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Contribution of bile and pancreatic juice to the control of pH in the pig duodenum

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Summary. To evaluate the respective contributions of biliary and pancreatic secretions in the formation of intraduodenal pH, 10 pigs were chronically fistulated in either the common bile duct (5 pigs) or the pancreatic duct (5 pigs). They were all fitted with a permanent catheter into the duodenum for the continuous return of secretion and with a duodenal T-cannula allowing the introduction of a combined glass electrode. pH was recorded over 8-hour periods in fed or fasted pigs in which both secretions were returned or in which either the bile or the pancreatic juice was not restituted. In fed pigs with secretions normally flowing into the duodenum, the pH fluctuated between 4 and 6 during 57 % of the recording time ; it exceeded 6 during 29 % of this period. The rest of the time, it remained between 2 and 4. Bile deprivation did not change the duration of the $4 < pH < 6$ and $2 < pH < 4$ ranges, although a slight decrease of the $pH > 6$ time-course was recorded. Deprivation of pancreatic secretion did not induce significant variations of the pH pattern. In fasted pigs with restituted biliary and pancreatic secretions, a $pH > 6$ was recorded for 70 % of the 8-hour period. The pH was lower in short periods : 19 % ($4 < pH < 6$), 8 % ($2 < pH < 4$) and less than 1 % ($pH < 2$) of the time. Total suppression of bile as well as that of pancreatic juice resulted in a significant decrease of the $pH > 6$ range to the benefit of the acid periods which were increased. These observations suggest that, in fed pigs, food temporarily surpassed the neutralizing abilities of the digestive secretions, but that, in fasted pigs, biliary and pancreatic secretions were important in the neutralization of duodenal juice.

Introduction.

Studies on patients with severely reduced exocrine pancreatic function have shown that increased acidity in the duodenum is a possible pathogenetic factor in duodenal ulcers and that it may also interfere with bile acid function (Go *et al.*, 1970) and intestinal pancreatic enzyme hydrolysis (Malagelada, 1980). It has been demonstrated in healthy men that most of the acid in the duodenum is neutralized by bicarbonate, while some diffuses into the mucosa (Winship and Robinson, 1974), the bicarbonate sources being secretions from the pancreas, liver and Brunner's glands (Brooks, 1970 ; Rhodes *et al.*, 1984 ; Florey and Harding, 1935).

According to Dutta, Russel and Iber (1979), pancreatic juice plays a large role in the control of endoluminal acidity. According to Flemstrom and Kivilaakso (1983), the most likely source of bicarbonate is the duodenal mucosa. The magnitude of liver bicarbonate contribution is not known.

The aim of the present study was to quantify the effect of a total and permanent extracorporeal diversion of bile or pancreatic juice in pigs in order to evaluate the respective contributions of these secretions to the formation of postprandial and interdigestive pH in the duodenum. The pig was chosen because it is a good experimental model for human digestive studies.

Material and methods.

Animal preparation. — Ten castrated male pigs of the Large-White breed, initially weighing 50.8 ± 1.5 kg, were used. After anesthesia they were fitted either with a chronic fistula in the bile duct (5 pigs) or with a catheter in the pancreatic duct (5 pigs), using techniques previously described (Corring, Juste and Jimenez, 1983 ; Corring, Aumaitre and Rérat, 1972). For re-infusion of secretion, all the pigs were fitted with a catheter in the duodenum at the natural junction of the fistulated bile or pancreatic duct. A T-cannula was introduced into the duodenal lumen downstream from the pancreatic duct junction ; after the animals were slaughtered, T-cannula was immediately localized at a mean 19 ± 1 cm downstream from the pylorus in the 10 pigs studied. The duodenal junctions of the bile and pancreatic ducts were an average distance of about 3 and 12 cm, respectively, from the pylorus.

The animals received a standard fattening diet (800 g of meal diluted in 1 600 ml of water) containing 16 % of protein and distributed in two meals at 9 h 00 and at 17 h 00.

Experimental design. — After surgery, the bile was returned immediately and the pancreatic juice 24 h later through the duodenal catheter. The diverted secretion was returned to the animal at a rhythm mimicking excretory output (Juste, Corring and Le Coz, 1983).

After one week of postoperative recovery, the experiment was carried out in two consecutive phases using the following schedule : 4 days of permanent restitution, 2-3 days without experimentation, 4 days of either bile or pancreatic juice deprivation. The pH was recorded continuously for 8 h (from 9 h 00 to 17 h 00) during each experimental day. These trials were done alternately in pigs fed at 9 h 00 or fasted for 16 h at the beginning of recording (previous meal eaten the afternoon before at 17 h 00). Secretion restitution was suppressed 24 h before the beginning of the second phase.

Measurement of pH and expression of data. — A small 12-mm combined glass electrode (Ingold model 440, Urdorf, Switzerland) was placed in the duodenal T-cannula ; the endoluminal part of this cannula was made so as to prevent any contact between the intestinal wall and the electrode.

The electrode was connected to a pH meter (Beckman Zeromatic SS3) coupled to a continuous pen recorder (Beckman Dynegraph R.M.). Paper speed was 6 cm.min^{-1}. The apparatus was calibrated using buffer solutions with known pHs of 2, 4 and 7, respectively, before and after recording. The temperature at which the electrode functioned was taken into account by adjustment of the pH meter (20 °C for buffer solutions ; 38 °C in the pigs).

In fasted pigs, pancreatic juice and bile volumes flowing through the duodenum amount to 50 ml.hour^{-1} (Corring, Aumaitre and Rérat, 1972 ; Juste, Corring and Le Coz, 1983). With additional gastric juice, the measurements of pH variation were assumed to be valid.

The pH recordings were analysed in terms of time-course and expressed by the percentage of recording time during which the pH stayed between the limit values that defined the four ranges of relative acidity : pH > 6 ; 6 > pH > 4 ; 4 > pH > 2 ; pH < 2. In the same situation (fasted or fed pigs and for each of the four pH ranges), the results of the restitution periods and deprivation periods were compared using Student's t-test for paired data.

Results.

Fed pigs. — During the 8 hours following meal intake in control pigs (bile and pancreatic secretions restituted), the pH fluctuated between 4 and 6 during 57 % of the recording time (fig. 1) ; it was more than 6 during 29 % of that time. The rest of the time, pH values were less than 4. When the bile was not restituted to the duodenal lumen, the pH was higher than 6 for a shorter time (17 % only). The duration of periods with a pH varying within the other ranges was not significantly longer. The deprivation of pancreatic juice did not significantly change the duration of any of the different pH ranges compared to those recorded in the controls. The pH changed more often between 4 and 6 (52 % of the experimental period).

Considering each consecutive postprandial 2-hour period separately, we compared, in the same way, pigs with secretions restituted and those with no restitu-

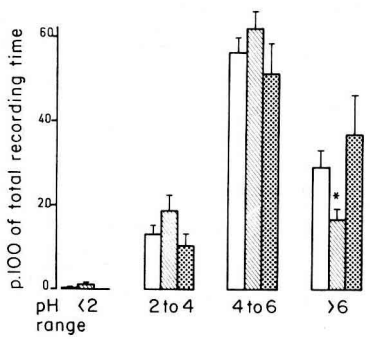

FIG. 1. — *Intraduodenal pH* (as per cent of the 8 hour-recording duration) *in fed pigs.* White bars : bile and pancreatic juice restituted (20 repetitions). Hatched bars : bile deprivation (10 repetitions). Dark bars : pancreatic juice deprivation (10 repetitions). Results are expressed as mean ± SEM. * $P < 0.05$ *vs* restitution period.

tion of bile or of pancreatic juice. Figure 2 shows that non-restitution of bile decreased the time-course of the pH range near neutrality, except for the period between 4 and 6 h after meal intake. During the first 2 postprandial hours, this acidification caused the time-course of the pH range between 2 and 4 to lengthen (17 % *vs* 4 % of the control time-course). During the other three periods (2-4, 4-6 and 6-8 h after meal intake), the time course for the pH ranges below 6 was not significantly different from that in the controls. In no postprandial period studied did the deprivation of pancreatic juice significantly change the time-course of the various pH ranges.

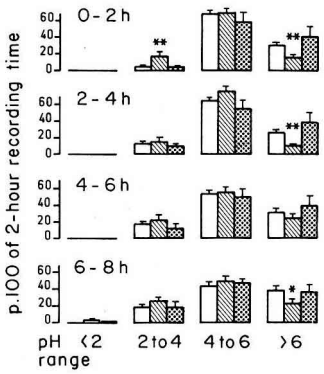

FIG. 2. — *Intraduodenal pH* (as per cent of each consecutive postprandial period of 2 h) *in fed pigs.* White bars : bile and pancreatic juice restituted (20 repetitions). Hatched bars : bile deprivation (10 repetitions). Dark bars : pancreatic juice deprivation (10 repetitions). Results are expressed as mean ± SEM. * $P < 0.05$ *vs* restitution periods, ** $P < 0.01$ *vs* restitution periods.

Fasted pigs. — In pigs fasted for 16 h (fig. 3) and with restitution of pancreatic and biliary secretions, the pH was higher than 6 for more than 70 % of the recording time. The pH showed only temporary recordings between 4 and 6 (19 %) and between 2 and 4 (9 %), and rarely fell below 2 (less than 1 %). Non-restitution of bile caused a very significant decrease in the length of time that the pH remained higher than 6 (40 % of the experimental period), while the time that pH ranged between 4 and 6, on one hand, and between 2 and 4 on the other (32 and 24.5 %, respectively, of recording time), increased significantly. Pancreatic juice deprivation increased this same type of effect even more : the time-course

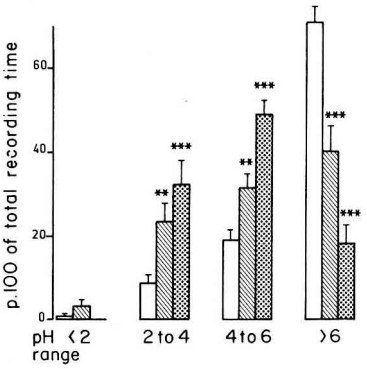

FIG. 3. — *Intraduodenal pH* (as per cent of the 8 hour-recording duration) *in fasted pigs.* White bars : bile and pancreatic juice restituted (20 repetitions). Hatched bars : bile deprivation (10 repetitions). Dark bars : pancreatic juice deprivation (10 repetitions). Results are expressed as mean ± SEM. * $P < 0.05$ *vs* restitution period, ** $P < 0.01$ *vs* restitution period, *** $P < 0.001$ *vs* restitution period.

of the pH range near neutrality decreased (18 *vs* 71 % in the controls). The pH particularly changed between 4 and 6 (49 % of the recording time) and between 2 and 4 (32 %). Excluding bile or pancreatic juice from the duodenal lumen did not cause any significant variation in the pH range of less than 2.

Discussion.

This study based on the continuous measurement of duodenal pH shows that preferential pH is between 4 and 6 in fed pigs with normal restitution of pancreatic juice and bile. These two secretions therefore seem to have very little part in neutralizing the gastric chyme entering the proximal small intestine in these conditions. On the whole, while the absence of bile caused a slight acidification in the duodenal lumen, the non-restitution of pancreatic juice did not significantly change the time-course during which the pH varied in the different ranges, whether during total recording (8 hours) or during each successive 2-hour period.

In humans with chronic pancreatitis associated with insufficient secretion, overall postprandial duodenal pH was more acid than that recorded in the control subjects (Regan *et al.*, 1979 ; Bommelaer *et al.*, 1984). This divergence may result either from a difference in the neutralizing ability of human pancreatic juice and pig pancreatic juice or from a variation in the amount of acid entering the duodenum : an increase in the amount of acid secreted from a gastric pouch was observed by Sekine *et al.* (1981a) in the ligated pancreatic duct of the dog. In the present study, neither the level of gastric acid secretion nor gastric emptying was measured in animals without restitution of the pancreatic juice.

Contrary to results in the postprandial period bile and pancreatic juice are the two main routes by which the contents of the duodenal lumen are neutralized during the interdigestive period. Bile deprivation, and particularly pancreatic juice deprivation, lead to clear acidification of intraduodenal pH. This result agrees with conclusions reported by Dutta, Russel and Iber (1979) in humans with chronic pancreatic insufficiency. The intraduodenal pH of these patients was often less than 6, although it exceeded this threshold during more than 90 % of the recording period in control subjects. However, duodenal acidification seems to be less than that recorded in pigs with pancreatic juice deprivation.

The present work is the first in any species to study the potential effect of bile on intraduodenal pH. The duodenal acidification we observed in pigs with no bile restitution might be a result of excess gastric acid secretion, as shown by Sekine *et al.* (1981b) in dogs after bile deprivation.

Finally, the large quantities of contents flowing into the duodenum during the postprandial period seem to play the main role in regulating endoluminal pH in pigs, suggesting that neutralizing duodenal mechanisms were transiently overcome.

The relative acidification of the duodenal lumen recorded in fed pigs compared to fasted ones might impair the catalytic activity of the main pancreatic enzymes, trypsin and lipase (Malagelada, 1980). Lipase, irreversibly destroyed at a pH of less than 4, loses 70 % of its activity when pH is less than 6 (Heizer, Cleave-

land and Iber, 1965) ; this situation occurred 60 % of the time in the present results. These conditions are usually considered as unfavorable to digestion. However, digestion did occur and with a satisfactory yield ; it might be that the enzymes were protected against acid. The physical-chemical heterogeneity of the duodenal contents after meal intake could lead to better adsorption of some enzymes by food particles or by the mucosa which would provide a more suitable environment for the activity of those enzymes. Moreover, the high transit rate of the alimentary bolus in the duodenum (Laplace, 1978) reduces the length of time the enzymes are exposed to acid there. Our study was limited to a recording of the pH in the proximal duodenum lumen, and it still remains to be determined over what length of small intestine there is complete neutralization. There is no doubt that it is difficult to determine a general relationship between duodenal pH and enzyme activities *in vivo*. Considering the enormous secretory capacity of the pig pancreas (Corring, 1980), it is necessary to define the quantity of enzymes rendered inactive by acid compared to the total amount excreted into the duodenum.

In *conclusion,* the present results show that pancreatic and biliary secretions in fed pigs have only a minimal role in the neutralization of proximal duodenum contents. On the contrary, bile, and particularly pancreatic juice, contribute highly to the neutralization of the gastric acid chyme arriving in the duodenum of fasted pigs.

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Résumé. *Rôle des sécrétions biliaire et pancréatique dans le contrôle du pH duodénal chez le porc.*

Afin d'évaluer les contributions respectives des sécrétions biliaire et pancréatique dans la formation du pH intraduodénal, 10 porcs ont été munis sous anesthésie d'une fistule chronique soit du canal cholédoque (5 porcs), soit du conduit pancréatique (5 porcs). Sur tous les animaux, un cathéter a été placé dans le duodénum pour une restitution continue de la sécrétion dérivée, et une canule en T permettant l'introduction d'une électrode combinée en verre a été implantée en aval de l'abouchement naturel du canal pancréatique. Les enregistrements de pH ont été réalisés pendant des périodes de 8 h chez l'animal alimenté ou à jeun, soumis soit à une irrigation duodénale des 2 sécrétions, soit à une dérivation extracorporelle totale de la bile, soit à une dérivation extracorporelle totale du suc pancréatique.

Chez le porc alimenté et bénéficiant d'une irrigation duodénale de ses sécrétions digestives, le pH a oscillé entre 4 et 6 durant 57 % de la durée de la période d'enregistrement ; il a excédé 6 pendant 29 % de cette durée, le reste du temps correspondant à des valeurs comprises entre 2 et 4. La non-réintroduction de la sécrétion biliaire n'a pas modifié significativement les durées des plages de pH acide ($4 < pH < 6$) et fortement acide ($2 < pH < 4$), bien qu'une légère diminution de la durée de la zone de $pH > 6$ ait été enregistrée. La dérivation de la sécrétion pancréatique n'a entraîné aucune variation significative des temps d'évolution du pH à l'intérieur de ces différentes zones.

En période interdigestive, un pH supérieur à 6 a été enregistré pendant 70 % de la durée de la période expérimentale lorsque la bile et le suc pancréatique étaient normalement présents dans la lumière intestinale. Le pH n'était qu'épisodiquement compris entre 4 et 6 (19 % du temps), 2 et 4 (8 % du temps) et était très rarement inférieur à 2 (moins de 1 %

de la durée de l'enregistrement). La dérivation extracorporelle totale de la bile, et plus encore du suc pancréatique, a entraîné une diminution significative de la durée de la zone de pH voisine de la neutralité (pH > 6) au bénéfice des plages de pH acide et fortement acide qui ont été augmentées.

Ces résultats suggèrent chez le porc alimenté une capacité réduite de neutralisation du bol alimentaire par les sécrétions pancréatico-biliaire, mais mettent en évidence en période interdigestive un rôle important de la bile, et plus encore du suc pancréatique, dans la neutralisation du chyme duodénal.

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