AMYLASE ACTIVITY IN THE BLOOD AND PANCREAS OF HYPOPHYSECTOMIZED AND TESTOSTERONEPROPIONATE TREATED MALE ALBINO RATS

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The activity of sex hormones is subjected to complex neuro-humoral regulation, with the participation of the brain cortex, subcortex and more particularly of the centers in the hypothalamus, gonadotropic hormones of the adenohypophysis and interaction with other endocrine glands (2, 11).

In previous studies (5, 7) the authors of the present paper have established the activying effect of the testosteronepropionate on the amylase in blood and pancreas of male rats. We also established the essential role for its effect played by the functional condition of the central nervous system, finding moreover that during an inhibition process this activying effect is not manifested (5), whereas during excitation of the central nervous system with coffeine the activying effect is enhanced (7).

The final goal of the present work is to elucidate the role played by the hypothalamo-hypophyseal system insofar action of male sex hormones is concerned on enzymic activity. With this purpose in view we carried out investigations on the amylase of rats deprived of the hypophysis and hypophysectomized, treated in addition with testosteronepropionate.

Method of investigations

The studies were carried out on 39 male albino rats, with approximately identical age and weight (160—220 gr), distributed into three series: control group (15 rats), hypophysectomized (10 rats) and hypophysectomized, injected with testosteronepropionate (14 rats).

For the operative removal of the hypophysis we resorted to the method as developed by *Ulovich* (10), with a number of improvements proposed by the same author. The hypophyseal tissue removed was histologically and macroscopically proved according to *Ulovich*. The criterium adopted as indication for the complete removal of the hypophysis was the decrease of the rat's weight over a period of 3 months postoperatively.

Following hypophysectomy the test animals underwent a regime assumed as optimal insofar reduction of mortality rate is concerned (10, 17), rich in proteins and vitamins diet, giving 5% glucose solution instead of water in the first postoperative month and maintaining environmental room temperature up to 26—28°C.

The test animals were studied three months after removal of the hypophysis, and 14 were additionally treated with oil solution of testosteronepropionate, with a total dosage 1 mg per 100 gr body weight, administered intramuscularly over a period of 10 days. Deterioration of the general condition was noted in the rats receiving testosteronepropionate. Three fatal cases also occured, which were omitted from the present investigation.

The animals of all three series were sacrificed by decapitation in a predetermined hour of the day (11 a. m.), keeping them for 15 hours previous to killing without food. The method of investigation is described with more details in previous publications by the same authors (5, 6). The blood for study was promptly obtained, and similarly without delay whatever the pancreas, seminal vesicles and prostate gland were removed and accordingly weighed.

The amylase activity was established according to the modifications of the *Engelhardt-Gerchuk* method (3), as proposed by *Kalitzin* (5, 6) for the blood amylase and according to the *Balzer-Schuster* technique (12) for the pancreas amylase.

Each of the results obtained represents the mean arithmetical value of four parallel determinations (5, 6).

Results

The data obtained were subjected to statistical elaboration (1). The results are presented in tables 1 and 2.

The results illustrated in table 1 show:

1. Decrease of amylase activity in hypophysectomized animals (series II) as compared to controls: in the blood with 46,9%, in the pancreas — 38,2%.

2. The hypophysectomized rats, injected with testosteronepropionate (series III) disclose a further lowering of amylase activity: in the blood with 62%, in the pancreas with 77,5% lower as compared to control animals.

The differences set forward in points 1 and 2 are statistically reliable ($P = 0,05$).

3. The large fluctuations in the activity of pancreatic amylase in the experimental rats are worth mentioning (series II and III). Moreover, lower amylase activities of the pancreas as well as a more substantial reduction in its weight are established in animals hypophysectomized in younger age.

The sequelas of hypophysectomy, already described in literature (8, 10, 21, 22), were also found in the test animals investigated (series II and III): general reduction of weight in the animals, atrophy of the sex glands and secondary sexual organs (prostate and seminal vesicles) and of the pancreas as well. These alterations are demonstrated in table 2.

Table 1

Amylase activity in the blood and pancreatic homogenate of control, hypophysectomized and hypophysectomized testosteronepropionate treated male albino rats

Blood Incubation 2 h. at 37°C pH=6,5 Increase of reduction substances (mg/l ml blood)				Pancreatic homogenate Incubation 30 min. at 37°C pH=6.8 Increase of reduction substances (mg/1mg fresh tissue)			
No of experiment	Control	Hypophysectomized	Hypophysectomized, injected testosteronepropionate	No of experiment	Control	Hypophysectomized	Hypophysectomized, injected testosteronepropionate
1	82,4	62,1	44,8	1	21,38	16,76	0.26
2	104,8	66,4	35,2	2	22,05	15,97	2.36
3	96,8	62,4	37,2	3	22,73	16,98	2.70
4	136,8	42,0	40,8	4	19.98	17,21	4.50
5	142,8	58.4	10,0	5	19.98	17,44	1,35
6	109,6	50,8	50,0	6	20,25	16,87	11,47
7	75,8	68.8	54,4	7	22,05	17,32	5.06
8	147,6	67.6	46.8	8	21,38	0,23	2,70
9	82,8	80,8	56,0	9	21,49	13,28	1,24
10	125,8	55,3	46,0	10	22.28	0,34	7.65
11	96,4	—	43,6	11	23,96	—	9,90
12	95.0	—	23,4	12	20.59	—	4.73
13	131.6	—	48,8	13	20,59	—	9.00
14	86,2	—	36,8	14	21,94	—	5,86
15	126,4	—	—	15	20,70	—	—
$\bar{X}$	109,3	61,5	40,98	$\bar{X}$	21,48	13,24	4,91
$\pm S$	21,3	10,7	12,30	$\pm S$	1,10	6,90	3,50
$\pm S\bar{X}$	5,8	3,4	3,20	$\pm S\bar{X}$	0,41	2,20	0,90

$\pm S$ — mean square deviation of individual experiment.
$\pm SX$ — standard error of mean arithmetical.

Table 2

The effect of hypophysectomy and testosteronepropionate treatment on the total weight, weight of pancreas and secondary sex organs in male albino rats

Series of animals studied	Number of animals	Weight in gr.		Weight of pancreas gr/100 gr weight	Weight of secondary sex organs (prostate and seminal vesicles gr/100 gr weight)
		At operation	During experiment		
Series I	15	—	180±10,5	0,53±0,072	0,98±0,150
„ II	10	214±16,5	170±15,4	0,13±0,010	0,30±0,071
„ III	14	165± 7,30	132± 6,6	0,10±0,003	0,47±0,080

Note: I series — controls; II series — hypophysectomized; III series — hypophysectomized + injected with tesfosteronepropionate.

Discussion

The data illustrated in table 1 show (in rats investigated 3 months after removal of the hypophysis) a reduction of amylase activity in the blood as well as in the pancreas as compared to control animals.

The equal degree of activity inhibition in pancreatic and blood amylase following hypophysectomy is in compliance with the conception for a certain relationship between the two amylases (23), which, anyway, does not rule out the possibility for other sources of blood amylase, as for instance from the liver tissue (20). It is acceptable that the decreased amylase activity in hypophysectomized rats be accounted for by the reduced biosynthesis of this particular enzyme, as the overall growth is reduced and the protein synthesis respectively lowered in the animals deprived of hypophysis. The major changes with respect to weight and amylase activity as well in younger rats subjected to hypophysectomy could very well be interpreted as a manifestation of the age-group peculiarities in the central nervous system and in the hypothalamo-hypophyseal-adrenal regulation of the protein and nucleinic metabolism (4, 9).

In a recently published work, the authors (7) have established the activying effect of testosteronepropionate as regards amylase. A similar activying effect of the testosterone is established in the studies of numerous authors in the past several years, concerining a number of enzymes with the assumption that this action is substantiated by inhibition of the biosynthesis of the enzyme protein component. Against the background of the data accumulated, we could afford assuming that one of the reasons for the decreased activity of amylase in the blood and pancreas in rats, studied 3 months following removal of the hypophysis, is due to the reduced testosterone production in these animals, since for its biosynthesis in the testes participation of gonadotropins is required (14, 15), and in the cortex of adrenals — that of ACTH (13, 15).

However, the activity of blood and pancreatic amylase in hypophysectomized male rats, injected with testosteronepropionate, not only fails full restoration, but is further decreased as compared to the controls. A greater reduction in weight is also marked of the animals and atrophy of the pancreas. The characteristic action of the testosteronepropionate is manifested merely with respect to secondary sex organs, whose growth is enhanced without reaching anyway, the weight of the control rats.

Explanation of these effects for the time being is impossible. It could be assumed however, that in order that testosteronepropionate action be manifested on the enzymes, in our case on the amylase, an intact hypophysis is required.

Moreover, against the background of a disturbed hypothalamo-neurohypophyseal system and disorders in the correlations with other endocrine glands consequent on hypophysectomy (21, 22), very probably the additionally administered testosteronepropionate leads to a number of intricate interactions with other glands, which on their turn also exert a certain influence upon the pancreas and pancreatic amylase, as for instance the thyroid gland (21).

The clarification of these interrelationships could be subject to further investigations.

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АКТИВНОСТЬ АМИЛАЗЫ В КРОВИ И ПОДЖЕЛУДОЧНОЙ ЖЕЛЕЗЕ У ГИПОФИЗЭКТОМИРОВАННЫХ И ТРЕТИРОВАННЫХ ТЕСТОСТЕРОНПРОПИОНАТОМ САМЦОВ КРЫС

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РЕЗЮМЕ

Исследования активности амилазы в крови и поджелудочной железе проведенные у 39 самцов крыс распределены в три серии: 15 контрольных, 10 гипофизэктомированных и 14 гипофизэктомированных и третированных тестостеронпропионатом (общая доза 1 мг/100 г веса, вводимая на три раза в продолжении 10 дней).

Подопытные животные гипофизэктомированы по методу Уловича, исследовались в продолжении трех месяцев после удаления гипофизарной железы. Установлено общее падение веса, а также уменьшение поджелудочной железы и вторичных половых органов. Активность амилазы понижена в сравнении с контрольными животными: в крови на 46,9%, в поджелудочной железе на 38,2%. Гипофизэктомированные крысы, которым впрыскивался тестостеронпропионат продолжают терять в весе и показывают еще более низкую активность амилазы в крови на 62%, а в поджелудочной железе на 77,5% ниже чем у контрольных.

Полученные при опыте данные обсуждаются.