being less than 5% of the quantity perfused in the absence of net fluid absorption (Wingate et al, 1973).

**Discussion**

The cause of the distress induced by dihydroxy bile acid perfusion in healthy subjects was at first obscure. Intraluminal conjugated bile acids are not recognized as a source of abdominal distress, and higher concentrations than those employed here may be found in the normal human intestinal lumen after gallbladder contraction. Reported effects in vitro of bile acids on enterocyte enzyme systems (Pope, Parkinson, and Olson, 1966; Hepner and Hofmann, 1973) do not appear to be of clinical significance. The symptoms experienced by the subjects were not sufficiently specific to indicate a precise pathophysiological origin. Since it was noted that symptoms were only associated with perfusates which either arrested net absorption or induced frank secretion, this prompted an analysis of the volume changes involved.

When a perfusate containing a non-absorbed secretogenic solute is used as a test solution in an intestinal perfusion system, and only partly recovered by siphonage at the end of the test segment, the residual solute will continue to induce secretion distal to the test segment. This distal secretion will continue until the solute is either diluted to a concentration (C') which does not provoke secretion, or removed from the lumen. The extreme case is shown by simple calculation. The data summarized in table I yield the information that C' for GDC is 4.3 mM/l. If 10 ml/min of a 10 mM/l solution of GDC is perfused into the intestine, and the GDC is neither absorbed nor inactivated, the volume will increase until 23 ml/min will pass a point distal to the perfusion point. The assumption underlying perfusion techniques is that distal recovery of the perfusate will prevent this volume expansion. The quantitative consequence of recovering less than 100% of a secretogenic perfusate is illustrated in figure 1. In unit time, a volume \( V_{in} \) is perfused into the intestinal

![Fig 1 Volume changes in the perfusion segment (AB) and the adjacent distal segment (BC) during perfusion of a secretogenic solution into the lumen at A, showing the dependence of the residual volume \( V_C \) at C on the proportion of fluid removed at B. Shading indicates volumes exceeding the volume perfused at A \( V_{in} \).](http://gut.bmj.com/10.1136/gut.15.9.728)
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lumen at point A, and a volume $V_{\text{out}}$ is removed by siphonage at point B. AB is the perfusion segment, and BC is the adjacent distal segment, equal in length to AB. $V_1$ is the residual volume which passes point C in unit time. Assuming steady state conditions, the percentage recovery (cf table III) is $R$, where

$$R = \frac{V_{\text{out}}}{V_{\text{in}}} \times 100 \quad \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots (1)$$

Figure I shows that in the presence of secretion, even when $R = 100\%$, there is residual perfusate which will increase in volume in segment BC, and as $R$ falls, $V_1$ increases and exceeds $V_{\text{in}}$.

For comparison, fig 2 illustrates the same perfusion geometry during absorption; in this case, no perfusate will reach C until R falls well below 100%. In this instance, the rate of volume change in AB and BC is constant; the problem of computing volume change in the presence of a secretogenic solute is that this is being constantly modified by dilution of the solute, and calculation of $V_1$ must take account of this by solving differential equations for volume changes between AB and BC.

![Diagram](image)

Fig 2 Volume changes in the perfusion segment (AB) and the adjacent distal segment (BC) during perfusion at A of a solution which is absorbed.

Even if our hypothesis is correct, some questions remain to be answered. Since intestinal perfusion is not a new technique, why should fluid distension be a 'new' phenomenon? It is possible that adverse effects of perfusion have occurred in the past, but have been attributed to idiosyncrasy or intolerance of intubation. A more probable explanation is to be found in the fact that the experimental use of perfusates containing secretogenic substances in human small intestine is a recent development. Allied to this, there has been an increasing tendency to perfuse the small intestine with a double-lumen system with an occlusive balloon (Phillips and Summerskill, 1966) rather than with a triple-lumen system with a mixing segment (Whalen, Harris, Geenan, and Soergel, 1966). The controversy over the respective merits of the two systems has concentrated on their accuracy in measuring absorption (Sladen and Dawson, 1968; Fordtran, 1969; Sladen and Dawson, 1970; Modigliani and Bernier, 1971). One difference between the two systems which has not so far received attention, probably because it was thought to be of little or no interest, is that the triple-lumen system has two siphonage tubes, compared to the single siphonage of the double-lumen system. It may be for this reason that Russell and his colleagues (Russell, Allan, Gerskowitch, and Cochran, 1973) apparently did not encounter adverse effects with bile acid perfusions similar to ours, since they combined the use of an occlusive balloon with a mixing segment and a triple-lumen perfusion system.

What significance should be attached to our findings? Our model demonstrates that secretogenic perfusions may generate substantial volumes of fluid in the lumen of the small intestine. Our hypothesis suggests that such volumes can cause discomfort in healthy subjects and also raises the possibility of hazard in the presence of intestinal disease. We conclude that when solutions are perfused into the small intestine in conditions associated with net fluid secretion, more attention should be paid to predicting the consequent volume changes in the intestine; in practical terms this means care in avoiding excess perfusion rates and ensuring adequate perfusate recovery.

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References


