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Comparison of the oral (PABA) pancreatic function test, the secretin-pancreozymin test and endoscopic retrograde pancreatography in chronic alcohol induced pancreatitis

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SUMMARY The oral (PABA) pancreatic function test (PFT), the secretin-pancreozymin test and endoscopic retrograde pancreatography (ERCP) have been carried out in 32 patients with suspected chronic alcohol induced pancreatitis (CAIP) in order to evaluate which, if any, test was most likely to confirm the provisional diagnosis.

Thirty one patients had changes of minimal (n=6) moderate (n=7) or advanced (n=18) chronic pancreatitis on pancreatography, whilst one patient had a pancreas divisum. Eight hour urinary PABA excretion was significantly reduced in patients with moderate and advanced structural changes ($p<0.001$) and correlated significantly with all parameters of the PFT, although eight patients with an abnormal pancreatogram and pancreatic function test had a normal PABA value. The PFT was abnormal in 23 patients, but normal in five patients with an abnormal pancreatogram and low PABA value. Most patients with minimal change pancreatitis had a normal PABA test and PFT. We conclude that pancreatography appears to be the most sensitive method for detecting chronic pancreatic damage and for confirming a clinical diagnosis of chronic alcohol induced pancreatitis. Both the PFT and PABA test are useful confirmatory tests and whilst the PFT is slightly more sensitive for assessing pancreatic exocrine function, the PABA test is well tolerated and simple to perform. It may therefore be the complementary investigation of choice for this group of patients.

The differentiation of small bowel malabsorption from chronic pancreatic disease with steatorrhoea is rarely a diagnostic problem in clinical practice as chronic pancreatitis, particularly when due to alcohol is accompanied by pain in over 90% of cases.[1] Nevertheless, the clinical problem is often to decide whether a patient with a suggestive history of pain and excess alcohol ingestion but without steatorrhoea does in fact have chronic pancreatitis. Because quite advanced morphological changes of chronic pancreatitis can occur in the absence of either overt evidence of malabsorption or comparable changes in standard tests of pancreatic exocrine function and *vice versa*, morphological and functional tests are probably complementary.[2]

Any method used to arrive at a diagnosis of chronic pancreatitis should ideally be evaluated by comparison with histology of the affected gland although in practice this is rarely available because only a small proportion of patients come to surgery. Structural damage must therefore be assessed by pancreatography using established criteria[3] although to obtain consistent good quality pancreatograms requires technical expertise and may be associated with a slight morbidity.

The secretin-pancreozymin test continues to be the standard test of pancreatic exocrine function against which newer tests should be compared.[2] It is, however, time consuming and requires consider-

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able technical skill for meaningful results to be obtained.

In the face of these drawbacks to established methods of diagnosing chronic pancreatitis, a simpler test for confirming a clinical diagnosis of chronic pancreatitis would be useful.

A new direct test of exocrine pancreatic function which uses a specific substrate for chymotrypsin is simple to undertake and does not require duodenal intubation.[4–7]

Briefly, chymotrypsin hydrolyses the p-aminobenzoic acid (PABA) moiety of an ingested synthetic peptide, N-benzoyl-L-tyrosyl-p-aminobenzoic acid (Bz Ty PABA). This moiety is then rapidly absorbed across the small intestinal mucosa, conjugated in the liver and excreted in the urine. Assuming normal small bowel and hepatic function the percentage recovery of PABA in the urine is proportional to chymotrypsin activity in the upper small bowel, although this is influenced by intraluminal pH.[8] This test has been shown to correlate with both the Lundh test, [4 5] the secretin-pancreozymin test[9 10] and also with faecal chymotrypsin activity[11] and faecal fat excretion.[12] More recently attempts to improve the diagnostic accuracy of the test have been reported.[13–15]

Despite this correlation with standard tests of pancreatic exocrine function, there have been no studies to determine the degree with which the PABA test correlates with pancreatic morphology and, therefore, its usefulness as a means of excluding or confirming a clinical diagnosis of chronic pancreatitis associated with structural damage. We have therefore prospectively compared the PABA test with endoscopic retrograde pancreatography and the secretin-pancreozymin test in patients with chronic alcohol induced pancreatitis.

Methods

PATIENTS

Thirty two consecutive patients with a clinical diagnosis of chronic alcohol induced pancreatitis based on a history of typical pancreatic pain and chronic excess alcohol ingestion (>80g alcohol per day) were studied as part of their routine pancreatic assessment.

A smaller number of patients were also diabetic (n=8), had biochemical steatorrhoea (n=7) or had a history of previous documented attacks of acute pancreatitis (n=13). There were 30 men and two women, mean age 45 years (27–58 years). No patient with an episode of acute pancreatitis within the previous six weeks or a pancreatic cyst was included. Renal and hepatic function were biochemically normal in all subjects and no patient had clinically overt small bowel disease.

For the PABA test the control group (HC) consisted of 18 healthy subjects from the hospital staff, mean age 41 years (27–55).

PABA TEST

After an overnight fast subjects took 0·5 g of the synthetic peptide N-benzoyl-L-tyrosyl-p-aminobenzoic acid (Fluorochem Ltd, Glossop, Derbyshire) dissolved in a Lundh test meal, which acted as a standard stimulus of pancreatic secretion. Water was allowed but food was witheld until four hours after ingestion of the test meal. Urine was collected for eight hours and assayed for aromatic amines as previously described.[6 16] No patient was taking any medication other than paracetamol for analgesia or pancreatic enzyme supplements. These were stopped for 48 hours before the test. All subjects were requested to avoid foods with a high aryl acid content for 24 hours before the test. To ensure that no interference with the laboratory analysis had occurred, a pretest urine sample was also collected and assayed for PABA content. An eight hour urinary excretion of PABA of less than 59% (control mean minus 2 SD) was regarded as abnormal.

SECRETIN-PANCREOZYMIN TEST

Pancreatic exocrine function (standard pancreatic function test) was tested by analysing duodenal contents for volume, mean and maximal bicarbonate output, amylase, trypsin, chymotrypsin and lipase concentration following an intravenous injection of secretin and pancreozymin as previously described.[17] Briefly, after an overnight fast two Salem Sump tubes (Argyle) were passed nasogastrically. Under fluoroscopic control, one tube was positioned in the distal second part of the duodenum and the other in the dependent part of the stomach for continual aspiration of gastric contents. After a 10 minute basal collection which was discarded, an intravenous bolus of Boots secretin 2 U/kg (batch no 91510/4) was given, and the duodenal contents aspirated for six consecutive 10 minute periods. After one hour an intravenous bolus of Boots pancreozymin 1·5 U/kg (batch no 91331/2) was given and a further two 10 minute collections were made.

The volumes of each sample were noted and the bicarbonate, trypsin, chymotrypsin, amylase and lipase concentrations and output analysed on the same day as the test. Trypsin was determined by the method described by Nardi[18] using benzoyl-I-arginine amide hydrochloride as the substrate, and was expressed as units of tryptic activity. Chymotrypsin was measured according to the method of Schwert and Takenaka[19] using N-acetyl-L-tyrosine

ethyl ester as substrate.

Amylase was determined using Pimstones modification of the Gomori method[20] and the result expressed as milligrams of starch digested by one millilitre of pancreatic juice in 30 minutes at 37°C.

Lipase was measured by Pimstone's modification of the Gomori method[21] using alpha-naphthyl laurate as substrate and a diazonium salt as the colour reagent. The result was expressed as the number of micromoles of alpha naphthol liberated by one ml of pancreatic juice in three hours at 37°C. The results for volume, mean and maximal bicarbonate output, amylase and lipase were scored from 0 to 2 and the total used as an arbitrary value for function on a scale from 10 to 0, where 10/10 represented normal function and increasingly abnormal function ranged from 9/10 to 0/10.[22]

ERCP

ERCP was carried out in the standard manner.[23] Pancreatograms were assessed blindly by three independent observers and pancreatic ductal abnormalities classifed as minimal, moderate or advanced changes according to established criteria. Lateral branch filling sufficient to evaluate minimal changes was achieved in all cases and a minimum of three abnormal side branches required for inclusion in this group. Advanced changes were not further sub-divided although no patient had a complete main duct obstruction. The presence or absence of pancreatic calcification was determined by plain abdominal radiograph and computed tomography.

STATISTICAL METHODS

Pearsons correlation coefficient was used for comparison of the PABA test and the standard pancreatic function test and Spearmans rank order correlation for comparison of the PABA test with ERCP grading. For comparisons within groups the Mann-Whitney U test or Newmans-Keuls technique of multiple comparisons was used.

Results

Twenty patients had non-calcific chronic pancreatitis (NCP) and 11 patients had chronic calcific pancreatitis (CCP). Six patients had minimal, seven moderate and 18 advanced changes of chronic pancreatitis based on the structural changes noted as pancreatography. All patients with calcification automatically entered the advanced group. One patient without calcification had a pancrease divisum and was excluded from further consideration because of a failure to delineate the duct of Santorini at ERCP.

PABA EXCRETION

The percentage of PABA excreted in the urine after eight hours (mean±SEM) was 71·6±1·5 for healthy controls, 52·6±3·2 in the NCP group and 39·2±4 in the chronic calcific pancreatitis group (Fig. 1). The results according to the pancreatogram gradings were 60·8±2 for minimal 50·8±6 for moderate and 42·9±3·8 for advanced changes (Fig. 2).

The differences between healthy controls, non-calcific chronic pancreatitis and chronic calcific pancreatitis were significant and for patients with chronic calcific pancreatitis there was no overlap with the healthy control group (HC *vs* CCP $p<0·001$; HC *vs* NCP $p<0·001$; CCP *vs* NCP $p<0·005$). The PABA values did not correlate with the ERCP grading overall (rho=0·32) although there were significant differences in PABA values between individual groups (HC *vs* moderate $p<0·001$; HC *vs* advanced $p<0·001$; minimal *vs* advanced $p<0·001$).

The PABA values also correlated significantly

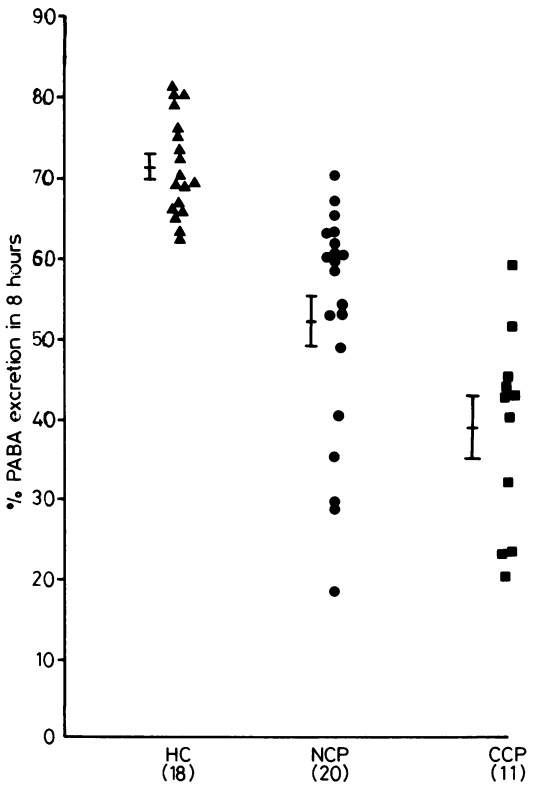

Fig. 1 *Eight hour urinary excretion of PABA in healthy controls (▲) and patients with non-calcifying (●) and calcifying pancreatitis (■).*

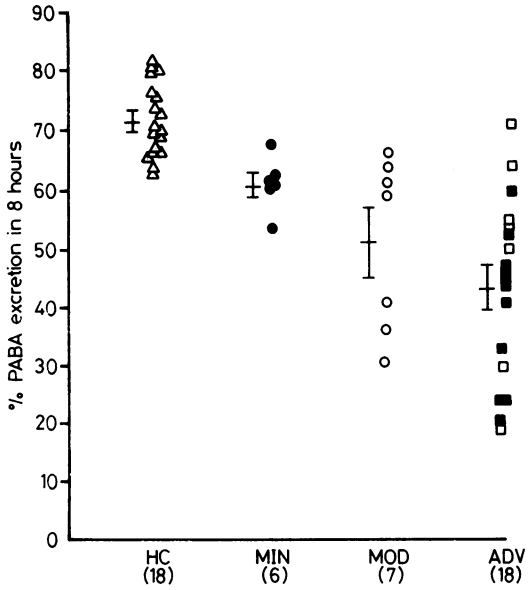

Fig. 2 *Eight hour urinary excretion of PABA in healthy controls (△) and patients with minimal (●), moderate (○) and advanced CAIP [calcifying (■), non-calcifying (□)].*

with all parameters of the standard pancreatic function test and the arbitrary pancreatic function test score (Table 1). As expected, chymotrypsin showed the best correlation with the PABA test. Some patients with abnormal PABA tests had normal standard pancreatic function tests (Table 2) and *vice versa* (Table 3).

Discussion

Since the introduction of the oral (PABA) pancreatic function test, it has been shown to correlate well with both the secretin-pancreozymin test[9 10] and the Lundh test[4 5] in patients with chronic pancreatitis. The present study in patients with painful chronic alcohol induced pancreatitis has also shown a significant association of the PABA test with differing degrees of structural damage as noted on pancreatography in addition to confirming that there is a significant correlation between the PABA test and the secretin-pancreozymin test. The diagnosis and evaluation of chronic pancreatitis using either functional or structural criteria continues to arouse controversy, primarily because morphological and functional tests vary. It is generally considered that the standard method of detecting and evaluating pancreatic function and the test with which others should be compared, remains the secretin pancreozymin test.[2] Some workers, however, prefer the somewhat simpler Lundh test for routine diagnostic use.[24] In comparing function tests with pancreatogram findings most studies have suggested that the secretin-pancreozymin test is a more sensitive indicator of chronic pancreatic damage[25–27] than the Lundh test[28 29] although it has been claimed that the latter may be a more sensitive indicator of minimal change pancreatitis.[30]

Nevertheless, in all these studies there are a number of patients with chronic pancreatitis who have an unequivocally abnormal pancreatogram and yet apparently normal pancreatic exocrine function. The converse is equally true, although in this series no patient had abnormal exocrine function and a

Table 1 *Correlation of eight hour urinary PABA recovery with standard pancreatic function test values*

% PABA excretion	*vs* Vol	r=0·52	p<0·01
	vs max HCO_3	r=0·62	p<0·001
	vs mean HCO_3	r=0·63	p<0·001
	vs amylase	r=0·42	p<0·05
	vs trypsin	r=0·57	p<0·001
	vs chymotrypsin	r=0·64	p<0·001
	vs lipase	r=0·52	p<0·01
	vs PFT score	r=0·62	p<0·001

Table 2 *Pancreatogram grading and PABA values in patients with chronic alcohol induced pancreatitis and a normal standard pancreatic function test and an abnormal PABA test*

Patients	*Calcification*	*Pancreatogram, grading*	*PABA value (<59%)*
JaP	Yes	Advanced	45·8
JP	No	Advanced	54·7
EK	No	Moderate	40·6
JG	No	Moderate	58
MH	No	Minimal	53·9

Table 3 *Pancreatogram grading and standard pancreatic function test score in patients with a normal oral pancreatic function (PABA) test (>59%) and an abnormal pancreatic function test*

Patient	*Calcification*	*Pancreatogram grading*	*PFT score 10 (normal) 0 (bad)*	*PABA value*
HA	Yes	Advanced	7	59·7
LP	No	Advanced	6	70·9
BP	No	Advanced	9	63·8
WVR	No	Moderate	6	60·9
GC	No	Moderate	8	66·0
AP	No	Moderate	8	63·5
LL	No	Minimal	8	62·1
LB	No	Minimal	9	67·5

normal pancreatogram. Some of these difficulties arise because of observer variation in pancreatogram interpretation,[31] particularly regarding the diagnosis of minimal change pancreatitis or because of different methodologies and interpretations of the results of the secretin-pancreozymin test.[27] As Tables 2 and 3 clearly illustrate, however, there remain patients with gross structural changes and normal exocrine function as measured by either the standard pancreatic function test or the PABA test. Although in this series the secretin-pancreozymin test appears slightly more sensitive than the PABA test (23/31 abnormal=74%, 20/31 abnormal=65%) this advantage does not outweigh the disadvantages of the test as outlined earlier. Furthermore, in the six patients in this study with minimal change pancreatitis the standard pancreatic function test was abnormal in two patients while the PABA test was abnormal in only one (Table 4), suggesting that neither test is of particular benefit in early cases of chronic alcohol induced pancreatitis. Both tests, however, remain useful as corroborative evidence of pancreatic disease in this difficult group.

The reason why the majority of patients with minimal structural damage do not have functional impairment remains unclear, although it has been shown that a significant percentage of alcoholic patients without clinical evidence of pancreatitis but with evidence of pancreatic cellular derangement at the electron microscopic level have increased 80 minute flow, and bicarbonate secretion rates.[32] It is possible therefore that a number of patients with minimal change pancreatitis pass through a stage of hypersecretion before functional impairment becomes apparent.

Histological proof must remain the final arbiter of whichever test is used to evalute a clinical diagnosis of chronic alcohol induced pancreatitis. Nevertheless, in the majority of patients surgical intervention is unnecessary and such proof unobtainable. In the absence of histological confirmation therefore in patients presenting with a suggestive history, this study would suggest that the most useful investigation for confirming a clinical diagnosis of chronic alcohol induced pancreatitis is ERCP. A test of exocrine function is a complementary procedure and when abnormal is useful corroboration. In this situation the PABA test is well tolerated, simple to carry out and correlates well with the degree of pancreatic damage as determined by pancreatography. Newer modifications offer the advantage of even greater sensitivity[33] and specificity although the simplicity of the test is compromised. The secretin-pancreozymin test may continue to be useful in patients with suspected chronic pancreatitis who have a normal PABA test and an equivocal or normal pancreatogram.

Table 4 *PABA values and pancreatic function test scores in patients with minimal changes on ERCP*

Patient	*PABA value*	*PFT score 10 (normal) 0 (bad)*
TB	61·0	10
SP	60·0	10
MM	60·4	10
LL	62·1	8
LB	67·5	9
MH	53·9	10

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