Effect of secretin on plasma motilin in man

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SUMMARY   Graded doses of 0·1, 0·3, 0·9, and 2·7 clinical units/kg/h of pure synthetic secretin were infused over 60 minute periods in six healthy volunteers. Duodenal bicarbonate output and pH were recorded and plasma secretin and motilin levels were measured by radioimmunoassay. During the infusions plasma motilin fell in a dose dependent manner to a nadir of 35%. This fall was linearly correlated with pancreatic bicarbonate output, whereas a non-linear correlation was observed between plasma motilin and both plasma secretin level and duodenal pH. It is suggested that plasma motilin levels are decreased by secretin-induced pancreatic bicarbonate juice flow. This may be important for the control of motilin secretion initiated by duodenal acidification and the concomitant delay in gastric emptying.

Motilin, a recently discovered hormonal peptide from the small intestine (Brown *et al.*, 1971), has been shown to increase lower oesophageal sphincter pressure and to delay gastric emptying in man (Ruppin *et al.*, 1975; Rösch *et al.*, 1976).

So far, no physiological regulation of motilin secretion has been ascertained. In the dog it has been reported that serum motilin levels are raised by strong duodenal alkalinisation (Dryburgh and Brown, 1975). We have shown recently that, in man, motilin is released by duodenal acidification and after intraduodenal instillation of tris buffer (pH 10·2) motilin fell (Mitznegg *et al.*, 1976). As, however, a duodenal pH of 10·2 is unlikely to be achieved under normal conditions, the significance of this finding is uncertain. The present study was designed to assess the effect on motilin release of secretin. This peptide initiates a physiological alkalinisation of the duodenum by stimulating endogenous alkaline pancreatic juice flow (Jorpes and Mutt, 1961).

Methods

Six healthy volunteer men (aged 23-42 years) were

studied after informed consent was obtained. Each subject underwent two tests, each after an overnight fast and separated by one week.

The first experiment was performed in order to establish the effect of graded infusions of secretin on pancreatic bicarbonate output and duodenal pH. The duodenum was drained *via* a modified Lagerlöf tube and duodenal fluid was collected throughout the experiment and corrected for recovery losses by continuous intraduodenal instillation of Co^{58}-labelled vitamin B_{12} as previously described (Tympner *et al.*, 1974). pH was measured conventionally and bicarbonate concentration was determined by titration in an atmosphere of N_2.

After a basal period of 20 minutes, synthetic secretin was infused via a cubital vein catheter into the upper caval vein. Graded doses of 0·1, 0·3, 0·9, and 2·7 clinical units (CU)/kg/h were administered for one hour each. The secretin was synthesised according to Wünsch *et al.* (1972). The material used had a biological activity of about 3·9 CU/μg, identical with the activity of pure natural secretin from G I H Research Unit, Karolinska Institute, Stockholm, Sweden (Lehnert *et al.*, 1973). The reproducibility of the effect of secretin infusions on pancreatic juice flow in the same subjects has been established (Tympner *et al.*, 1974).

In the second experiment the effect of secretin infusions on plasma motilin was studied. The experiment was carried out exactly as described above except that no duodenal fluid was collected. Instead

blood was withdrawn from an antecubital vein at 10 minute intervals during the basal period, at five minute intervals during the secretin infusions and thereafter at 10 minute intervals until 270 minutes. The samples (4·5 ml) were mixed with 1000 kallikrein inactivating units heparinised aprotinin (Trasylol) per ml, immediately centrifuged, and the plasma stored at − 20°C until assay. The immunoassayable plasma secretin was determined as previously described (Bloom, 1975). The results were pooled to calculate the mean plateau levels of secretin for each dose of synthetic secretin infused.

Plasma motilin was measured by a highly specific and sensitive radioimmunoassay. Details of the assay are given elsewhere (Bloom *et al.*, 1976). In brief, antibodies were raised to pure porcine motilin (gift of Professor J. C. Brown) and used at a final dilution of 1 in 320 000 to bind approximately 50% of the added motilin Iodine[125]. There was no cross-reactivity with other known gastrointestinal peptides and changes of 5 pmol/l plasma could be detected with 95% confidence. It has been shown previously that fasting human plasma motilin levels are fairly constant for each individual but vary considerably between subjects (Bloom *et al.*, 1976). In order to put equal weight on each individual's response, the plasma motilin levels in the present study were normalised. In the figures the motilin data are expressed as percentage (± SEM) of the individual mean baseline.

Results

In the present study the mean basal plasma motilin level (± SEM) was 62·2 ± 9·4 pmol/l. During the one-hour infusions of graded doses of 0·1, 0·3, 0·9, and 2·7 CU/kg/h secretin the mean motilin level fell to 50·9 ± 3·6 (p > 0·2), 44·0 ± 3·9 (p < 0·2), 26·0 ± 1·9 (p < 0·01), and 21·8 ± 1·8 (p < 0·002) pmol/l respectively and rose to 42·8 ± 11·1 (p > 0·2) pmol/l 25 minutes after termination of the infusions (p value from Student's t test as compared with baseline level).

It can be seen from Fig. 1 that, whereas at lower infusion rates of secretin, some fluctuation of plasma motilin is observed, at higher doses (0·9, 2·7 CU/kg/h) a stable depression was achieved.

Figure 2 shows the semilogarithmic correlation between plasma secretin levels achieved during the infusions and the concomitant decrease of motilin. A typical S-shaped dose response curve was obtained. It demonstrates a maximal motilin decrease of about 65% corresponding to a plasma secretin level of 163 ± 0·8 pmol/l.

In Fig. 3 the plasma motilin levels are plotted against bicarbonate output and duodenal pH. Whereas bicarbonate output had a linear correlation with motilin inhibition over the range observed (r = 0·99 for mean values), pH had not. The initial fall in motilin was parallelled by a rise in pH though
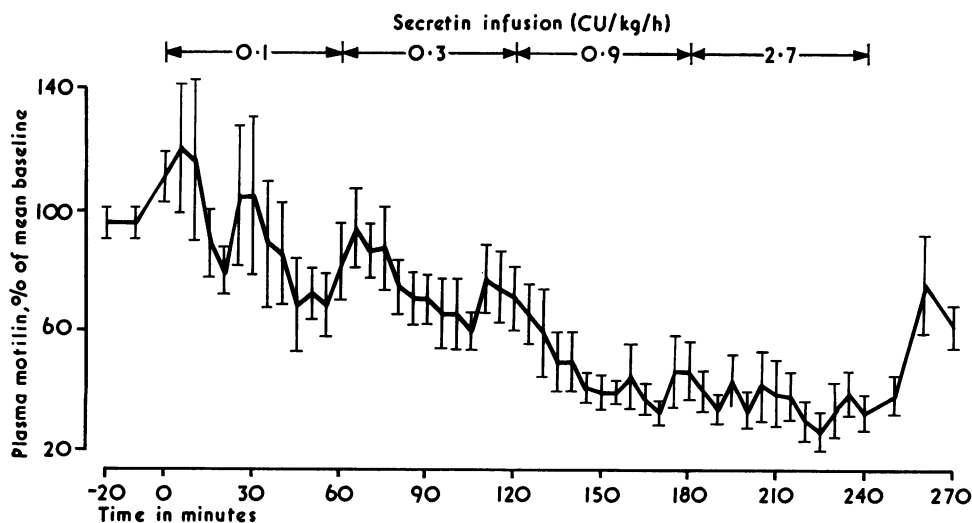

Fig. 1 *Plasma motilin in six volunteers during infusion of synthetic secretin. Motilin values are expressed as a percentage of each individual's baseline (bars = SEM).*
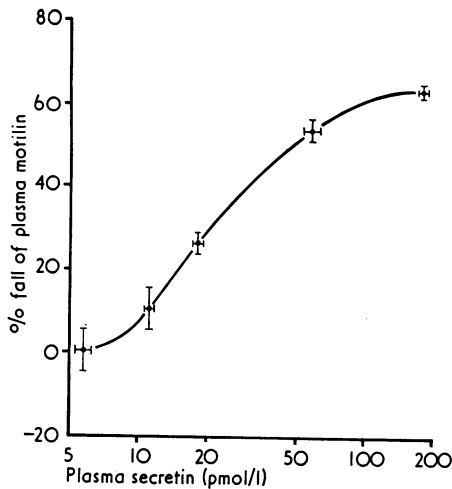
Fig. 2 *Semilogarithmic plot of percentage fall in plasma motilin versus plasma secretin concentrations for six subjects. Each data point represents the mean values of a 60-minute period during infusion of 0·1, 0·3, 0·9, and 2·7 CU/kg/h synthetic secretin, respectively. The vertical line through each point indicates the SEM of percentage of motilin depression and the horizontal line the SEM of plasma secretin.*
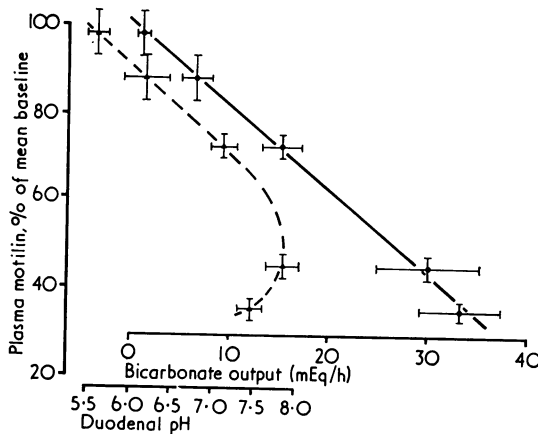

Fig. 3 *Relation of plasma motilin to duodenal pH (---) and bicarbonate output (—) during infusion of synthetic secretin in six subjects. Each data point represents the mean values of a 60-minute period during infusion of synthetic secretin as in Fig. 2. The vertical line indicates the SEM of plasma motilin expressed as percentage of each individual's mean baseline and the horizontal line the SEM of duodenal fluid pH and pancreatic bicarbonate output, respectively.*

at higher levels of bicarbonate output this effect was no longer seen.

Discussion

Our experiments show for the first time that the infusion of graded doses of pure synthetic secretin leads to a dose-dependent decrease of plasma motilin in man.

This finding may be explained as a result of the secretin-induced output of endogenous alkaline pancreatic juice flow: A close linear correlation was observed between bicarbonate secretion and motilin decrease (Fig. 3).

On the other hand the plot of motilin suppression against plasma secretin (Fig. 2) demonstrates levelling off in motilin suppression at 65 %. This occurs at a secretin concentration of approximately 160 pmol/l, at which level a maximal pancreatic bicarbonate output is seen (Haecki *et al.*, 1976). This suggests that the motilin inhibition is probably the direct result of increased bicarbonate production.

It has previously been reported that, in contrast with the dog (Dryburgh and Brown, 1975), alkalinisation of the upper small intestine in man does not produce an increase but a fall in circulating motilin (Mitznegg *et al.*, 1976) and our present results confirm this finding. This may be explained by species differences.

Plasma motilin can be suppressed by the peptide hormone somatostatin (Mitznegg *et al.*, 1977). Recently a direct mechanism has been postulated for the secretin-induced inhibition of gastrin release (Thompson *et al.*, 1972). Therefore we cannot exclude the possibility that the plasma motilin levels were suppressed by a direct action of secretin rather than the secretin-mediated effect of bicarbonate. Further experiments—for example, the investigation of the effect on motilin release of secretin in pancreatectomised subjects—will be needed to obtain conclusive data.

Motilin causes inhibition of gastric emptying (Ruppin *et al.*, 1975). We have shown previously that endogenous motilin is released after duodenal acidification (Mitznegg *et al.*, 1976) and report now that its release may be inhibited by the pancreatic bicarbonate juice flow. One might speculate therefore that motilin is involved in the physiological regulation of gastric emptying and its secretion is controlled by the release of gastric acid and duodenal bicarbonate in response to a meal.

References

Bloom, S. R. (1975). The development of a radioimmunoassay for secretin. In *Gastrointestinal Hormones*, pp. 257-268. Edited by J. C. Thompson. Texas University Press: Austin.

Bloom, S. R., Mitznegg, P., and Bryant, M. G. (1976), Measurement of human plasma motilin. *Scandinavian Journal of Gastroenterology*, 11, suppl. 39, 47-52.

Brown, J. C., Mutt, V., and Dryburgh, J. R. (1971). The further purification of motilin, a gastric motor activity stimulating polypeptide from the mucosa of the small intestine of hogs. *Canadian Journal of Physiology and Pharmacology*, **49**, 399-405.

Dryburgh, J. R., and Brown, J. C. (1975). Radioimmunoassay for motilin. *Gastroenterology*, **68**, 1169-1176.

Haecki, W. H., Belohlavek, D., Bloom, S. R., Demling, L., Domschke, W., Domschke, S., Galariotis, C., Mallinson, C., Mitznegg, P., and Wuensch, E. (1976). Secretin pharmacokinetics and plasma levels for half maximum bicarbonate response in man. *Gut*, **17**, 393.

Jorpes, E., and Mutt, V. (1961). On the biological activity and amino acid composition of secretin. *Acta Chemica Scandinavica*, **15**, 1790-1791.

Lehnert, P., Stahlheber, H., Röder, O., Zoelch, M., Forell, M. M., Wünsch, E., Jaeger, E., and Martens, H. L. (1973). Bestimmung der biologischen Aktivität von synthetischem Secretin. *Klinische Wochenschrift*, **51**, 44-46.

Mitznegg, P., Bloom, S. R., Domschke, W., Domschke, S., Wünsch, E., and Demling L. (1976). Release of motilin after duodenal acidification. *Lancet*, **1**, 888-889.

Mitznegg, P., Bloom, S. R., Domschke, W., Domschke, S.,

Wünsch, E., and Demling, L. (1977). Pharmacokinetics of motilin in man. *Gastroenterology*, **72**, 413-416.

Rösch, W., Lux, G., Domschke, S., Domschke, W., Wünsch, E., Jaeger, E., and Demling, L. (1976). Effect of 13-nle-motilin on lower esophageal sphincter pressure in man. (Abstr.) *Gastroenterology*, **70**, 931.

Ruppin, H., Domschke, S., Domschke, W., Wünsch, E., Jaeger, E., and Demling, L. (1975). Effects of 13-nle-motilin in man—inhibition of gastric evacuation and stimulation of pepsin secretion. *Scandinavian Journal of Gastroenterology*, **10**, 199-202.

Thompson, J. C., Reeder, D. D., Bunchman, H. H., Becker, H. D., and Brandt, E. N. (1972). Effect of secretin on circulating gastrin. *Annals of Surgery*, **176**, 384-393.

Tympner, F., Domschke, S., Domschke, W., Classen, M., and Demling, L. (1974). Reproducibility of the response to secretin and secretin plus pancreozymin in man. *Scandinavian Journal of Gastroenterology*, **9**, 377-381.

Wünsch, E., Jaeger, E., Deffner, M., Scharf, R., and Lehnert, P. (1972). Zur Synthese des Sekretins, VI Reindarstellung des synthetischen Sekretins. *Chemische Berichte*, **105**, 2515-2522.