Leucine aminopeptidase in the pancreatic juice during ethionine pancreatitis

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EDITORIAL SYNOPSIS In the early stages of ethionine pancreatitis leucine aminopeptidase output was increased while the excretion of amylase, trypsin, and lipase was diminished.

Müller-Wieland and Becker (1962) have reported an increase of leucine aminopeptidase in the duodenal juice of man after stimulating the exocrine pancreas. The present experiments were designed to investigate the pattern of leucine aminopeptidase output in normal rats and in animals poisoned with ethionine.

METHODS

Pancreatic juice was withdrawn from 41 male albino rats ranging in weight from 200 to 300 g. in an acute experiment. The exocrine pancreas was stimulated by secretin and pancreozymin (Boots). The technical details have already been described in full (Müller-Wieland, 1964). The activity of the enzymes amylase (Norby, quoted by Lagerlöf, 1942) trypsin (Gowenlock, 1953), lipase (Marchis-Mouren, Sarda, and Desnuelle, 1959), and leucine aminopeptidase (Goldberg and Rutenburg, 1958) was estimated in fractions of pancreatic juice. Pancreatitis was caused by ethionine.

Male rats only were used, because of the excessive fatty degeneration of the liver in female rats in the early stage of ethionine toxicity (Farber, Koch-Weser, and Popper, 1951; Becker 1957).

RESULTS

CONTROLS In six animals the standard errors of the mean for volume, and output of amylase, trypsin, lipase, and leucine aminopeptidase were, in the secretin period, 8.13, 7.57, 10.7, 6.68, and 9.25%, and in the pancreozymin period 12.6, 12.7, 10.7, 11.2, and 5.82%. The relationship of enzyme output during the post-secretin and post-pancreozymin phases was 5.56:1, 2.95:1, 2.43:1, and 2.40:1 (Table I).

INCREASING DOSES OF ETHIONINE (EXPERIMENTAL GROUPS 1-5) In rats given from 0.33 to 1.00 mg./g. body weight of ethionine the volumes of the pancre-

atic juice remained within the normal range with a variation of one standard deviation from the mean in most animals (groups 1-3). With doses of 1.33 and 1.66 mg./g. body weight (groups 4 and 5) the volumes decreased. The excretion of leucine aminopeptidase increases in the early stages of the ethionine pancreatitis. This was particularly noticeable in the post-pancreozymin phase. In group 3 the output was 260% of the normal while the excretion of amylase, trypsin, and lipase was impaired. The leucine aminopeptidase output remains increased in spite of higher doses of ethionine (groups 4 and 5) which caused considerable damage to acinar cell function (Table I and Figure 1).

EXOCRINE FUNCTION OF THE PANCREAS ONE, TWO, THREE, AND FIVE DAYS AFTER ETHIONINE POISONING (EXPERIMENTAL GROUPS 3, 6-8) The rats were given 1.00 mg./g. body weight of ethionine as three single doses (each 0.33 mg./g. body weight). The exocrine pancreatic function was tested after 24, 48, 72, and 120 hours.

Following the injections of ethionine the output of amylase, trypsin, and lipase showed a fall which became maximal on the second day. The excretion of leucine aminopeptidase was diminished during the post-secretin phase and increased in the post-pancreozymin period. Thereafter the output of amylase, trypsin, and lipase was gradually restored to normal by the fifth day. However, at this time the rise in leucine aminopeptidase output reached 264% of normal in the post-secretin phase (Table I and Figure 2).

DISCUSSION

In the pancreatic secretion of normal rats the pattern of leucine aminopeptidase excretion was comparable with that of trypsin and lipase. In the earlier stages of ethionine pancreatitis leucine aminopeptidase out-

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TABLE I

VOLUME AND OUTPUT OF AMYLASE, TRYPSIN, LIPASE, AND LEUCINE AMINOPEPTIDASE AFTER SECRETIN AND PANCREOZYMIN AND MEANS AND SIGNIFICANCE OF DIFFERENCE BETWEEN MEANS OF NORMAL AND ABNORMAL GROUPS OF RATS WITH ETHIONINE PANCREATITIS

Group No.	No.	Volume (ml. 10 ³)				Amylase Output (Norby units)				Trypsin (Gowenlock)	
		Post-secretin Periods 1 + 2		Post-pancreozymin Period		Post-secretin Period 1 + 2		Post-pancreozymin Period		Secretin Periods 1 + 2	
		Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.
Controls	6	534	116	232	71.1	584	108.2	105	32.8	676	178
Increasing doses of ethionine											
1	5	490	50	205	26	510	22	107	34	904	192
2	7	464	26	180	40	570	108	110	66	813	214
3	4	488	22	198	37	362	357	61	32	394	116
4	4	413	110	146	54	195	47	31	15	453	97
5	4	424	72	169	24	260	68	39	21	438	45
Test 1, 2, 3, and 5 days after ethionine poisoning											
3	4	488	26	198	37	362	357	61	32	394	116
6	4	389	124	159	47	246	42	51	12	248	50
7	4	418	86	190	38	343	109	152	65	296	57
8	4	600	71	253	75	493	55	132	39	778	130

FIG. 1. Means of volume and output of amylase, trypsin, lipase, and leucine aminopeptidase in the post-secretin and post-pancreozymin period of groups of rats with pancreatitis induced by increasing doses of ethionine correlated with the means of normals.

TABLE I—continued

VOLUME AND OUTPUT OF AMYLASE, TRYPSIN, LIPASE, AND LEUCINE AMINOPEPTIDASE AFTER SECRETIN AND PANCREOZYMIN AND MEANS AND SIGNIFICANCE OF DIFFERENCE BETWEEN MEANS OF NORMAL AND ABNORMAL GROUPS OF RATS WITH ETHIONINE PANCREATITIS

Units)		Lipase (Desnuelle units)				Leucine Aminopeptidase (units)					
Pancreozymin Period		Secretin Periods 1 + 2		Pancreozymin Period		Secretin 1		Period 2		Pancreozymin Period	
Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.	Mean	S.D.
232	61	235	38.7	100	16.4	34.1	7.55	25.9	5.58	24.1	3.43
284	62	295	41	84	27.2	68.2	9.74+++	45.2	5.67+++	42.2	4.77+++
257	58	276	49	79	16.8	36.3	4.03	44.9	5.20++	51.3	4.68+++
138	55	147	55	44	17.7	35.0	3.36	53.3	3.65++	62.8	2.31+++
108	16	135	50	30	9.2	51.7	9.34+	24.6	3.21	30.1	19.7
95	11	131	28	30	1.0	51.4	15.28	22.6	5.82	35.6	16.2
138	55	147	55	44	17.7	35.0	3.36	53.3	3.65++	62.8	2.31+++
105	33	114	32	35	9.1	30.1	3.82	14.4	3.92++	31.2	4.01+
123	39	219	35	69	18.2	48.8	9.39	28.1	1.68	34.5	3.51++
254	62	249	57	83	17.6	125	22.8+++	35.2	35.3++	31.3	5.39

p = = 0.02-0.01
 f = ++ = 0.01-0.001
 = +++ = 0.001-0.0001

FIG. 2. Means of volume and output of amylase, trypsin, lipase, and leucine aminopeptidase in the post-secretin and post-pancreozymin period of groups of rats with ethionine pancreatitis in the recovery period correlated with the means of normals.

put increased while the poisoning impaired the excretion of amylase, trypsin, and lipase. Ethionine thus affects the excretion of these enzymes in a different manner.

In the groups receiving small doses of ethionine the increasing output of leucine aminopeptidase during the post-pancreozymin period showed that it is not likely to be synthesized and excreted with the other three enzymes.

CONCLUSION

The pancreatic juice of the rat contained leucine aminopeptidase. The pattern of output of amylase, trypsin, lipase, and leucine aminopeptidase was similar in normals. Ethionine affected the excretion of leucine aminopeptidase and the other enzymes in a different manner. At the beginning of ethionine pancreatitis the secretion of leucine aminopeptidase increased, particularly in the post-pancreozymin phase. Leucine aminopeptidase is not likely to be synthesized and excreted with the other three enzymes.

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