performed, the weight in grams of the 10 control mice was 39.8 ± 1.4 SEM The GTG-treated mice averaged 60.7 ± 2.0 g. The difference in weight was highly significant ($P < 0.001$). All of the GTG-treated mice differed from the control mean weight by more than 2 standard deviations (which was our criterion for ensuring that hypothalamic damage had resulted from the GTG).

2-Deoxy-D-glucose (500 mg/kg, s.c.) or saline was injected at noon into 5 mice per group. 2 h later, the mice were killed by decapitation. Blood was collected into heparinized tubes; plasma was prepared and analyzed for glucose by a glucose oxidase method. The livers were rapidly removed and frozen on dry ice. Later they were homogenized, and the $100{,}000 \times g$ supernatant fraction was used for enzyme assays[20,21].

The Figure a shows plasma glucose levels. The GTG-lesioned mice in this experiment were hyperglycemic. The percentage increase in blood glucose concentration caused by 2-deoxyglucose was about equal in control and in GTG-treated mice. Basal activity of hepatic tyrosine aminotransferase (Figure b) in GTG-lesioned mice was approximately equal to that in intact mice. 2-Deoxyglucose significantly increased the enzyme in both groups, although the response in the GTG-lesioned mice was only about half that in the controls. YUWILER et al.[22] have shown that glucose feeding antagonized the induction of tyrosine aminotransferase by glucocorticoids, tryptophan, and casein. Possibly the high blood glucose level in the GTG mice treated with 2-deoxyglucose suppressed the induction of tyrosine aminotransferase. But YUWILER et al. also found that glucose antagonized tryptophan oxygenase induction, and tryptophan oxygenase induction by 2-deoxyglucose was as great in the GTG mice as in controls (Figure c). Indeed, hepatic tryptophan oxygenase activity was almost identical in GTG-treated and in control mice both before and after treatment with 2-deoxyglucose. These experiments show that 2-deoxyglucose induces hepatic tyrosine aminotransferase and tryptophan oxygenase both in intact mice and in mice with GTG-induced hypothalamic lesions. An intact ventromedial hypothalamus is thus not required for these effects of 2-deoxyglucose.

The mechanism by which 2-deoxyglucose induces these enzymes is not established. We have earlier shown that tyrosine aminotransferase is induced by epinephrine[23], so that stimulation of epinephrine secretion from the adrenal medulla is a possible mechanism. That stimulation of epinephrine secretion in fact occurred after 2-deoxyglucose administration to both groups of mice is suggested by the similar elevation in blood glucose levels. Plasma free fatty acid levels measured at the time of sacrifice of the mice in our study were not elevated in the 2-deoxyglucose-treated group, but that is no doubt because the time interval of 2 h after drug treatment was too long; RICHARDSON and HOKFELT[24] found maximum increase in plasma FFA $^1/_2$ h after 2-deoxyglucose treatment with little effect remaining at 2 h.

In summary, 2-deoxyglucose increased hepatic tyrosine aminotransferase and tryptophan oxygenase activity in intact mice and in mice with hypothalamic lesions induced by GTG treatment. The induction of these enzymes may have been mediated by stimulation of adrenal medullary secretion.

Zusammenfassung. 2-Deoxyglukose führt bei Mäusen zu Blutzuckersteigerung und in der Leber zu Aktivierung der Tryptophan-Pyrrolase und Tyrosine-Transaminase bei durch Goldthioglukose verursachten hypothalamischen Schäden.

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Early and Selective Increase in Brain Dopamine Levels after Axotomy

The dopamine (DA) in the neostriatum (the caudate nucleus plus putamen) disappears within a week after making lesions in the cell bodies or in the non-terminal axons belonging to the nigro-neostriatal DA neurons[1-3]. This is probably due to degeneration and death of the DA-containing axon terminals of these neurons. Similarly, ascending noradrenaline (NA) neurons have been demonstrated and mapped out[2,4]. In this investigation, the time courses of the changes in the DA and NA concentrations of the forebrain have been studied both biochemically and histochemically in more detail, and have been compared in the same animals, after lesions were made in the ascending DA and NA pathways.

Material and methods. In the biochemical experiments, hooded rats of both sexes weighing about 200 g were used. Under pentobarbital sodium anaesthesia (about 40 mg/kg i.p.), a complete and almost frontal hemisection of the forebrain through the caudal hypothalamus was made by means of a blunt-edged spatula[5]. In the control rats, only the skull and dura were opened. At different time intervals after the operation, the rats were decapitated and the lesion was extended to the other side. The DA and NA on both sides of the forebrain frontal to the lesion were determined spectrophotofluorimetrically after cation exchange chromatography and oxidation[6-8].

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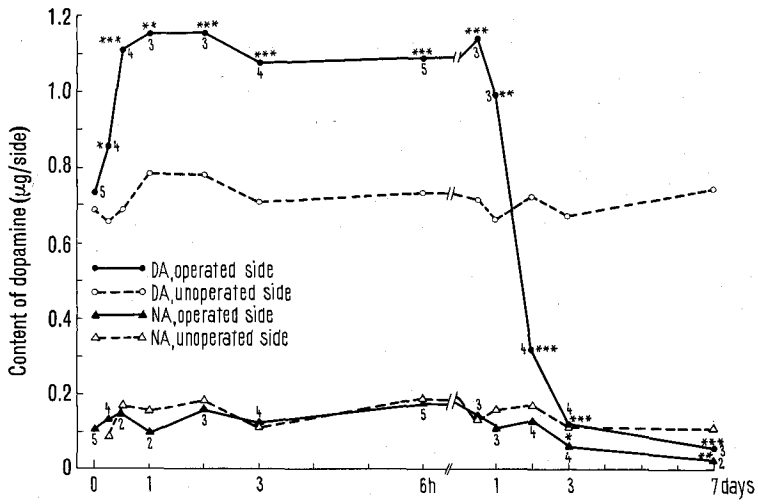

Contents (µg/side) of dopamine (DA) and noradrenaline (NA) on the 2 sides of the rat forebrain at various time intervals after a complete hemisection of the brain. Means and number of experiments are indicated. Differences between the operated and unoperated side are indicated by ***, ** and * for $P < 0.001$, $P < 0.01$ and $P < 0.05$, respectively. Statistical evaluation by t-test after single-factor analysis of variance (variances within groups 0.01779 and 0.001437 with 66 and 60 degrees of freedom within groups for DA and NA, respectively).

In the histochemical experiments, Sprague-Dawley rats weighing 150 g were operated on unilaterally under N_2O–O_2-halothane anaesthesia[4]. The electrothermic lesion was about 1 mm in diameter and was placed stereotaxically at the level of the mamillary body. It involved the lateral part of the lateral hypothalamic area, the ventral part of the crus cerebri and the medial parts of the subthalamus. At different time intervals after the operation, the brains were taken for histochemical analysis of monoamines which involved freeze-drying, formaldehyde treatment and embedding in paraffin[9,10]. In order to observe increases in the DA fluorescence more easily, relatively dry reaction conditions were used.

Results and discussion. Using biochemical techniques, a marked and statistically highly significant rise in DA was observed on the lesion side (Figure). The increase was already evident 15 min after the operation and the DA content seemed to remain at about 150% of the unoperated side for about 24 h. Thereafter, the DA concentration fell sharply to very low values after 7 days. Previous reports have indicated that a lesion of the nigro-neostriatal DA pathway may cause a transient rise in the striatal DA before its disappearance[11–13]. In contrast to the DA, the amounts of NA remained virtually unchanged until the third day after the operation when a fall then occurred. No early clearcut changes were observed for 5-hydroxytryptamine according to our preliminary data. The losses of DA and NA can be accounted for by anterograde degeneration of the ascending neurons.

In the histochemical experiments, similar increases were observed for the DA fluorescence in the nerve terminals of the caudate nucleus-putamen (Table). Less marked increases were seen in the DA terminals of the olfactory tubercle and nucleus accumbens. Both the nigro-neostriatal and the meso-limbic DA neurons were severed by the lesion, as evidenced by the vast accumulation of catecholamines in the DA bundles caudal to the lesion and by retrograde changes in the mesencephalic DA cell bodies. Since a similar accumulation was observed in the ventral NA bundle on the cell body side of the lesion, this pathway was, in all probability, also affected. However, the large NA nerve terminals of this system in the hypothalamus, preoptic area and basal forebrain did not display any increase corresponding to that of the DA terminals.

In conclusion, a lesion of the ascending catecholamine neurons in the brain produces an increase in the DA but not in the NA concentration of the corresponding nerve terminals before the disappearance associated with nerve degeneration occurs. This rise must be due to an imbalance between formation and loss of DA but the exact mechanism needs further investigations[14].

The fluorescence intensity in the neostriatal dopamine nerve terminals of rat at different time intervals after acute unilateral electrothermic lesion

Hours after operation	Fluorescence intensity[a] Lesion side				Control side	
0	2	(7)			2	(7)
1/4	2 1/2	(4)	3	(2)	2	(6)
1/2	2 1/2	(2)	3	(2)	2	(4)
1	3	(2)	4	(4)	2	(5)
3	3	(1)	4	(12)	2	(13)
6	4	(7)			2	(7)
18	4	(5)			2	(5)
24	2	(2)	3	(5)	2	(7)
36	0	(3)	1	(5)	2	(8)
48	0	(5)			2	(5)

[a] A semiquantitative estimation of fluorescence intensity was made on coded slides: 4 = very strong, 3 = strong, 2 = moderate, 1 = weak, 0 = no fluorescence. Number of rats within parentheses.

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Résumé. Nous avons étudié les concentrations de dopamine et de noradrénaline du prosencéphale après lésion unilatérale des neurones monoaminergiques correspondants. Les deux méthodes employées soit biochimique et histochimique ont mis en évidence sensiblement le même phénomène, à savoir: une augmentation importante et rapide du taux de dopamine entre 15 min et 24 h suivie d'une chute due selon toutes probabilités à la dégénerescence axonale. Quand à la noradrénaline, seule la deuxième phase, la chute, a pu être observée.

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Inhibition of Acute Inflammation by Hyperlipemia

Dextran-induced anaphylactoid inflammation is inhibited by diabetes[1]. It has been suggested that this is due to the elevation of the blood sugar level[2]. Hyperlipemia is also very characteristic of diabetes. Therefore, experiments were carried out to determine the influence of the artificially induced hyperlipemia on the inflammatory response evoked by dextran.

Methods. Female rats of R Amsterdam strain weighing 100–200 g were used. Inflammatory response was produced by subplantar injections of dextran (300 µg Intradex in 0.1 ml) and carrageenin (500 µg in 0.1 ml). The volumes of the hind paws were measured by a mercury displacement method before and 60 min after dextran and 4 h after carrageenin injection. The degree of inhibition was expressed as percent of the control value.

Hyperlipemia was induced by oral administration of sunflower seed oil (oleum helianthi) and oleic acid by a stomach tube and by i.v. injection of Lipofundin® (B. Braun, Melsungen, 10% soybean oil emulsion (and oleic acid)/3% emulsion prepared by Tween 20 in isotonic saline). Inflammation was induced 2 h after oral and 30 min following i.v. administration. Serum samples were withdrawn at the time when inflammation was provoked. Lipids were extracted[3] and evaporation was done under N_2 atmosphere[4]. Triglyceride and free fatty acids (FFA) were determined by thin layer chromatography[5]. Blood sugar was measured by orthotoluidine reagent[6].

Results and discussion. The experiments showed that oleum helianthi (5.0 g/kg by mouth) and Lipofundin® (0.5–1.0 g/kg i.v.) considerably inhibited the anaphylactoid inflammation (Table I). The effect depended upon the dose applied and the body weight of the animals. The inhibition was only moderate in rats weighing 100 or 150 g, whereas animals weighing 200 g responded more intensively to the pretreatment. This fact emphasizes the significance of age in the inhibitory response induced by fat load.

Oleic acid in a dose of 150 mg/kg i.v. was ineffective in blocking dextran edema. Oleic acid and glycerol (5.0 g/kg) given by mouth had no significant effect on the inflammatory process evoked by dextran (Table I).

Lipid determinations indicated that 1.0 g/kg of oleum helianthi which had no effect on the inflammation did not

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Table I. Effect of oleum helianthi, Lipofundin®, oleic acid and glycerol on the acute inflammation induced by dextran

Pretreatment	Dose (g/kg)	Route of administration	Body weight[a] (g)	Inhibition (%)	*P*
Oleum helianthi	1.0	per os	200 (6)	11	N.S.
	5.0		150 (6)	33	0.01
	5.0		200 (10)	51	0.001
Lipofundin®	0.25	i.v.	200 (6)	28	0.01
	0.5		200 (10)	55	0.001
	1.0		100 (6)	36	0.01
	1.0		150 (6)	36	0.01
	1.0		200 (10)	61	0.001
Protamine sulphate	0.005	i.v.	200 (10)	0	N.S.
Protamine sulphate	0.005	i.v.			
plus Lipofundin®	0.5		200 (10)	75	0.01[b]
Oleic acid	0.15	i.v.	200 (10)	0	N.S.
	5.0	per os	200 (10)	12	N.S.
Glycerol	5.0	per os	200 (6)	0	N.S.

[a] In parentheses number of experiments. [b] Compared to the group treated with only Lipofundin®. [c] Inflammation was evoked 1 h after the administration of oleic acid. N.S.: not significant.