A STUDY OF PANCREATIC ENZYMES BY DUODENAL INTUBATION IN NORMAL SUBJECTS AFTER PROSTIGMINE ADMINISTRATION*

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Summary : Twenty healthy subjects have been selected for the present study. There was significant parallism between trypsin and lipase secretion but frequent dissociation between trypsin and amylase. Lower values of pancreatic volume, bicarbonate and enzymes secretion as compared to the Western reports are explained by the lower weight of Indians in comparison to Western people. Prostigmine an inexpensive and reliable alternative to pancreozymine is recommended for routine clinical studies of pancreatic enzymes.

Key words : pancreatic enzyme duodenal intubation prostigmine

INTRODUCTION

While the analysis of blood for pancreatic enzymes is useful in the diagnosis of acute pancreatitis yet the estimation of serum enzymic levels is rather erratic in chronic pancreatic disorders (5, 17). The estimation of the pancreatic enzymes in pancreatic juice is a better reliable and accurate method in diagnosis of chronic pancreatic disroders (6, 9). Two main difficulties encountered are :

Collection of pancreatic fluid and use of pancreozymin and secretin as stimulants for its secretion, which are expensive and difficult to obtain in India. 'Lundh Test Meal' (4, 13) has not been used for this purpose in the present study because of the complexity of the composition of the same which is purely based on Western lines. Therefore, an alternative i.e., prostigmine which is easily available and inexpensive has been used as an agent for stimulation of the pancreatic enzymes in place of 'Lundh Test Meal'.

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MATERIALS AND METHODS

Twenty healthy subjects having no past history of abdominal operation or, gastrointestinal disease were selected on the basis of criteria of Cohen and Shock (7). They included 12 males and 8 females of 24 to 60 years of age (Table I). The socio-economic status of the patients was also recorded and found good.

TABLE I : Physical characteristics of the subjects and other parameters.

Sex	*Age* (*yrs*)	*Height* (*cms*)	*Weight* (*kg*)	*Surface area* (m^2)	*Temp.* (°C)	*Socio-economic status*
Male (12)						
Range	24–55	156–180	44–71	1.39–1.88	14–23	Good
Mean	33.41	165.5	51.5	1.55	18.5	
Female (8)						
Range	29–60	148–165	40–61	1.28–1.65	14–23	Good
Mean	38.62	158.4	48.3	1.45	18.5	

The patients were advised not to take any thing orally after 10 p.m. on previous night to avoid nauseas and vomiting while passing tube orally. Next day early in the morning at 6 A.M., a specially designed radio-opaque duodenal tube with a mercury bulb attached to its tip and having four holes in the distal 5 cms was passed orally under fluoroscopic control till the bulb reached the pylorus. The position of the bulb at the end of 3rd part of duodenum was confirmed by fluoroscopy. Now a Ryle's tube was passed through the nose into the stomach and continuous suction of gastric contents was done by a water suction pump to prevent mixing of the gastric and the duodenal contents. Intermittent suction was done from the duodenal tube. When alkaline and bile stained fluid started coming, the collection was done in an ice-cold flask containing glycerol. Two basal samples of duodenal fluid of 10 mins each were aspirated. Now prostigmine (6.1 *mg/kg* body weight) was injected intramuscularly and duodenal contents were collected for 2 hours, three samples of 20 mins each in 1st hour and 2 samples of 30 mins each in 2nd hour. The final concentration of glyceroloid juice mixture was made 50% by adding glycerol (Vol/Vol.). Each sample was then analysed for following (a) Total volume, (b) pH., (c) Bicarbonate concentration by method of Lagerlof (12), (d) Amylase activity by method of Norby (15), (e) Lipase activity was determined by Comfort and Osterbero method (8), (f) Trypsin activity was measured by Gowenlock's method (11).

RESULTS

Twenty healthy subjects consisting of twelve males and eight females of age from 24 to 60 yrs (mean age 36.01 yrs) were taken for present study. Colour of fasting as well as of 120 mins post stimulation duodenal aspirate was dark yellow. Fluid was moderately viscid and clear. Its pH value ranged from 7.5 – 9.0 in post stimulation duodenal aspirate.

The volume and enzyme contents of duodenal fluid following prostigmine administration over a period of 120 mins has been depicted in Table II.

TABLE II : Showing the total volume, bicarbonate concentration and enzyme contents of duodenal fluid in 120 mins post stimulation period (20 normal subjects).

Case	*Volume*	*Bicar–bonate*	*Amylase (units)*		*Lipase (units)*		*Trypsin (units)*	
No.	*(ml)*	*(mEq/l)*	*Total*	*per min*	*Total*	*per min*	*Total*	*per min*
1	120	23.20	443.70	3.69	8545	71.2	8763	73.02
2	70	28.80	349.80	2.91	3925	32.7	4040	38.16
3	61	21.00	254.30	2.11	971.5	8.1	2617	21.80
4	65	37.60	445.40	3.71	2750	22.9	2220	18.50
5	62	32.00	57.10	0.47	1240	10.3	3336	27.80
6	76	18.40	291.00	2.42	5425	45.2	2568	21.40
7	105	36.00	728.50	6.07	6390	53.2	2865	23.87
8	80.5	28.80	440.00	3.66	4230	35.2	4117	34.30
9	65.5	30.00	466.00	3.88	3205	26.7	2990	24.91
10	95	25.60	638.80	5.32	5850	48.7	970	8.09
11	107	21.60	530.00	4.41	6960	58.0	6345	52.87
12	45	33.00	327.80	2.73	1200	10.0	245	2.04
13	121	35.20	692.90	5.77	5530	46.1	7545	62.87
14	174	15.20	466.40	3.88	6660	55.5	1143	95.26
15	105	38.40	389.20	3.24	6150	51.2	3590	29.90
16	60	36.00	367.40	3.06	2800	23.3	4525	37.70
17	112	24.80	353.90	2.94	4860	40.5	5956	49.63
18	95	25.60	639.80	5.33	5095	42.4	5770	48.08
19	78	25.00	821.20	6.84	4595	38.3	7047	58.72
20	145	20.00	331.10	2.75	6100	50.8	7435	61.95
RANGE	45–174	15.2–38.4	57.1–821.2	47–6.84	971–8545	8.1–71.2	245–11432	2.04–95.26
Mean ± S.D.	92.10 ± 31.40	27.80 ± 6.70	451.70 ± 176.6	3.76 ± 1.51	4624.07 ± 2023.20	38.53 ± 17.05	4718.8 ± 2713.6	39.32 ± 25.09

The use of prostigmine was found to be safe. Only in four cases nausea and mild abdominal colic occurred, but the tests could even be carried out in such cases also.

DISCUSSION

Pancreozymin and secretion are now well known as stimulants for pancreatic juice production but these can not be used in India due to their extremely high price and non-availability. Prostigmine was therefore used for this purpose. Prostigmine has also been used for the same purpose by Simko (16).

Mean volume of duodenal content after stimulation with prostigmine per 60 min was 46.05 *ml* in our series. These figures are slightly lower than those reported by Sun *et al.* (18). Marin *et al.* (14) have reported post secretion volume per 60 *min/kg* body weight ranging from 1.61 *ml* to 4.9 *ml.*

Choi *et al.* (6) reported post pancreozymine secretin volume after 60 min ranging from 75 to 339 *ml* (Mean 181 *ml*). Indian subjects weigh on an average 15 *kg* less than American and approximately 10 *kg* less than British subjects. These lower values in our subjects could be due to lower dose of prostigmine administration and as well as less body weight of Indians. Repetition of duodenal fluid collection within 1–5 days in 10 subjects gave reproducible results in our series indicating satisfactory technique of fluid collection.

Sun *et al.* (17) reported bicarbonate concentration after pancreozymin stimulation as ranging from 8 to 84 *mEq/litre* (mean 43±15.3), while maximal bicarbonate concentration following secretin ranged from 69 to 126 *mEq/litre* (mean 100±14.3). Sun *et al.* (18) and Marin *et al.* (14) have put maximum normal bircabonate concentration more than 70 *mEq/litre.* Our values of bicarbonate concentration are again at a lower level (15.2 to 38.4 *mEq/litre*) and this can be explained by the lower dose of prostigmine which provides mainly enzymic stimulation due to lower weight of Indian subjects.

In the present study the rate of amylase secretion for the post-stimulation period of 120 mins ranged from 57.1 to 821.2 units. Our results are in close agreement with those reported by Goldberg and Wormsley (10) showing thereby that the pancreatic enzymic stimulating capacity of prostigmine closely parallels that of pancreozymin, although weaker than secretin. It is difficult to compare our results with other reports, due to different techniques and units used. When suitably adjusted for unit, amylase secretion figures compare very well with those of other workers like Choi *et al.* (6).

The rate of secretion of trypsin in the post stimulation period averaged at 4718.8 units. These figures agree with those reported by Goldberg and Wormsley (10).

The lipase content (971.5 – 8545 units/120 mins) in our post stimulation samples agree closely with the results shown by Choi *et al.* (6) who have reported maximal lipase concentration unit/c.c. at 20.9 and total output over a 70 mins period averaging at 3672.0 units.

The pancreatic enzymes are secreted in parallel *in vivo* (2 and 12) and also in response to injection of secretin (3) and pancreozymin (1).

There was a definite relationship (Fig. 1 and 2), statistically significant correlation ($t = 2.7$) between trypsin and lipase. Goldberg and Wormsley (10) have shown that the

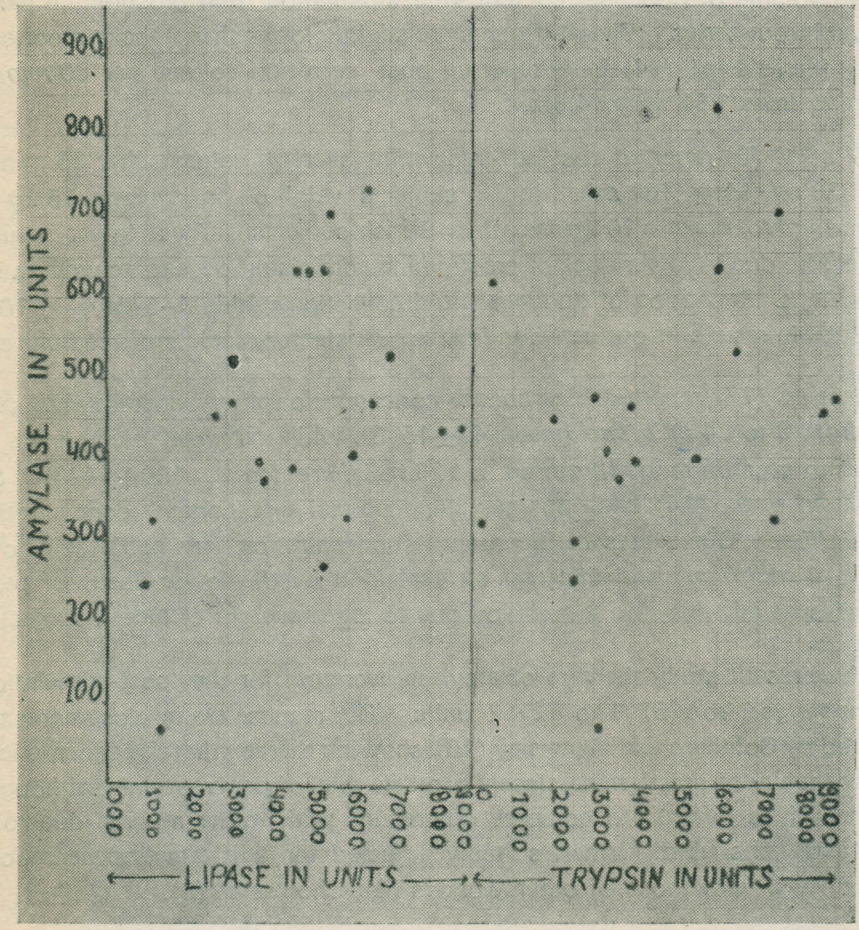

Fig. 1 : Diagram showing statistically insignificant correlation between pancreatic amylase, lipase and trypsin.

relationship between amylase and proteolytic enzyme is not constant. Normal response of the pancreas to stimulation is a greater increase in the secretion of proteolytic enzyme than of amylase. Our findings of insignificant parallelism between trypsin and amylase secretions is in keeping with the observations of above workers. Diamond *et al.* (9) has found frequent dissociation between trypsin and amylase secretions.

It thus appears from the present study that prostigmine provides a pancreatic stimulation very similar to pancreozymin and probably more reliable than test meals (19). Secretin has been shown to cause gradual disappearance of lipase from human duodenal contents without change in the activities of other enzymes (18). The study also reveals that

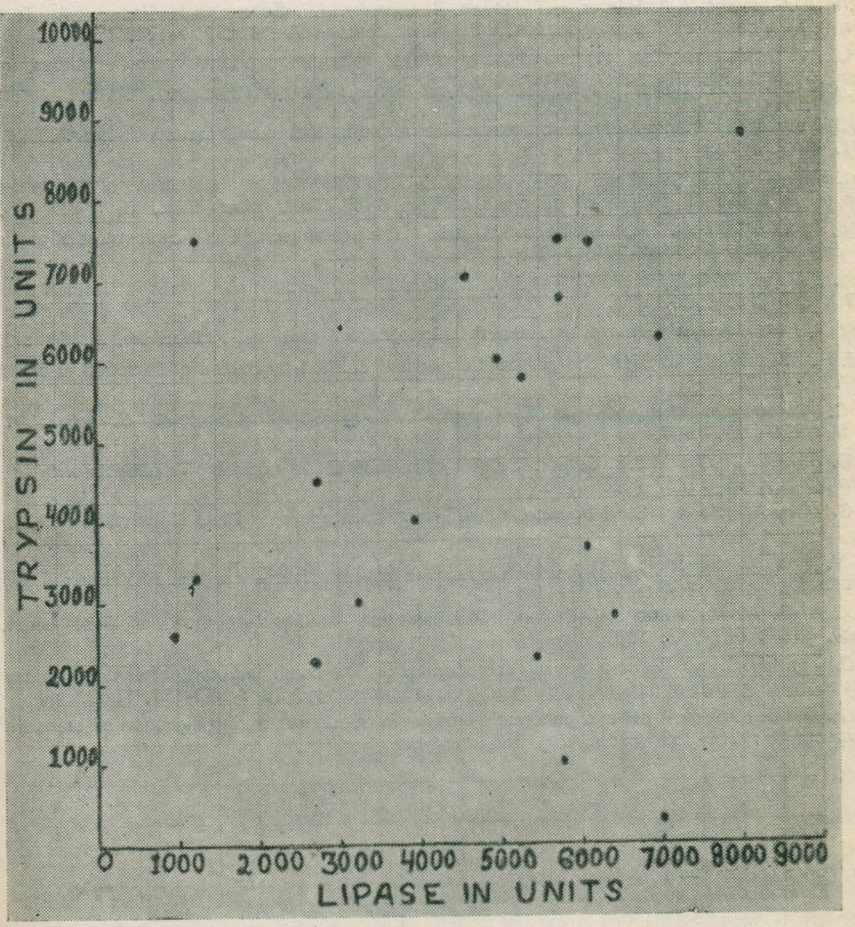

Fig. 2 : Diagram showing statistically significant positive correlation between pancreatic lipase and trypsin contents (t=2.7).

estimation of only one enzyme is not an adequate measure of total pancreatic capacity to secrete enzyme and it is preferable to measure all the three enzymes in routine clinical study.

REFERENCES

1. Ammann, R.W., B. Tagwercher, H. Kashiwagi and H. Rosenwund. Diagnostic value of fecal chymotrypsin and trypsin assessment for detection of pancreatic disease. *Amer. J. Diq. Dis.*, **13** : 123-146, 1968.
2. Babkin, B.P. Secretory mechanism of the digestive glands. New York, Paul B. Hoeber., Inc. 1944.
3. Bockus, H.L., M.H. Lopusnaik and V. Tachdjian. Tests of pancreatic secretion via duodenal intubation. Gastroenterology Ed. by H.L. Bokus III, 907-914, W.B. Saunders Company, Philadelphia, 1965.
4. Borgstrom, B., A. Dahlquist, G. Lundh and J. Sjovall. Study of intestinal digestion and absorption in the human. *J. Clin. Invest.*, **36** : 1521-1536, 1957.
5. Burton, P., D.G. Evans, A.A. Harper, H.T. Howet, S. Oleesky, J.E. Scott and H. Varley. A Test of Pancreatic function in man based on the analysis of duodenal contents. *Gut*, **1** : 111-124, 1960.
6. Choi, H.J., F. Goldstein, G.W. Write and H. Menduke. Normal duodenal trypsin values in response to secretion-pancreozymin stimulation with preliminary data in patients with pancreatic disease. *Gastroenterology*, **53** : 397-402, 1967.
7. Cohen, J.E. and N.W. Shock. Blood volume studies in middle and elderly males. *Amer. J. Med. Sc.*, **217** : 388-391, 1949.
8. Comfort, M.W. and A.F. Osterberg. The value of determinations of concentration of Serum lipase and serum amylase in diagnosis of disease of pancreas. *Proc. Staff Meet. Mayo Clin.*, **15** : 427-433, 1940.
9. Diamond, J.S., B.A. Siegal, M.B. Gall and S. Karten. The use of secretin as a clinical test of pancreatic function. *Am. J. Digest. Dis.*, **6** : 366-372, 1939.
10. Goldberg, D.M. and K.G. Wormsley. The inter-relationship of pancreatic enzymes in human duodenal aspirate. *Gut.*, **11** : 859-866, 1970.
11. Gowenlock, A.H. The estimation of trypsin activity in duodenal contents. *Biochem. J.*, **53** : 274-277, 1953.
12. Legerlof, H. Pancreatic function and pancreatic diseases studied by means of secretion. *Acta. Medica Scand. Suppl.*, 128, 1942.
13. Lundh, G. Deleneation of trypsin and chymotrypsin in human intestinal content. *Scand. J. Clinc. and Lab. Invest.*, **9** : 229-232, 1957.
14. Marin, G.A., M.C. Clark and J.B. Senior. Studies of malabsorption occurring in patients with Laenic's cirrhosis. *Gastroenterology*, **56** : 727-736, 1969.
15. Norby, G. Om Amylasen i Blod ogurine. Levine and Munksgaard. Diss. Gopenhagen 1935. From Lagerlof (1942).
16. Simko, V, and R. Simoneic. The diagnostic value of the Prostigmine test in chronic pancreaticopathies. *Gastroenterologica*, **97** : 21-25, 1962.
17. Sun, D.C.H., R. Albacete and J.K. Cohen. Malabsorption studies in cirrhosis of liver. *Arch. Intern. Med. (Chicago)*, **119** : 567-572, 1967.
18. Sun, D.C.H. and H. Shey. Pancreozymin - secretion test. The combined study of serum enzymes and duodenal contents in the diagnosis of pancreatic disease. *Gastroenterology*, **38** : 570-581, 1960.
19. Wornig, H. and S. Mullertz. pH and pancreatic enzyme in duodenum during digestion of standard meal. *Scand. J. Gastroent.*, **1** : 268-283, 1966.