Plasma Catecholamines in Long-Term Diabetics with and without Neuropathy and in Hypophysectomized Subjects

NIELS JUEL CHRISTENSEN

From the Second Clinic of Internal Medicine, Kommunehospitalet, Århus, Denmark

ABSTRACT Employing a precise and sensitive double-isotope derivative technique, plasma catecholamine concentration (PCA) was measured in four groups of subjects: (*a*) long-term diabetics with neuropathy, (*b*) long-term diabetics without neuropathy, (*c*) hypophysectomized long-term diabetics with neuropathy, and (*d*) nondiabetic control subjects. Blood samples were obtained from subjects in the supine and in the standing position.

In nondiabetic control subjects, PCA (mainly noradrenaline) increased from 0.26 ng/ml in the supine position to 0.69 and 0.72 ng/ml 5 and 10 min after assuming the standing position. By plotting this increase in PCA on the *y* axis in a coordinate system vs. increase in pulse rate, PCA was divided into two components: one of these depended on the rise in pulse rate on standing (called CAH) and the other corresponded to the intercept on the *y* axis where rise in pulse rate equals zero (CAP).

Long-term diabetics with neuropathy showed a significant reduction in PCA in both the supine and the standing position. Further analysis demonstrated that CAP was considerably reduced whereas CAH was normal. Long-term diabetics without neuropathy showed normal PCA values.

Surprisingly, hypophysectomized diabetics with neuropathy exhibited mean PCA values in both the supine and the standing position which were similar to those found in the nondiabetic subjects and considerably elevated compared with the findings in the nonoperated, long-term diabetics with neuropathy. Further analysis in terms of CAP and CAH demonstrated, however, that CAP was just as abnormally reduced in the hypophysectomized as it was in the nonoperated patients whereas CAH was considerably increased.

In contrast to the findings in the nonoperated diabetics with neuropathy, the hypophysectomized diabetic patients with neuropathy demonstrated a negative correlation between rise in PCA and blood pressure on standing indicating that the increase in PCA was at least partially a compensatory phenomenon in the interest of the maintenance of a normal level of blood pressure.

An increased sympathetic tone (vasoconstriction) is believed to be at least partially responsible for the increased capillary resistance and decreased capillary permeability occurring after hypophysectomy.

Received for publication 26 July 1971 and in revised form 30 November 1971.

INTRODUCTION

Autonomic neuropathy is a common finding in diabetic patients, particularly in diabetes of long standing. The disease is most pronounced in the lower extremities; it can, however, be demonstrated in most parts of the body. Noradrenaline is released from the sympathetic nerve endings in response to nerve impulses and a certain amount of the noradrenaline secreted is delivered to the blood. The first objective of the present study was to determine whether autonomic neuropathy in diabetics could be revealed by measuring the concentration of plasma noradrenaline during various conditions.

Hypophysectomy reduces the progression of diabetic retinopathy and visual impairment (1, 2). Capillary resistance is also considerably increased in such patients (3, 4). Circulatory changes occurring after hypophysectomy are not well understood. Vasoconstriction is known to take place and might in part explain the beneficial effects of the operation (4). Accordingly, the status of the sympathetic nervous system in hypophysectomized diabetics deserves investigation, and the second aim of this study was to examine whether hypophysectomized diabetics demonstrate any changes in plasma catecholamine concentration.

PROCEDURE

The principal procedure was to measure the concentration of plasma catecholamines in nondiabetic control subjects and in long-term diabetics. Samples were obtained while the subjects rested in the supine position and 5 and 10 min after they had assumed the standing position. The experiments were always performed in the morning after a fast of at least 8 hr. The subjects were not allowed to leave their beds before the experiments. Smoking was prohibited, but water intake was not limited.

Blood was collected from an antecubital vein via an indwelling catheter, and at least 15 min elapsed between the time when the catheter was inserted and withdrawal of the first blood sample. 20 ml of venous blood was collected twice while the subjects rested in the recumbent position, and 20 ml was withdrawn 5 and 10 min after the patients had assumed the upright position. In addition, a small amount of blood was collected for determination of the blood glucose concentration.

To avoid changes in blood volume, isotonic sodium chloride was given intravenously to all test subjects immediately after withdrawal of a blood sample.

The blood pressure and pulse rate were measured before and 1 min after the patients had assumed the standing position and again just before collection of blood samples in the upright position.

The vibratory perception threshold was measured in the big toe. Three measurements were performed on each side, and the mean of the six values used in the calculations.

In two cases, arterial and venous blood was obtained simultaneously while the subjects rested in recumbency and again 5 and 6 min after the subjects had assumed the standing position.

Some subjects were re-examined, 20 ml of venous blood being collected 1, 3, 5, and 10 min after the upright position had been attained. On this occasion isotonic sodium chloride was not given.

PATIENTS

A total of 34 subjects were examined, 4 of whom were re-examined. The general procedure as outlined above was performed in the following groups of subjects. (*a*) Seven nondiabetic control subjects (cases 1–7) with a mean age of 42 yr. None of them had diseases of the cardiovascular system; (*b*) nine long-term diabetics (cases 8–16) with neuropathy as judged by measurements of the vibratory perception threshold in the big toe; (*c*) six long-term diabetics (cases 17–22) without neuropathy; and (*d*) eight hypophysectomized long-term diabetics (cases 23–30) with neuropathy.

The diabetic patients were treated with diet and insulin. None of them had ketosis at the time of examination, and all of them exhibited a good to fairly well-controlled metabolic state at the time of examination. On the other hand, hypoglycemia was avoided.

The hypophysectomized patients were examined from 3 to 10 yr after operation (mean, 7 yr). A detailed description of the operation as well as the results of follow-up examination have been reported by Lundbæk et al. (1). All the patients had proliferative retinopathy at the time of the operation. Pituitary ablation was performed via the transsphenoidal approach. All patients developed signs and symptoms of hypothyroidism postoperatively, whereafter thyroid substitution was instituted with desiccated thyroid extract 60 mg 3–5 times daily. In addition, the patients received replacement therapy with cortisone. The mean decrease in basal metabolic rate despite treatment with thyroid hormone was 15%.

One hypophysectomized patient was re-examined and two additional control subjects (cases 31 and 32) examined employing the general procedure outlined above in order to measure separately plasma adrenaline and plasma noradrenaline in the upright position.

Two control subjects (cases 33 and 34) were investigated for venous-arterial differences in plasma catecholamine concentration in the supine and upright position.

METHODS

For the measurement of total plasma catecholamine (PCA)[1] concentration, i.e. the sum of adrenaline and noradrenaline, and for the separate determination of plasma adrenaline and plasma noradrenaline, the double-isotope derivative technique described by Engelman and Portnoy (5) was employed with minor modifications. This method permits the determination of the plasma catecholamine content in a sample without the use of standard solutions. We have, however, in each analysis included one or two samples containing a known amount of catecholamine, 0.8 ng/ml noradrenaline for the determination of PCA and 0.6 ng/ml of adrenaline and of noradrenaline for separate determination of adrenaline and noradrenaline, in order to assure continuous control of the method. 10 ml of plasma was used in each analysis.

The precision of the method was calculated on the basis of 19 double determinations performed on 19 consecutive days. The samples contained 0.8 ng/ml of noradrenaline and 10 ml of the sample was used in each analysis. The standard deviation of the single determination was ±0.03 or 4%. The mean recovery was 99 ±8% (standard deviation of the single recovery).

The glucose oxidase method was used to determine blood glucose concentration (6).

Arterial blood pressure was measured in the arm employing the indirect auscultatory method with a mercury manometer.

Vibratory perception threshold was measured in the big toe with a biothesiometer (Bio-Medical Instrument Co., Newbury, Ohio). The threshold is expressed in volts, 50 v on the biothesiometer corresponding to the maximal amplitude of oscillations. The vibratory perception threshold in the lower extremities increases slightly with age in the normal population (7). Values obtained in a large group of diabetics have been presented elsewhere (8). A threshold value of 20 v was considered indicative of neuropathy. As previously shown, there is a strong correlation in individual diabetic patients between the presence of autonomic neuropathy as judged by circulatory studies in the feet and abnormalities of vibratory sensation measured in the same place (9).

Conventional probability levels of significance were used in the statistical analysis, a P value greater than 0.05 being considered nonsignificant.

---

[1] *Abbreviations used in this paper:* CAH, one component of PCA which depends on the rise in pulse rate on standing; CAP, a component of PCA where rise in pulse rate equals zero; PCA, plasma catecholamine concentration.

TABLE I

Nondiabetic Control Subjects

Case	Sex	Age	VPT	Pulse rate/min				Blood pressure/mm Hg				PCA ng/ml		
				0	1	5	10	0	1	5	10	0	5	10
		yr	*v*	*min*				*min*				*min*		
1	m	52	15	48	56	60	68	120/80	130/90	120/90	120/100	0.26	0.64	0.53
2	m	22	10	88	124	108	108	120/80	110/90	110/90	110/80	0.24	0.67	0.75
3	f	48	16	52	52	52	56	130/80	130/100	130/90	130/90	0.31	0.58	0.58
4	m	39	11	68	72	72	84	140/110	150/110	150/110	150/110	0.31	0.69	0.82
5	m	38	12	68	84	96	?	120/80	130/100	120/90	?	0.36	1.00	?
6	f	47	25	72	108	96	96	110/70	120/90	110/80	110/90	0.12	0.58	0.79
7	m	46	13	52	68	64	64	130/90	140/100	140/100	140/100	0.24	0.66	0.87
Mean		42	15	64	81	78	79	124/84	130/97	126/93	127/95	0.26	0.69	0.72
±SD				14	27	22	20	10/13	13/8	15/10	16/10	0.09	0.15	0.13

Pulse rate, blood pressure, and total plasma catecholamine concentration (PCA) in recumbency and in the upright position. Pertinent clinical data are also included. VPT, vibratory perception threshold in the big toe. (?) denotes that the measurement or the blood sample could not be obtained because the subject fainted.

RESULTS

Nondiabetic subjects. Pertinent clinical and laboratory data appear in Table I; Fig. 1 presents the PCA concentration in recumbency and in response to standing. The mean value in the supine position averaged 0.26 ng/ml plasma and rose to 0.69 and 0.72 ng/ml 5 and 10 min after assuming the standing position. The blood pressure and pulse rate showed the expected rise in response to standing. One of the subjects fainted after the 5 min sample had been taken and this person showed the highest rise in PCA and the highest pulse rate on standing. One of the control subjects (case 6) had a rather high vibratory perception value in the big toe, but the PCA response to standing was not different from that found in the other patients.

Long-term diabetics with neuropathy. The mean value for PCA in the recumbent position averaged 0.11 ng/ml and differed significantly from the mean value obtained in the nondiabetic control subjects (P less than 0.001) (Table II); on standing the PCA level was significantly reduced compared with the level obtained in the controls (P less than 0.001 [5 min]; 0.001 [10 min]). None of the control subjects had a PCA level less than 0.5 ng/ml after 5 min of standing, whereas this was seen in all of the long-term diabetics with neuropathy. PCA concentrations were similar in the 5- and 10-min samples in most of the subjects examined. However, there was a large difference in case 14. This subject also showed a considerable increase in pulse rate between measurements taken after 5 and 10 min of standing. Four of the nine patients had a decrease in systolic blood pressure in the upright position of at least 20 mm Hg. These patients had a lower mean value of PCA compared with those with a normal blood pressure response, and the 10-min values are significantly different (P less than 0.02). All diabetic patients with neuropathy also demonstrated proliferative retinopathy.

Long-term diabetics without neuropathy. The mean values for PCA were not different from the values obtained in the control subjects (Table III). One of the subjects showed an abnormal fall in blood pressure in response to standing and this person exhibited a pronounced increase in pulse rate and also showed the largest rise in PCA on standing. Three of the patients had proliferative retinopathy, one showed slight changes in the retina, and two had no abnormalities.

From the data presented above, it appears that there is a correlation in long-term diabetic patients between threshold values for vibratory perception in the lower extremities and rise in PCA in response to standing (P less than 0.001) (Fig. 2). It should be emphasized that this correlation is due to an agreement in these values in individual diabetic patients and not due to a

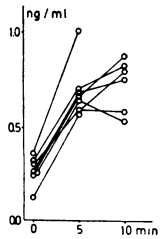

FIGURE 1 Nondiabetic control subjects. Total plasma catecholamine concentration (ng/ml) in recumbency and 5 and 10 min after assuming the standing position.

TABLE II

Long-Term Diabetics with Neuropathy

Case	Sex	Age	Duration	VPT	Pulse rate/min				Blood pressure/mm Hg				PCA ng/ml			Blood glucose
					0	1	5	10	0	1	5	10	0	5	10	
		yr	*yr*	*v*	*min*				*min*				*min*			*mg/100 ml*
8	m	55	30	28	68	70	72	72	150/80	130/80	130/80	130/80	0.09	0.20	0.25	87
9	f	42	25	20	84	100	104	108	170/100	130/80	130/80	120/80	0.13	0.30	0.25	143
10	m	41	25	20	78	82	94	97	170/90	170/80	170/90	170/90	0.21	0.46	0.46	164
11	m	51	21	32	84	96	102	100	140/80	140/90	140/90	140/90	0.10	0.26	0.36	71
12	m	43	31	45	88	96	96	100	170/80	170/90	170/90	?	0.08	0.15	0.22	271
13	m	31	31	25	60	88	84	84	130/90	130/100	130/100	130/100	0.18	0.35	0.43	239
14	m	37	27	30	72	100	104	116	150/100	150/110	150/110	150/110	0.08	0.37	0.61	163
15	m	59	41	38	48	64	68	72	120/70	120/80	120/80	110/80	0.08	0.30	0.37	209
16	f	26	14	35	80	104	104	104	120/70	120/80	120/80	80/?	0.08	0.35	0.36	165
Mean		43	27	30	74	89	92	95	147/84	140/88	140/89		0.11	0.30	0.37	168
±SD					13	14	14	16	21/11	19/11	19/11		0.06	0.09	0.12	65

Pulse rate, blood pressure, and total plasma catecholamine concentration (PCA) in recumbency and in the upright position. Pertinent clinical data are also included. VPT, vibratory perception threshold in the big toe. (?) denotes that the measurement or the blood sample could not be obtained because the subject fainted.

common dependency of both parameters on the duration of diabetes.

Hypophysectomized long-term diabetics with neuropathy. Obviously these patients had proliferative retinopathy but they also demonstrated neuropathy. It was not possible to obtain a group of hypophysectomized long-term diabetics with normal vibratory sensation. The mean vibratory threshold was slightly higher in these patients than it was in the nonoperated patients with neuropathy (Table IV). The hypophysectomized patients did not show the low PCA values that were expected (Table IV). The mean recumbent value was 0.28 ng/ml plasma and 0.74 and 0.73 ng/ml, 5 and 10 min after rising. These values are similar to those found in the nondiabetic control subjects and in the diabetics without neuropathy, but they differ significantly from the values obtained in the nonoperated patients with neuropathy (P less than 0.01 [0 min]; 0.01 [5 min]; 0.01 [10 min]). Of the 13 values obtained in the upright position in these patients, all except two were above 0.5 ng/ml plasma. It can be seen from Table IV that the variance is rather large; some patients demonstrated values just below 0.5 ng/ml whereas in other very large increments were seen. It is obvious that this cannot be explained by a difference in the degree of neuropathy. The hypophysectomized patients showed the highest pulse rate in the supine as well as in the upright position. Comparing mean values in the four groups by variance analysis, the difference between mean pulse rates were not significant. There seems to be a relationship between pulse rate and PCA level and this will be discussed in more detail below.

TABLE III

Long-Term Diabetics without Neuropathy

Case	Sex	Age	Duration	VPT	Pulse rate/min				Blood pressure/mm Hg				PCA ng/ml			Blood glucose
					0	1	5	10	0	1	5	10	0	5	10	
		yr	*yr*	*v*	*min*				*min*				*min*			*mg/100 ml*
17	m	33	26	15	84	96	88	96	130/90	140/100	140/110	130/100	0.28	0.89	0.92	142
18	m	32	20	15	76	108	112	132	120/80	120/90	120/80	120/90	0.14	0.42	0.55	392
19	m	43	39	18	60	76	72	72	130/80	140/100	140/90	130/90	0.31	0.69	0.72	221
20	m	33	32	12	68	84	84	88	120/80	120/90	120/90	120/90	0.33	0.60	0.52	111
21	m	60	23	13	68	88	88	88	140/80	130/90	130/90	130/90	0.30	0.72	0.72	237
22	f	32	20	15	72	120	116	136	120/80	110/90	120/90	90/80	0.17	1.16	0.90	113
Mean		39	27	15	71	95	93	102	127/82	127/93	128/92	120/90	0.26	0.75	0.72	203
±SD					8	16	17	26	8/4	12/5	10/10	15/6	0.09	0.26	0.17	107

Pulse rate, blood pressure, and total plasma catecholamine concentration (PCA) in recumbency and in the upright position. Pertinent clinical data are also included. VPT, vibratory perception threshold in the big toe.

TABLE IV

Hypophysectomized Long-Term Diabetics with Neuropathy

Case	Sex	Age	Duration	VPT	Pulse rate/min				Blood pressure/mm Hg				PCA ng/ml			Blood glucose
					0	1	5	10	0	1	5	10	0	5	10	
		yr	*yr*	*v*	*min*				*min*				*min*			*mg/100 ml*
23	m	45	27	47	68	84	84	76	120/80	120/90	120/80	100/?	0.23	0.55	0.58	154
24	m	38	32	26	84	108	108	124	170/120	170/130	170/130	170/130	0.27	0.78	0.87	101
25	m	34	29	50	88	112	112	?	150/100	140/100	?	?	0.51	1.04	?	233
26	m	36	28	50	80	80	84	82	130/90	140/100	130/100	130/90	0.21	0.42	0.57	139
27	m	43	40	28	72	112	116	116	130/70	100/70	110/70	110/70	0.28	1.11	?	130
28	f	31	15	46	72	108	104	?	140/100	120/100	120/100	?	0.18	0.86	?	290
29	f	53	25	33	76	84	92	104	140/80	140/80	130/80	110/70	0.18	0.39	0.54	168
30	f	38	28	28	88	120	120	128	130/90	120/90	110/90	90/80	0.38	0.79	1.07	257
Mean		40	28	39	79	101	103	105	139/91	131/95			0.28	0.74	0.73	184
±SD					8	16	14	22	16/16	21/18			0.10	0.27	0.23	68

Pulse rate, blood pressure, and total plasma catecholamine concentration (PCA) in recumbency and in the upright position. Pertinent clinical data are also included. VPT, vibratory perception threshold in the big toe. (?) denotes that the measurement or the blood sample could not be obtained bacause the subject fainted.

Furthermore, the 5 min PCA value in the hypophysectomized patients is correlated to a decrease in blood pressure on standing (P less than 0.02) (Fig. 3). Despite the large increase in PCA in the standing position, many of the hypophysectomized patients exhibited an abnormal fall in blood pressure.

Pulse rate and PCA concentration. A significant relationship was obtained between the rise in pulse rate and the rise in PCA concentration with regard to the 5-min values. This was observed in the control subjects, in the long-term diabetics with neuropathy and in the hypophysectomized patients (Fig. 4). A significant correlation was also found in the long-term diabetics with neuropathy when the 10-min values were used. Table V presents a closer analysis of the relationship obtained between rise in PCA and rise in pulse rate. The total rise in PCA after 5 min in the upright position can be divided into two components. The first component (CAP) is the value obtained at the point where the regression line associating rise in pulse rate with rise in PCA intersects the y-axis (Fig. 4), corresponding to no rise in pulse rate after the attendance of the standing position. The second component (CAH) is dependent on the rise in pulse rate and can be calculated from the regression line. In the nondiabetic, control subjects the total rise in PCA was 0.43 ng/ml plasma, with CAP equaling 0.29 ng/ml.

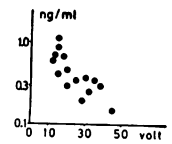

FIGURE 2 Total plasma catecholamine concentration (ng/ml) plotted on the ordinate on a log scale against the vibratory perception threshold (volts) in the big toe. Results obtained in nonhypophysectomized, long-term diabetics with and without neuropathy.

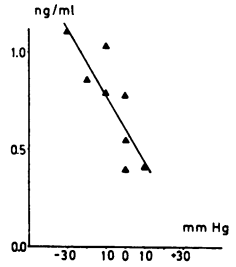

FIGURE 3 Ordinate, total plasma catecholamine concentration (ng/ml) (5 min value). Abscissa, change in systolic blood pressure (mm Hg) after assuming the standing position (1 min value). Results obtained in hypophysectomized, long-term diabetics with neuropathy.

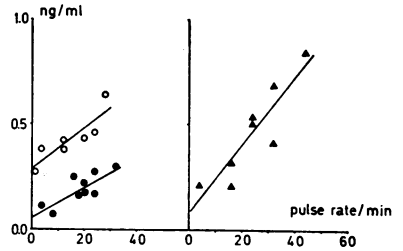

FIGURE 4 The relationship between increase in total plasma catecholamine concentration (ng/ml) and rise in pulse rate after 5 min in the standing position. The regression lines are also plotted on the figure. Left upper curve: (○), nondiabetic control subjects; left lower curve: (●), long-term diabetics with neuropathy; right curve: (▲), hypophysectomized, long-term diabetics with neuropathy.

TABLE V

Relationship between Increase in PCA and Rise in Pulse Rate

	Equation of the regression line	*P* less than	Total rise in PCA	Increase in pulse rate/min	CAP	CAH
			ng/ml		*ng/ml*	*ng/ml*
Nondiabetic subjects	$y = 0.0096x + 0.29$	0.01	0.43	14	0.29	0.13
Long-term diabetics with neuropathy	$y = 0.0069x + 0.06$	0.02	0.19	18	0.06	0.12
Hypophysectomized diabetics with neuropathy	$y = 0.0160x + 0.08$	0.01	0.46	24	0.08	0.38

Results of analysis of the relationship between increase in pulse rate (x) on standing (5 min) and rise in total plasma catecholamine concentration (PCA) (y). The P value indicates the probability that the slope of the regression line differs significantly from zero. CAP, the intersection between the regression line and the y axis. CAH, the slope multiplied by the mean rise in pulse rate.

This latter value differs significantly from zero (P less than 0.001). The pulse-related component averaged 0.13 ng/ml, or 31% of PCA, corresponding to a mean rise in pulse rate of 14. In the long-term diabetics with neuropathy, the CAP component is considerably reduced, being only 0.06 ng/ml. This value differs significantly from the corresponding value obtained in the control subjects (P less than 0.002). The CAH component is normal (0.12 ng/ml) in the long-term diabetics with neuropathy, but it should be noted that this is partly due to the slightly higher pulse rate in these patients. The slope of the regression line was less in these patients than in the nondiabetic control subjects; the difference was, however, not significant. In the hypophysectomized patients the total rise in PCA in response to standing was normal. The CAP component was, however, just as abnormal as in the nonoperated diabetic patients with neuropathy whereas the CAH component was considerably increased. This increase is readily explained by the steeper slope of the regression line associating rise in PCA with rise in pulse rate. The higher pulse rate of the hypophysectomized patients in the upright position is of little importance in this regard. There was a significant difference between the slopes of the regression lines in the hypophysectomized patients and in the nonoperated patients with neuropathy (P less than 0.05). PCA values were clearly higher in the hypophysectomized patients, also when their higher pulse rate is taken into account (P less than 0.01). In the hypophysectomized patients the total rise in PCA in response to standing was similar to that seen in the nondiabetic control subjects, but the mutual relationship between CAP and CAH was not the same.

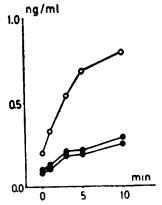

FIGURE 5 Ordinate, total plasma catecholamine concentration (ng/ml). Abscissa, time after assuming the standing position (min). (○), nondiabetic control subject; (●), two long-term diabetics with neuropathy.

It should be noted that the PCA concentrations obtained in the hypophysectomized patients in the supine position were also considerably increased in comparison with the nonoperated long-term diabetics with neuropathy. It was not possible to analyze these data in terms of CAP and CAH. There is, however, a significant relationship between PCA in recumbency in the hypophysectomized patients and the absolute values of the pulse rate (P less than 0.05). This relationship was not found in the three other groups investigated.

PCA after more frequent sampling. The experiments were performed in two long-term diabetics with neuropathy and in one nondiabetic subject (Fig. 5). In the normal person, PCA rose steeply during the first few minutes after the upright position was attained and thereafter a level is reached. In the long-term diabetics with neuropathy, the values for PCA were low throughout the period of measurement.

Separate determination of plasma adrenaline and noradrenaline. Measurements were performed on one of the sample obtained in the supine position in four control subjects and in three of the hypophysectomized patients (Table VI). As expected, noradrenaline constituted the largest part of PCA in the control subjects, but this was also the case in the hypophysectomized

patients. Plasma adrenaline and noradrenaline were also determined on samples obtained in the standing position in two control subjects (cases 31 and 32) and in one of the re-examined hypophysectomized patients. The rise in PCA was found to be due to a rise in plasma noradrenaline in both the control subjects and the single hypophysectomized patient examined (Table VI).

Venous-arterial difference in the forearm. This experiment was performed in order to examine the possibility that the CAP component in the control subjects (equal to 0.29 ng/ml) represented the venous-arterial difference. This value is very large compared with the fraction of PCA which is due to adrenaline and therefore PCA was measured in most of the arterial and venous samples. The study was performed in two nondiabetic persons (case 33 and 34). Arterial blood was obtained from the left brachial artery and venous blood in the usual way from the right arm. Blood was collected while the subjects rested in the supine position and 5 and 6 min after they assumed the upright position. There was a slight, negative difference in venous-arterial concentration of plasma noradrenaline in the recumbent position (Table VII). The PCA values were rather similar in the standing position although the arterial concentration was slightly higher than the venous concentration. It is, therefore, obvious that CAP does not represent the venous-arterial difference.

Blood glucose concentration. The results of these determinations are given in Table II, III, and IV. It can be seen that none of the diabetic patients had hypoglycemia at the time of the examination. The mean values obtained in the different groups do not differ significantly. No significant correlation was obtained between blood glucose concentration and PCA.

TABLE VI

Separate Determination of Plasma Adrenaline and Noradrenaline (Mean Values) in Hypophysectomized Long-Term Diabetics with Neuropathy and in Nondiabetic Control Subjects

	Recumbent position		
	A	NA	A/NA + A
	ng/ml	*ng/ml*	%
Hypophysectomized patients (n = 3)	0.03	0.27	9
Nondiabetic controls (n = 4)	0.05	0.21	14

	Standing position			
	5 min		10 min	
	A	NA	A	NA
	ng/ml			
Hypophysectomized patient (n = 1)	0.01	1.22	0.02	1.01
Nondiabetic controls (n = 2)	0.03	0.73	0.04	0.76

TABLE VII

Venous-Arterial Difference in the Forearm

		Supine position		Standing position			
				5 min		6 min	
Case		Artery	Vein	Artery	Vein	Artery	Vein
33	NA	0.23	0.21	0.59	0.60	0.69	0.65
	A	0.11	0.03				
34		0.23	0.26	0.49	0.45	0.54	0.45

Total plasma catecholamine concentration (ng/ml) and in one instance plasma noradrenaline and adrenaline estimated on samples obtained from the brachial artery and an antecubital vein. Values obtained in the supine position and in the upright position in two nondiabetic control subjects.

DISCUSSION

The circulatory mechanisms brought into play on assumption of the upright position consist fundamentally of two components, vasoconstriction and cardiac acceleration. These changes are mediated via the sympathetic nervous system in order to maintain an adequate level of blood pressure. The increase in PCA in response to standing also consists of two components. It appears from Fig. 4 that the regression line has a considerable positive intercept on the *y* axis (CAP) and there is a positive straight-line correlation between rise in pulse rate and increase in PCA on standing (CAH). CAP is probably derived from the sympathetic nerve endings around the vessels and particularly those of the lower extremities. This assumption is supported by the observation that this PCA fraction was considerably reduced in the long-term diabetics with neuropathy. Clinical and physiological studies have repeatedly shown that diabetic neuropathy is most severe in the lower extremities and lower part of the body. In the present study a correlation was obtained between PCA and vibratory perception threshold measured in the big toe. It has previously been shown that diabetics with decreased vibratory sensation in the feet also show autonomic neuropathy from the functional point of view (9). CAH correlated to the heart rate and might represent noradrenaline release from the sympathetic nerve endings in the heart. However, the cardiac fraction of PCA is large, approximately 30%, compared with the

fraction of the cardiac output which makes up coronary blood flow, but it must be remembered that the heart has a rather pronounced sympathetic innervation. Our data indicate that CAP is rather constant from person to person whereas CAH is a variable factor.

Four of the nine long-term diabetics with neuropathy showed an abnormal fall in blood pressure in response to standing and these patients had significantly lower PCA values than the patients with neuropathy and a normal blood pressure response to standing.

The hypophysectomized diabetics with neuropathy did not show the expected reduced values of PCA, despite a degree of neuropathy somewhat worse than in the nonoperated patients. Mean PCA values after hypophysectomy were similar to the values obtained in the nondiabetic control subjects both in recumbency and in response to standing. However, in terms of CAP and CAH the situation differed from that of the nondiabetic control subjects. CAP was very abnormal in the hypophysectomized patients and thus it cannot be presumed that the autonomic neuropathy had disappeared after the operation. The correlation obtained between decrease in blood pressure on standing and rise in PCA suggests that the change is a compensatory phenomenon, i.e., an attempt to maintain a normal blood pressure.

It should be emphasized that the increased level of PCA in the hypophysectomized patients cannot be a simple consequence of the decrease in blood pressure in response to standing. The same degree of blood pressure abnormality was seen in some of the nonoperated patients with neuropathy and these patients had in fact the lowest PCA values. Furthermore, PCA was also elevated in the supine position in the hypophysectomized patients where blood pressures were identical with those of the nonoperated patients with neuropathy. It is unlikely that the increased levels of PCA in the hypophysectomized patients are due to a reduced degrading rate of PCA compared with the diabetic control subjects. Neural uptake is not impaired in hypophysectomized rats (10).

Circulatory changes in hypophysectomized diabetic patients are not well understood and observations are complicated by the effects of diabetes of long standing on vascular and nervous function. Landsberg and Axelrod (10) observed an increased turnover of noradrenaline in the heart and spleen of hypophysectomized rats and normalization occurred after treatment with thyroid hormone and restoration of normal adrenal cortical function. The increased noradrenaline turnover was mediated by an increase in the sympathetic nervous activity. They also observed a correlation between noradrenaline turnover and blood pressure in the rats. Basal metabolic rate is reduced in thyroid- and cortisone-substituted hypophysectomized diabetics (1, 11) due to abolition of growth hormone secretion. Cardiac output is decreased (12) probably secondary to a decreased venous return. Arterial blood pressure in the supine position remains unchanged (1, 3, 12). Blood volume is reduced (13). Vasoconstriction is known to take place following hypophysectomy. Falkheden (12) found an average increase in total vascular resistance of 30% in a group of hypophysectomized patients. Renal blood flow is reduced (14, 15), and we have recently found a considerable decrease in hand blood flow.[2]

It seems likely that venous return and cardiac output in the hypophysectomized diabetics are reduced to such a degree that an unchanged blood pressure can be maintained only by a considerable increase in the activity of the sympathetic nervous system. The increased levels of PCA may also be due to other regulatory mechanisms than that of blood pressure. The decreased flow in the hand of hypophysectomized diabetics is probably necessary for thermoregulation and likely to be mediately via the sympathetic nervous system.

Hypophysectomy reduces the progression of diabetic retinopathy and visual impairment (1, 2). Skin capillary resistance is considerably increased in such patients (3, 4), and the abnormal leakage of dye from the retina vessels decreases after the operation (16). Capillary resistance and capillary permeability are influenced by the level of the blood flow (17–19), and it is therefore possible that the increased capillary resistance and the decrease in permability observed after hypophysectomy are functional phenomena caused by a pronounced vasoconstriction, which in part is mediated via the sympathetic nervous system. Thus, an increase in sympathetic tone in the central retinal artery, which has a very pronounced sympathetic innervation (20), reducing retinal blood flow and intravascular pressure could be one of the factors responsible for the effect of hypophysectomy on diabetic retinopathy.

ACKNOWLEDGMENTS

Miss K. Carlsen is thanked for skillful technical assistance. Cand. med. M. S. Christensen is thanked for her preparation of the enzyme catechol-*o*-methyltransferase.

REFERENCES

1. Lundbæk, K., R. Malmros, H. C. Andersen, J. H. Rasmussen, E. Bruntse, P. H. Madsen, and V. A. Jensen. 1969. *In* Symposium on the Treatment of Diabetic Retinopathy. M. F. Goldberg and S. L. Fine, editors. Public Health Service Publication No. 1890. Washington, D. C. 291.

[2] Unpublished observation.

2. Oakley, N. W., G. F. Joplin, E. M. Kohner, and T. R. Fraser. 1969. *In* Symposium on the Treatment of Diabetic Retinopathy. M. F. Goldberg and S. L. Fine, editors. Public Health Service Publication No. 1890. Washington, D. C. 317.
3. Christensen, N. J. 1968. Increased skin capillary resistance after hypophysectomy in long-term diabetics. *Lancet.* **2**: 1270.
4. Christensen, N. J., and A. B. Terkildsen. 1971. Quantitative measurements of skin capillary resistance in hypophysectomized long-term diabetics. *Diabetes.* **20**: 297.
5. Engelman, K., and B. Portnoy. 1970. A sensitive double-isotope derivative assay for norepinephrine and epinephrine. *Circ. Res.* **26**: 53.
6. Christensen, N. J. 1967. Notes on the glucose oxidase method. *Scand. J. Clin. Lab. Invest.* **19**: 379.
7. Steiness, I. 1963. Diabetic neuropathy. Vibration sense and abnormal tendon reflexes in diabetics. *Acta Med. Scand. Suppl.* **173**: 394.
8. Christensen, N. J. 1968. Muscle blood flow, measured by xenon^{133} and vascular calcifications in diabetics. *Acta Med. Scand.* **183**: 449.
9. Christensen, N. J. 1968. Spontaneous variations in resting blood flow, postischaemic peak flow and vibratory perception in the feet of diabetics. *Diabetologia.* **5**: 171.
10. Landsberg, L., and J. Axelrod. 1968. Influence of pituitary, thyroid, and adrenal hormones on norepinephrine turnover and metabolism in the rat heart. *Circ. Res.* **22**: 559.
11. Falkeheden, T., T. Norin, B. Sjögren, and B. Skanse. 1962. Thyroid function and basal metabolic rate following hypophysectomy in man. *Acta Endocrinol.* **41**: 457.
12. Bojs, G., T. Falkheden, B. Sjögren, and E. Varnauskas. 1962. Haemodynamic studies in man before and after hypophysectomy. *Acta Endocrinol.* **39**: 308.
13. Falkheden, T., B. Sjögren, and H. Westling. 1963. Studies on the blood volume following hypophysectomy in man. *Acta Endocrinol.* **42**: 552.
14. Falkheden, T. 1963. Renal function following hypophysectomy in man. *Acta Endocrinol.* **42**: 571.
15. Isaacs, M., A. G. Pazianos, E. Greenberg, and B. J. Koven. 1969. Renal function after pituitary ablation for diabetic retinopathy. *J. Amer. Med. Ass.* **207**: 2406.
16. Balodimos, M. C., S. B. Rees, L. M. Aiello, R. F. Bradley, and A. Marble. 1969. *In* Symposium on the Treatment of Diabetic Retinopathy. M. F. Goldberg and S. L. Fine, editors. Public Health Service Publication No. 1890. Washington, D. C. 153.
17. Rossman, P. L. 1940. Capillary resistance in artificially induced fever. *Ann. Intern. Med.* **14**: 281.
18. Thomson, J. A. 1964. Alterations in capillary fragility in thyroid disease. *Clin. Sci. (London).* **26**: 55.
19. Mellander, S., and B. Johansson. 1968. Control of resistance, exchange, and capacitance functions in the peripheral circulation. *Pharmacol. Rev.* **20**: 117.
20. Ehinger, B. 1966. Adrenergic nerves to the eye and to related structures in man and the cynomolgus monkey (Macaca irus). *Invest. Ophthalmol.* **5**: 42.