CHAPTER 8A

NEUROENDOCRINE REGULATION OF BLOOD SUGAR LEVEL IN PIGEON

The reactive hypoglycaemia is a syndrome that results in lowered glucose levels and hyperinsulinemia. Though conclusive evidences are not there, this syndrome is believed to be due to hyperactivity of parasympathetic system (Nielson, 1926). Vagotomy was found to cure this syndrome (Boulet et al., 1954) while anticholinergic drugs were found to be useful in reducing the effect of this syndrome (Portis and Zitman, 1943; Veverbrants et al., 1969). The reactive hypoglycaemia is reflexely mediated whenever a carbohydrate meal is ingested. The earlier works on this syndrome are suggestive of vagal mediation in bringing about hypersecretion of insulin and thereby the hypoglycaemia, and do not consider the direct action of cholinergic system in the uptake of glucose by tissue cells. Recently Mondon and Burton (1971) showed clearly that acetylcholine together with insulin

increases the uptake of glucose by liver. The acetylcholine (ACh) is secreted at sinusoidal linings of the hepatic cords where the parasympathetic-cholinergic nerve plexus are demonstrated (Sutherland, 1964). In the avian liver the sinusoidal linings show the localization of acetylcholinesterase (AChE) (Pilo, 1969; Shah et al., 1972b). These authors suggested that the acetylcholine-acetylcholinesterase (ACh-AChE) system is helpful in the assimilation of glucose by the hepatocytes.

If acetylcholine has a direct effect on the uptake of glucose then a glucose load should increase the secretion of ACh at the hepatic sinusoidal linings. In the present chapter an attempt is made to correlate the ACh secretion in the liver, pancreas and skin in response to glucose administration, through the study of AChE in these tissues in pigeon.

MATERIALS AND METHODS

Adult pigeons (Columba livia) reared and maintained in the aviary of the department were starved for an overnight period and injected with 70 mg glucose per 100 gm body weight in normal saline as 30% solution intravenously. The control pigeons received normal saline only. After the administration of glucose the birds were sacrificed (five

at a time) at regular intervals of 30, 60, 90 and 120 minutes. The pieces of pancreas, liver and skin (from the abdomen region) as well as blood serum were quickly collected and were subjected to AChE and glycogen assays. The AChE determination was carried out using acetylcholine chloride as the substrate following the method described by de la Huerga et al. (1952). The activity is expressed as µ mole ACh hydrolysed per 100 mg of tissue or 1 ml serum per hour. The glycogen was determined using the method described by Seifter et al. (1950). The estimation of glucose in the blood was performed by micromethod described by Folin and Malmros (1929) and expressed as mg/100 ml, while glucose in skin was estimated by method described by Nelson and Somogyi (1944) and is expressed as mg/100 g skin. For comparison the estimations were carried out in the tissues of 48-hrs starved pigeons also.

The histochemical localization of AChE in the liver and pancreas was demonstrated by the method of Koelle and Friedenwald (1949) as modified by Coupland and Holmes (1957) using acetylthiocholine iodide (Sigma Chemical Co., U.S.A.) as the substrate.

RESULTS

The results obtained from the study of acetylcholinesterase in the pancreas, liver and skin after the glucose load (Table 1 and Fig. 1) showed that there is an elevated activity of this esterase by about 30 minutes. AChE value in the skin and pancreas of 48-hrs starved, overnight starved (normal) or control pigeons showed no significant variations. However, AChE activity was considerably low in the liver of 48-hrs starved birds as compared to those of overnight starved or control birds. A concomitant increase was noticed in the level of AChE in the serum at 30 minutes after the administration of glucose. The increased activity of AChE at 30 minutes in these tissues coincided with the peak glucose level in the blood. Likewise, skin glucose also showed maximum concentration at 30 minutes after the glucose administration. The glycogen value in the liver exhibited a corresponding increase at 30 minutes. However, skin glycogen content did not register any increase after glucose load.

In the liver of all pigeons [starved (Fig.2), control (Fig.3) or experimental (Fig.4)] the AChE was found to be localized in the sinusoidal linings corresponding to the parasympathetic nerve plexus. A very low histochemical

CHAPTER 8A

EXPLANATIONS FOR FIGURES

Fig. 1. Graph showing quantitative analyses of acetylcholinesterase activity in the liver, pancreas, skin and serum along with blood and skin glucose levels and glycogen contents of liver and skin in starved, normal and glucose injected pigeons.

Figs. 2 to 6. Photomicrographs showing histochemical localization of acetylcholinesterase activity.

Fig. 2. Section of liver of starved pigeon. 75X.

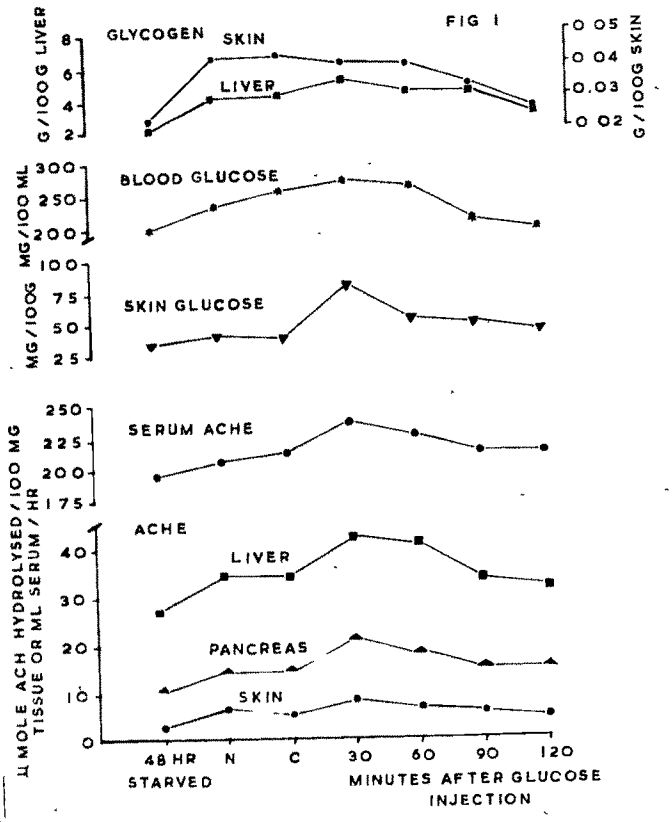
Fig. 3. Section of normal pigeon. 200X.

Fig. 4. Section of glucose injected (30 minutes) pigeon. 200X.

Fig. 5. Section of islet of Langerhans of starved pigeon. 200X.

Fig. 6. Section of islet of Langerhans of glucose injected (30 minutes) pigeon. 200X.

FIG 1


GLYCOGEN
SKIN
LIVER
G/100G LIVER
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6
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G/100G SKIN
BLOOD GLUCOSE
MG/100 ML
300
250
200
SKIN GLUCOSE
MG/100G
100
75
50
25
SERUM ACHE
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225
200
175
ACHE
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PANCREAS
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μ MOLE ACH HYDROLYSED/100 MG TISSUE OR ML SERUM/HR
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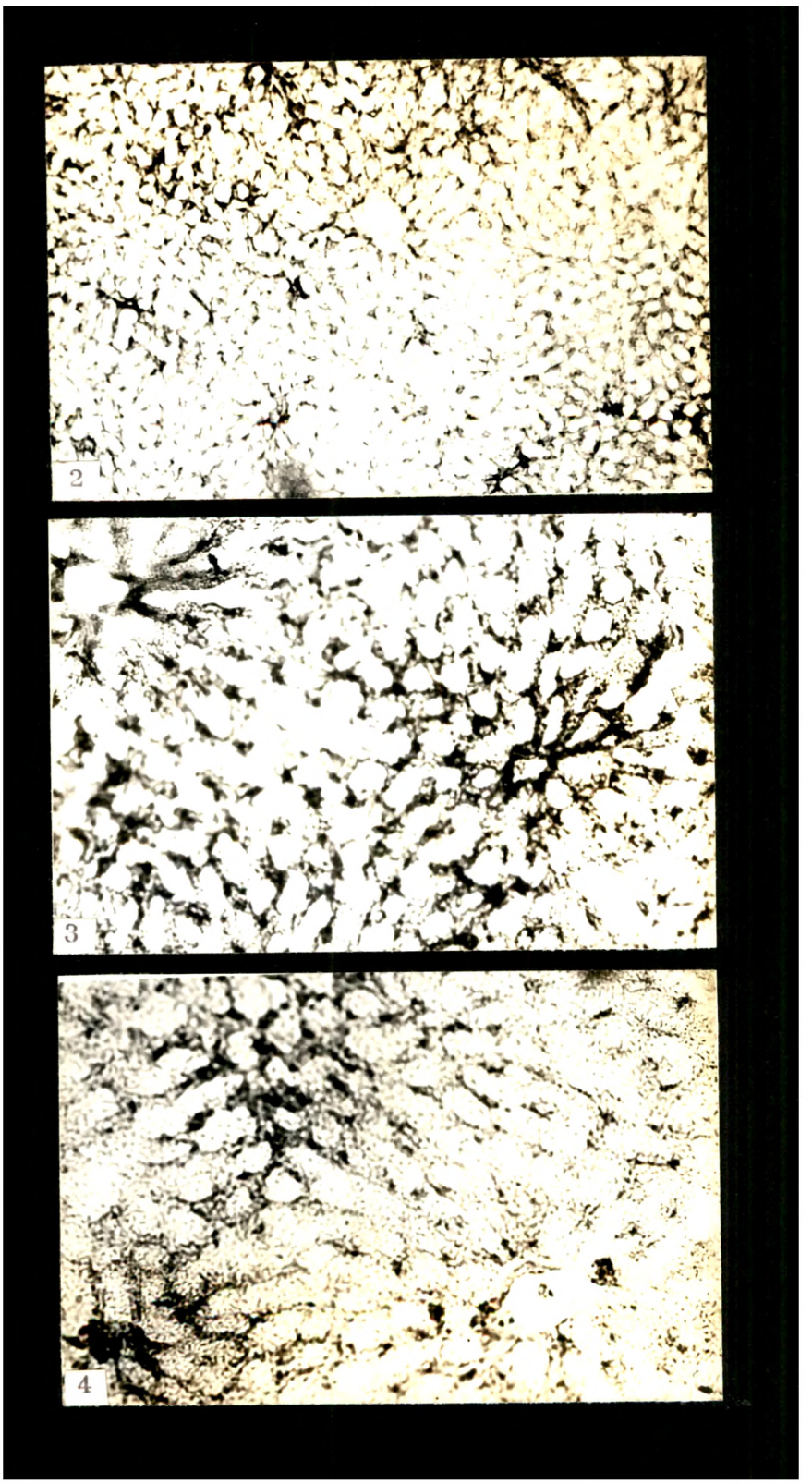
MINUTES AFTER GLUCOSE INJECTION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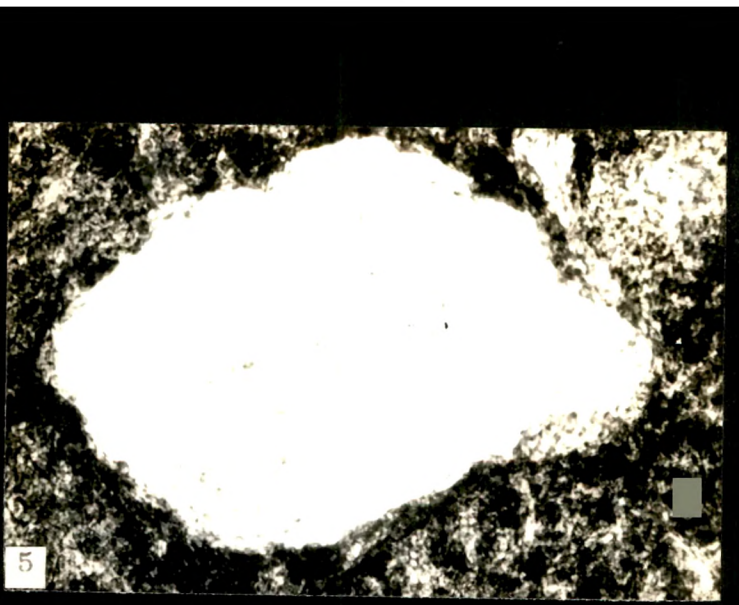

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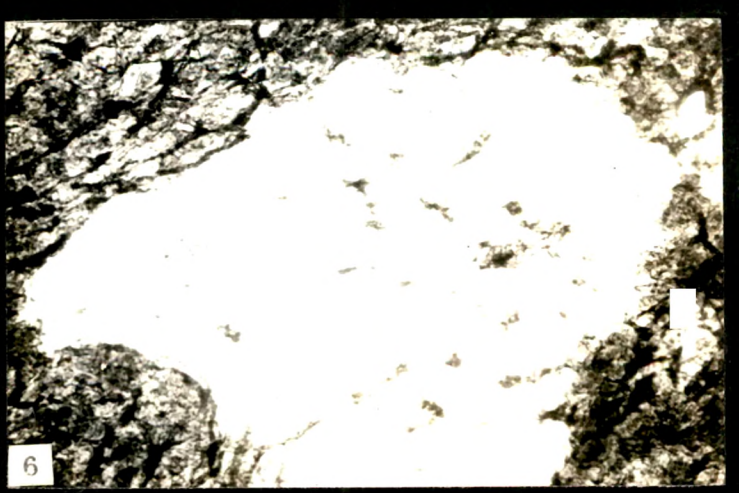

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TABLE 1

Acetylcholinesterase activity and glycogen content in the tissues of domestic pigeon and the corresponding blood and skin glucose values after the administration of the glucose[1]

TIME	ACETYLCHOLINESTERASE[2]				BLOOD[3] GLUCOSE	SKIN[4] GLUCOSE	GLYCOGEN[5]	
	PANCREAS	LIVER	SKIN	SERUM			LIVER	SKIN
48 hr starved	11.036 ± 3.858	26.948 ± 8.486	3.885 ±0.371	194.443 ± 12.026	196.91 ± 29.19	34.04 ± 5.16	2.170 ±0.057	0.0234 ±0.0063
NORMAL	14.812 ± 3.917	34.044 ± 1.741	6.713 ±0.427	206.50 ± 21.43	233.51 ± 27.57	41.22 ± 6.23	4.009 ±0.126	0.0441 ±0.0098
CONTROL	14.483 ± 2.552	36.412 ± 3.866	5.1043 ±0.1048	213.45 ± 18.761	252.32 ± 28.34	40.303 ± 6.23	4.199 ±0.023	0.0441 ±0.0120
30 min.	20.785 ± 6.357	42.778 ± 6.594	7.851 ±0.807	234.84 ± 15.28	268.79 ± 43.32	80.184 ±35.772	5.065 ±1.947	0.0417 ±0.0085
60 min.	18.024 ± 5.0311	40.864 ± 4.758	6.536 ±0.315	225.31 ± 14.89	261.53 ± 31.72	52.648 ±22.710	4.412 ±0.216	0.0411 ±0.0092
90 min.	15.876 ± 4.605	32.168 ± 4.993	5.391 ±0.564	213.48 ± 30.26	206.534 ± 37.685	49.867 ± 6.663	4.209 ±0.311	0.0326 ±0.0082
120 min.	15.528 ± 6.624	31.843 ± 5.795	4.993 ±0.538	212.42 ± 25.74	191.68 ± 12.54	41.475 ±10.735	3.018 ±0.218	0.0276 ±0.0018

1 - 70 mg glucose/100 g body wt. in saline.

2 - µ mole of ACh hydrolysed/100 mg tissue or 1 ml serum/hr.

3 - mg glucose/100 ml blood.

4 - mg glucose/100 g skin.

5 - g glycogen/100 g tissue.

reactivity was noticed in the islets of 48-hrs starved pigeons (Fig.5). However, in the normal, control or experimental birds, the islets exhibited a good reactivity, especially in the islet sinusoidal spaces (Fig. 6).

DISCUSSION

Since the distribution of acetylcholinesterase parallels the distribution of acetylcholine (Feldberg, 1950; Burgen and Chipman, 1951; Bennett et al., 1958; Quastel and Quastel, 1961; Gaddum, 1963), it is reasonable to believe that the elevation of the glucose level in the blood resulted in an increased release of acetylcholine in the liver, pancreas and skin. The ACh must also be finding its way to the serum as the serum AChE too showed an increase at 30 minutes.

In the liver, the AChE was found to be localized in the linings of the sinusoids (Figs. 2-4) where reticular fibres are usually situated. Pilo (1969) and Shah et al. (1972b), observing similar distribution of AChE in the liver of birds, suggested that ACh-AChE system must be aiding the uptake of metabolites by hepatocytes. It has been shown that ACh together with insulin increases the glucose uptake in the liver cells (Mondon and Burton, 1971).

If ACh facilitates glucose transport, it must be through coupling it with the flow of ions (see Wilbrandt, 1975). The ACh can bring about changes in the membrane permeability as well as movement of ions. By coupling with Na^+ - K^+ transport mechanism and utilizing the same energy the glucose molecules could then move across the hepatic cell membrane. The dependence of sugar transport on the presence of Na^+/K^+ was seen in the intestinal absorption of sugar (Riklis and Quastel, 1958; Crane, 1962; Schultz and Curran, 1970). It is also possible that not only Na^+ accelerates sugar transport but sugar in turn can enhance Na^+ transport. If transport of glucose and ions goes hand in hand then the ionic concentration in the hepatic cells should increase along with glucose uptake and glycogen deposition. This has been actually observed in the liver of growing pigeons where increased glycogen deposition was accompanied by not only an increased hepatic AChE activity but also an increased concentration of ions (Chapter 2). The glycogen content of the liver was also maximum at 30 minutes after the intravenous injection of glucose. Thus, in all probability, the major part of glucose entering the hepatic cells of the pigeons must have been through flow coupled transport mediated by ACh. This method of glucose transport eliminates, to a certain extent, the complete dependence of blood sugar level regulation on insulin in birds. Perhaps this is the

reason why in birds the insulin has only a secondary role in the regulation of carbohydrate metabolism while glucagon exerts a powerful control over it (Hazelwood, 1977; Epple, 1977). However, if the glucose entering the hepatic cells is not readily phosphorylated and deposited or disposed off, the flow of sugar molecules across the membrane may get retarded. Insulin, by activating the enzymes such as hexokinase, readily phosphorylates and diverts the glucose molecules that enters the hepatocytes. In all probability, in the avian liver glucose might be phosphorylated only as it enters or after its entry into the cells unlike in mammalian species where chemi-osmotic coupling (phosphorylation) is one of the way by which glucose is drawn into the cells.

Since certain amount of insulin is ~~also~~ necessary for the sugar transport the B-cells also might be stimulated to release the hormone after the glucose load. In most of the mammals, the insulin released by the B-cells could be stimulated by the cholinergic fibres. From the observation that the AChE activity increased in the pancreas after glucose administration, it could be deduced that ACh might be increasingly released in the pancreas. But the nervous stimulation of the B-cells secretion is questionable in birds as several studies have failed to show the presence of nerves in the avian islet tissue (Kern and Grube, 1972;

Kobayashi and Fujita, 1969; Sims et al., 1971; Epple, 1977). Histochemically, the AChE activity was observed in islets in the pigeon pancreas (Fig.6). In the pancreatic islets of the 48-hrs starved pigeon, the AChE activity was very sparingly localized in the linings of the sinusoidal spaces (Fig.5). In the normal (overnight starved), control and experimental (glucose injected) pigeons, the islets showed a considerable amount of AChE activity (Fig.6). However, histochemically a difference in the degree of elicitation of AChE activity between the control or experimental birds was not discernible as in the quantitative estimations. The presence of AChE in the islets is indicative of the ACh either secreted by minute nerve plexus (neuro-insular complexes) or brought by the blood. In fact, the blood should be containing more ACh 30 minutes after administration of glucose as AChE was maximum at this period. Perhaps the insulin release may be triggered by the ACh brought by the blood or by the direct action of glucose on the B-cells.

The increased level of AChE in the blood, observed soon after the glucose administration, must be due to the fact that the AChE is getting released even into the blood stream. Since the ACh secretion takes place at the sinusoidal linings this should be a likely eventuality.

At this time the AChE itself is being released into the blood stream from the liver to counteract the ACh that gets liberated into the blood.

In the skin, the AChE activity showed a maximum concentration at 30 minutes but there was no change in the glycogen content. In the skin the glucose uptake did not coincide with the maximum ACh release even though dermal glucose level was found to increase tremendously by 30 minutes. It may be reasoned here that the function of ACh in the skin is that a vasodialator thereby increasing the dermal glucose compartment. The dermal glucose compartment is long known to be a temporary regulator of blood sugar level by accomodating extra glucose for a short period.

Thus in pigeon ACh plays an important role in the regulation of blood sugar level by initiating the glucose uptake by the liver, by perhaps stimulating the insulin secretion by B-cells of islets of Langerhans as well as by vasodialatory effect on the dermal blood vessels.

CHAPTER 8B

CHOLINERGIC ASSISTANCE TO THE RELEASE AND ACTION OF INSULIN IN RATS

The insulin secretion by the B-cells of the islets of Langerhans is reflexly carried out through the mediation of autonomous nervous system (Woods and Porte, 1974). These reflexes are (1) in response to change in the glucose concentration in the blood, (2) in response to the stimuli related to food (smell, taste etc.) and (3) conditioned through learning. As cholinergic and adrenergic nerve fibres reach the islets both are implicated in the control of hormone secretion from islets. Stimulation of vagus nerve generally elicites secretion of insulin. Chieri et al. (1975) showed that, in dogs, a glucose load to the brain induces pancreatic insulin secretion mediated particularly by the vagus nerves.

Conversely, the rate of insulin secretion can be inhibited by direct neural input to the pancreas and this inhibition is mediated by alpha adrenergic receptors (Miller, 1975). The control of insulin release from the islets is normally the function of glucoregulator centre in the brain which is sensitive to both glucose load and insulin (Szabo and Szabo, 1975a; 1975b).

The cholinergic nerves not only participate in insulin release from pancreas but also figure in increasing glucose uptake facilities in the liver. Mondon and Burton. (1971) reported that acetylcholine together with insulin increases the glucose uptake by the liver cells. The glucose uptake by the cells in general is effected through different mechanisms: (1) carrier mediated transport, (2) flow coupled transport, (3) active transport (4) chemiosmotic coupled transport (phosphorylation) (Wilbrandt, 1975). The hepatic cell membrane, being less sensitive to insulin than the adipose and muscle tissues, perhaps utilizes the flow coupled transport for the uptake of glucose, in which the influx of sugar is coupled with the flow of ions. The acetylcholine (ACh), which is secreted by the cholinergic nerve plexus that reaches the liver lobules, by initiating the flow of ions, thus can facilitate the glucose uptake by the hepatocytes. The secretion of ACh by the nerve plexus can be easily perceived from the fact that

the liver lobules show the presence of acetylcholine hydrolising enzyme, the acetylcholinesterase(AChE) (Pilo, 1969). Gerebtzoff (1959) and Shah et al. (1972b) suggested that ACh-AChE system must be concerned with the assimilation of glucose into the liver cells. However, it is not clear whether or not the glucose load will increase the secretion of ACh in the liver.

The present study is an attempt to find the degree of acetylcholine secretion in pancreas, liver and skin of rat in response to glucose load as well as to assess the influence of acetylcholine in the release and action of insulin. Since the degree of acetylcholine released can be judged by the level of AChE activity, the AChE was quantitatively measured in the pancreas, liver, skin and serum of rats after glucose administration.

MATERIALS AND METHODS

Male albino rats of 100 to 150 gm weight, starved for an overnight period, were injected with glucose (15 mg glucose in 0.5 ml saline) intraperitonially. The control rats received 0.5 ml saline only. Five rats were sacrificed at regular intervals (viz., 30, 60, 90 and 120 minutes) after glucose administration. The pancreas, liver, skin and serum were quickly collected and were subjected to AChE

assay using acetylcholine chloride as the substrate according to the method of de la Huerga et al. (1952). The activity is expressed as μ mole ACh hydrolysed/100 mg tissue or ml serum/hour. Along with the determination of AChE activity, glycogen content of liver and skin as well as glucose concentration in the blood were ~~also~~ measured. Glycogen was estimated by using anthrone reagent according to the method described by Seifter et al. (1950) and glucose was estimated by the micromethod described by Folin and Malmros (1929). For comparison these estimations were also carried out in the tissues of 48-hrs starved rats.

Histochemical demonstration of acetylcholinesterase in the liver and pancreas was carried out by the method of Koelle and Friedenwald (1949) as modified by Coupland and Holmes (1957) using acetylthiocholine iodide as the substrate.

RESULTS

The results are presented in Table 1 and Fig. 1. The acetylcholinesterase activity in the pancreas, liver, skin and serum of 48-hrs starved rats registered the lowest values. Similarly, the blood glucose level and the glycogen content in the liver and skin were also at the minimum in 48-hrs starved rats. The control rats exhibited much higher values of AChE activity, glucose and glycogen. The administration

CHAPTER 8B

EXPLANATIONS FOR FIGURES

Fig. 1. Graph showing quantitative analyses of acetylcholinesterase activity of the liver, pancreas, skin and serum along with the blood glucose level and glycogen contents of liver and skin in starved, normal and glucose injected rats.

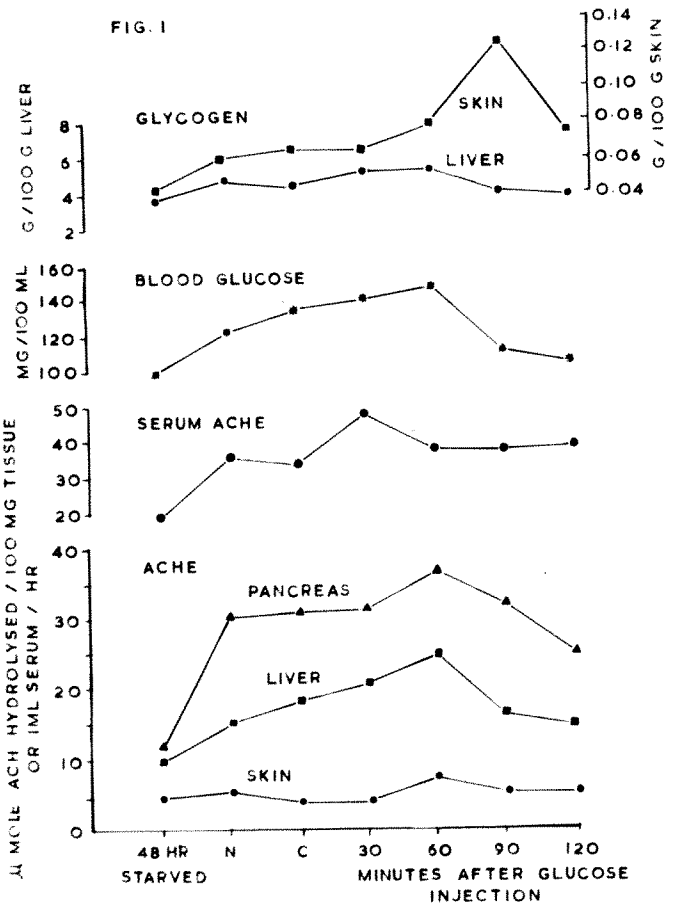
Fig. 2 to 4. Photomicrographs showing histochemical localization of acetylcholinesterase activity.

Fig. 2. Section of islet of Langerhans of normal rat. 200X.

Fig. 3. Section of liver of normal rat. 200X.

Fig. 4. Section of liver of glucose injected (60 minutes) rat. 200X.

FIG. 1


GLYCOGEN
SKIN
LIVER
G / 100 G LIVER
G / 100 G SKIN
BLOOD GLUCOSE
MG / 100 ML
SERUM ACHE
ACHE
PANCREAS
LIVER
SKIN
μ MOLE ACH HYDROLYSED / 100 MG TISSUE OR 1ML SERUM / HR
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MINUTES AFTER GLUCOSE INJECTION

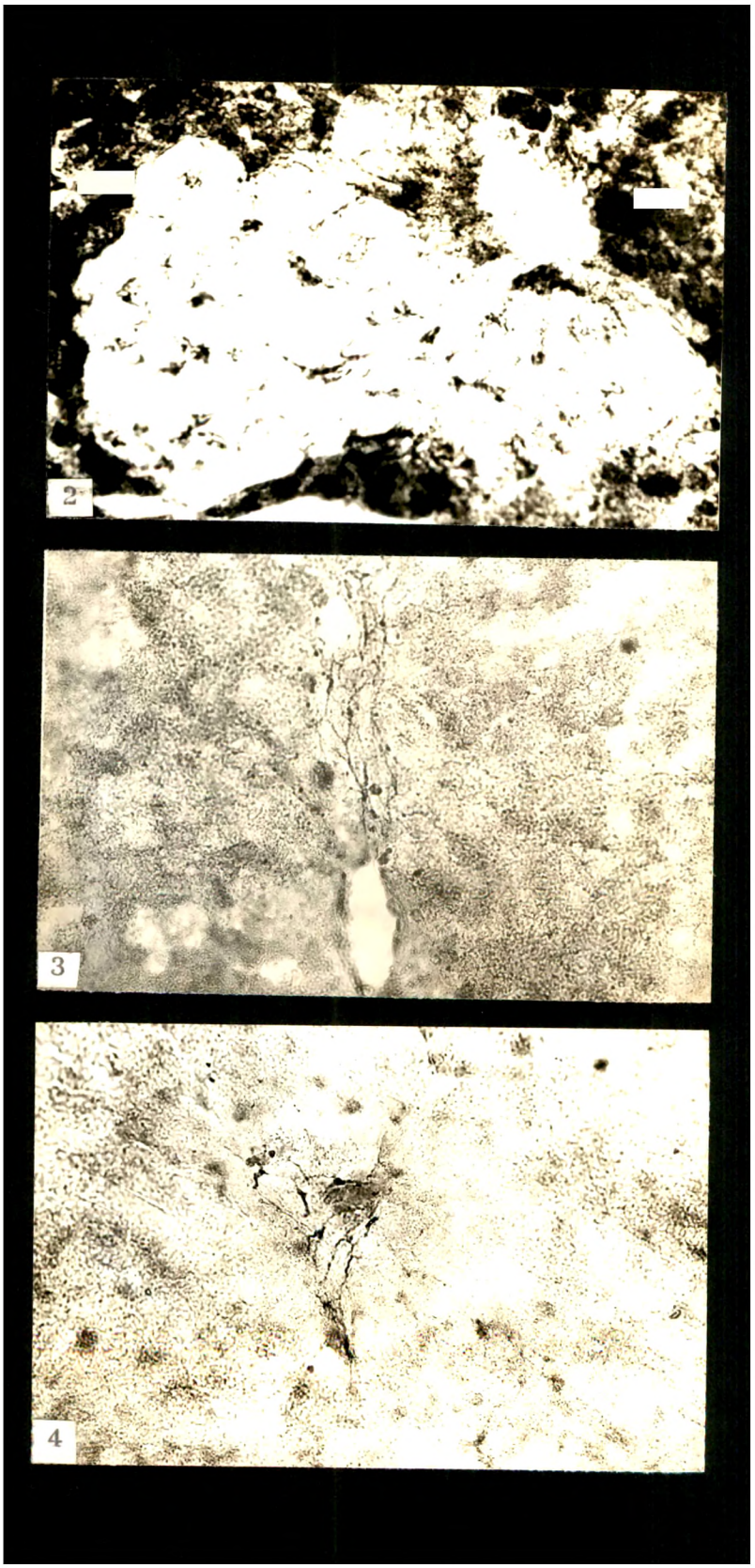
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TABLE 1

Acetylcholinesterase activity and glycogen content in the tissues of rat and the corresponding blood glucose values after the injection of the glucose[1]

TIME	ACETYLCHOLINESTERASE ACTIVITY[2]				BLOOD[3] GLUCOSE	GLYCOGEN[4]	
	PANCREAS	LIVER	SKIN	SERUM		LIVER	SKIN
48-hr starved	12.785 ± 3.673	9.650 ±3.110	4.563 ±1.260	18.901 ± 5.845	92.56 ±11.59	3.567 ±1.345	0.043 ±0.007
Normal	30.054 ± 4.498	15.005 ±4.692	5.247 ±2.669	36.320 ± 3.301	120.00 ±16.64	4.359 ±1.445	0.060 ±0.036
Control	31.043 ± 2.749	18.435 ±1.452	3.495 ±1.234	34.375 ± 5.209	133.223 ± 9.130	4.406 ±1.002	0.065 ±0.015
30 min.	30.867 ± 3.212	20.696 ±4.674	3.369 ±0.103	47.467 ± 7.182	141.306 ±14.451	5.169 ±2.178	0.067 ±0.019
60 min.	36.243 ± 2.037	24.675 ±2.328	6.852 ±1.429	37.080 ± 8.513	145.352 ± 8.889	5.173 ±1.352	0.078 ±0.044
90 min.	31.641 ± 2.675	16.170 ±4.782	4.934 ±0.105	37.216 ± 8.437	110.139 ±24.178	4.091 ±1.489	0.125 ±0.049
120 min.	24.480 ± 1.670	14.120 ±1.726	4.740 ±1.030	36.997 ±10.629	129.428 ±19.400	3.738 ±0.973	0.074 ±0.047

1 - 15 mg glucose in 0.5 ml of saline.

2 - μ mole of ACh hydrolysed/100 mg tissue or 1 ml serum/hr.

3 - mg glucose/100 ml of blood

4 - g glycogen/100 g tissue.

of glucose resulted in a further increase of the values of AChE in all tissues studied and increased the blood sugar and hepatic as well as cutaneous glycogen levels. The maximum AChE activity was observed at 60 minutes after glucose administration in pancreas, liver and skin while in the serum, AChE activity was maximum at 30 minutes. The blood sugar level was maximum at 60 minutes as it was with hepatic glycogen content. The glycogen content of skin, however, reached a peak level only at 90 minutes.

The histochemical studies on the AChE revealed that in the islet tissue of pancreas, this enzyme was found to be diffusely distributed in cells. The minute nerve plexus (intrainsular plexus) were also traceable with AChE technique (Fig. 2). In the liver, acetylcholinesterase was found to be generally diffused in the parenchymal cells but definite plexuses were not visible (Fig.3). The minute branches of portal vessels however, showed AChE reactivity (Fig. 3). When glucose was administered the portal areas showed higher reactivity than the areas around the central collecting vein (Fig.4).

DISCUSSION

The data obtained from the experiments clearly show that the elevation of glucose level in the blood caused a

general stimulation of ACh secretion by the cholinergic fibres reaching the pancreas, liver and skin. The maximum AChE activity observed at 60 minutes in these organs coincided with the maximum blood glucose level. The increased cholinergic activity might be due to the influence of glucose load on the glucoregulator centre of central nervous system (CNS) as Chieri et al. (1975) reported in dogs. It has been shown that, electrical stimulation of the venrolateral hypothalamic nuclei (VLH) causes a decrease of blood glucose (Gellhorn et al., 1941; Kuzuya, 1962; Shimazu et al., 1966), an increase of plasma insulin (Kuzuya, 1962; Steffens et al., 1972) and the release of humoral factor that elicits pancreatic insulin secretion (Idahl and Martin, 1971). Szabo and Szabo (1975a) concluded that it is the cholinergic system that is in operation in the glucoregulator centre of the CNS. In response to a glucose load, the glucoregulator centre (VLH) communicates with pancreas and other tissues through vagus nerve. Vagal stimulation elicites the insulin secretion from islets (Findlay et al.,1969; Porte et al., 1973). The vagal stimulation was also observed to be more effective when glucose level was elevated (Bergman and Miller, 1973; Brilton, 1925). It could be concluded here that the elevated ACh secretion in the pancreas as judged from the increased AChE activity could be mediating partially the release of insulin from B-cells. The acetylcholine is also capable of releasing membrane bound

Ca^{++} into the cytosol. The Ca^{++} as a secondary messenger could also influence the release of insulin together with cyclic adenosine monophosphate (c-AMP) (Porter *et al.*, 1976).

Along with the increased cholinergic activity in the pancreas, the liver and skin also showed increased acetylcholinesterase activity in the corresponding period. Since it is probable that AChE activity is proportional to the ACh release, it could be reasonably assumed that glucose load has brought about an increased release of ACh in these tissues. It has been reported that insulin together with ACh, increases the glucose uptake in the liver cells (Mondon and Burton, 1971). The ACh must be facilitating glucose transport through coupling it with flow of ions. The ACh can cause changes in membrane permeability, release of membrane bound Ca^{++}, as well as an increase in the c-AMP through inhibiting phosphodiesterase (Rasmussen, 1975). By coupling with the Na^{+}-K^{+} transport mechanism (thereby utilizing the same energy) the glucose molecules could then move across the hepatic cell membrane. The dependence of sugar transport on the presence of Na^{+}/K^{+} was reported in the intestinal absorption of sugar (Riklis and Quastel, 1958; Crane, 1962; Schultz and Curran, 1970). It is also possible that not only Na^{+} accelerates sugar transport but

sugar in turn can enhance Na^+ transport. The movement of Na^+ is believed to induce or modify the movement of sugar and other transport molecules either by co-transport or by counter transport (Wilbrandt, 1975).

Histochemically, the localization of AChE in the liver of rats was observed generally in the hepatic cells (Fig.3) unlike in the pigeon liver where it was seen in the linings of sinusoids (Pilo, 1969; Chapter 8a). However, after the glucose administration many blood vessels and sinusoids near the portal area were observed to show increased enzyme activity (Fig. 3). Since, hepatic nerve plexus runs along with the blood vessels, these sites represent the site of ACh release too. So the increase in the AChE activity in the rat liver at 60 minutes after glucose injection also represents the probable increase in the ACh release by the nerve plexus that line the portal vessels as well as the sinusoids near the hepatic portal spaces. Since the ACh is not released all along the sinusoidal linings as in the case of pigeon liver, ACh may not be influencing all the hepatic cells in the uptake of glucose, but only those in the periportal areas. In all probability only a part of glucose enters the hepatic cells through the flow coupled transport facilitated by ACh. In the case of rat liver major part of the glucose that enters the hepatic cells is through other means where insulin also plays a role by activating the membrane bound hexokinase.

While in the liver the maximum AChE activity observed at 60 minutes coincided with maximum glycogen deposition, in the skin these two were not found to coincide at all. The glycogen in the skin was maximum at 90 minutes rather than at 60 minutes. Perhaps in the skin AChE has only a vasodialatory influence.

In the rat liver it has already been seen that the ACh is released by the nerve plexus present along the blood vessles and in all probability some of the ACh might also be finding its way into the blood. The serum AChE is for such contingencies, and this enzyme can readily demolish the ACh released into the blood stream. In fact, serum AChE is also secreted by the liver cells. Since the AChE in the serum is found to become maximally active at 30 minutes, it is possible that this enzyme is released by the liver in anticipation of an increased liberation of ACh by the cholinergic nerve fibres.