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Neurocircuitry of the Basal Ganglia Studied by Monitoring Neurotransmitter Release

Effects of Intracerebral and Perinatal Asphyctic Lesions

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Abstract

The neurocircuitries of the basal ganglia are studied with in vivo microdialysis, with special consideration to dopamine transmission and its interaction with other neurotransmitter systems. The aim is to develop experimental models to study the pathophysiology and therapy of neurodegenerative disorders of the basal ganglia, as well as to develop models to study the short- and long-term consequences of perinatal asphyctic lesions. A main goal of these studies is to find and to characterize new treatments for these disorders.

Index Entries: Basal ganglia; neurotransmitter release; microdialysis; chemical lesions; asphyxia; hypothermia; rats.

Introduction

In this article, we present our recent investigations about the neurocircuitry of the basal ganglia. The aims of these investigations have been:

1. To study the neurocircuitry and neuropharmacology of the basal ganglia, with special consideration to dopamine (DA) transmission and its interaction with other neuronal systems;
2. To develop experimental models to study the pathophysiology of neurodegenerative disorders of the basal ganglia;
3. To study new pharmacological approaches for the treatment of these disorders; and
4. To develop a novel model to study the short- and long-term consequences of perinatal asphyctic lesions.

The basal ganglia provide a complex neuronal network that conveys and integrates signals from and to the cerebral cortex. Neurocircuitries of the basal ganglia have been associated with several neurodegenerative diseases. Thus, it is well established that in Parkinson's disease degeneration of the nigrostriatal DA system constitutes the most critical abnormality. In Huntington's chorea the most prominent feature is a marked loss of

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striatal neurons. However, although Alzheimer's disease and dementia of Alzheimer's type are primarily associated with deficits of cerebral cortex and hippocampal formation, some of the symptoms presented by these diseases are also linked to dysfunctions of the basal ganglia *(1)*. Furthermore, deficits of mesolimbic and mesocortical DA transmission have been associated with several functional syndromes, such as psychosis *(2)* and drug addiction *(3)*.

Whereas DA is an essential neurotransmitter of the basal ganglia, it exerts modulatory actions on neuronal circuitries that utilize amino acids and/or neuropeptides as chemical messengers. In the striatum, DA terminals directly link with medium-size spiny neurons utilizing γ-amino butyric acid (GABA) as a principal neurotransmitter, but also with neurons utilizing opioid or tachykinin peptides *(4)*. In the striatum, acetylcholine (ACh) is found in intrinsic large size neurons *(5)*. It has been suggested that striatal ACh neurons are not synaptically linked with DA terminals *(6)*. However, in an electron microscopy study, Kubota et al. *(7)* presented evidence that striatal cholinergic neurons may indeed receive direct inputs from dopaminergic axons. In agreement, we have recently shown, in an *in situ* hybridization study, that approx 95% of large size neurons in the striatum express mRNA for DA D-2 receptors *(8)*.

The medium-size spiny neurons project axons to the globus pallidus and to the substantia nigra. Neurons projecting to the globus pallidus contain both GABA and enkephalin, whereas striato-nigral fibers contain GABA, substance P, and dynorphin *(9)*. There are studies showing that in the striatum, DA exerts, via different receptors, selective actions on neuropeptides associated to different projection systems *(10)*. Dopamine acts on striato-pallidal neurons via D-2 type of receptors and on striato-nigral neurons via D-1 receptors. Thus, DA receptor multiplicity provides a gating system funneling striatal activity via different efferent pathways, a hypothesis largely developed in our own laboratory *(11–19)*.

The interaction between DA and the neuropeptide cholecystokinin (CCK) has been an important issue for describing the functioning of the basal ganglia. The issue has been discussed in rather oversimplified terms and without morphological background. In the striatum, several different CCK systems exist, with both intrinsic and extrinsic origin *(20)*. A proportion of mesencephalic DA neurons projecting to the telencephalon utilizes CCK as a cotransmitter *(21)*, and this neuropeptide is also present in cortico-striatal projections, probably colocalized with glutamate. We have recently reported evidence for a partly crossed CCK cortico-striatal pathway in the rat *(22–24)*. Little is, however, known about the functional interactions between CCK and DA and the contributions from each particular system to the final output of the basal ganglia.

Several methods have been applied to study interactions between monoamines and neuropeptides in the brain, the majority of them utilizing indirect neuroanatomical or behavioral techniques. We have chosen to utilize the novel technique, largely developed in our laboratory, of in vivo microdialysis *(25)*, which allows the simultaneous monitoring of the release of monoamines, ACh, amino acids, and peptides in restricted regions of the brain. Thus, the interactions among different nuclei of the basal ganglia have been studied with in vivo microdialysis, in normal and lesioned rats. DA, ACh, glutamate, aspartate, GABA, purines, lactate, and pyruvate are assayed with high-performance liquid chromatography (HPLC), coupled to electrochemical (EC), fluorescence (F), or ultraviolet (UV) detection systems. Neuropeptides are measured by sensitive radioimmunoassays (RIA) utilizing selective antibodies. Rotational behavior *(11,12,26)*, simultaneously with microdialysis or in parallel experiments has also been recorded. Histochemistry and *in situ* hybridization studies have also been performed.

The main aim of these studies is, however, to characterize pharmacological treatments. Drugs have been analyzed in experimental models mimicking Parkinson's or Alzheimer's diseases. Attention has been given to the effects of treatments with endogenous and exogenous trophic factors, such as nerve growth factor (NGF), the monosialoganglioside GM1, and nicotine.

Functional Neuroanatomical Studies

Modulation of Striatal DA Release by Striato-Nigral Pathways

Striatal DA release is differently modulated by striato-nigral GABA, dynorphin, substance P, and neurokinin A pathways. GABA and dynorphin exert a negative feedback on striatal DA, whereas substance P and neurokinin A provide a positive

feedback (27–29). The effects produced by substance P and neurokinin A are conveyed via different receptors (30,31), and via different neuronal (32,33) and metabolic (34,35) pathways (36). We have found that exogenously administered substance P may be cleaved to a shorter active fragment (substance P [1–7]), which can then have antagonistic properties against substance P (34), in contrast to the C-terminal substance P fragment (substance P [6–11]), which does not exert any significant modulation on the effects of substance P (35).

In order to monitor closely the neurotransmitter cascade connecting the substantia nigra and the striatum, the sensitivity of the analytical assays had to be in the lower pico- and femtomole levels for monoamines and amino acids, and peptides, respectively. Thus, following implantation of microdialysis probes into the striatum and into the substantia nigra, we can now simultaneously monitor monoamines, amino acids, and neuropeptides in both regions. Figure 1 shows dynorphin B and GABA levels simultaneously monitored in left striatum and left substantia nigra under basal and K^+-depolarizing conditions. The GABA antagonist biccuculine has been included together with the KCl, in order to block the effect produced by a massive release of GABA.

The effects of striatal or intranigral administration of the dopamine D-1 agonist SKF 38393, and the D-2 agonist quinpirole on GABA and dynorphin release have been studied. We have found that 100 μM of SKF 38393 included in the striatal perfusion medium produces a greater than twofold increase in nigral GABA and dynorphin levels, whereas no such effect has been observed after quinpirole. In contrast, nigral quinpirole, but not SKF-38393, produces a concentration-dependent decrease in striatal dopamine levels. In the nigra, SKF-38393 induces an increase of nigral GABA and dynorphin levels (85).

Modulation of Striatal DA Release by Cortico-Striatal Pathways

There is evidence that striatal DA release is presynaptically modulated by glutamatergic cortical inputs. It was proposed that the DA stimulation produced by glutamate reflected direct axonal interactions between glutamatergic and dopaminergic terminals in the striatum. This hypothesis received some support from biochemical and histochemical studies showing direct intrastriatal axonal interactions. However, the majority of the striatal afferents from the cortex and substantia nigra make axodendritic synaptic contacts with striatal neurons (37), giving a basis for polysynaptic loops, including GABA and/or ACh neurons, by which cortical glutamate neurons can also modulate striatal DA release (38,39).

We have found that cortical stimulation produces an increase in striatal ACh and DA release (39). These effects, however, seem to be mediated by different glutamate receptors; kainate agonists produced an increase in striatal DA release, whereas NMDA agonists produced an increase in striatal ACh release (38,40). The development of this model will enable us to follow the changes in extracellular striatal monoamine, amino acid, and neuropeptide levels during cortical stimulations. We have found, however, that the study of the interactions between cortex and striatum is complicated by the fact that inputs onto the striatum are partly originating in the contralateral side. Thus, we have reported that glutamate and CCK are released from a partly crossed cortico-striatal pathway (41,22–24).

Evidence for the presence of DA nerve terminals in the deep layers of the fronto-parietal cortex of the rat has been presented (42). In this cortical region, extracellular DA is found in a 1-nM range and can be increased by K^+-depolarization or amphetamine stimulation, and suppressed by mesencephalic 6OHDA lesions (42,43). Nigral administration of substance P produces an increase in both cortical and striatal DA release, whereas neurokinin A stimulates striatal DA release only (43). Differences in striatal and cortical DA functions have also been found in studies measuring the mRNA expression of several putative neurotransmitters with *in situ* hybridization and RNA blots. We have found that glutamic acid decarboxylase (GAD, a marker for GABA neurons), somatostatin, and NPY mRNA gene expression were increased in the striatum, but decreased in the cortex following DA deafferentation (44,45), suggesting therefore that DA has different functional roles in the striatum and frontoparietal cortex.

Extracellular levels of ACh can be simultaneously measured in the cortex and striatum of rats (46). These levels could be selectively stimulated by several pharmacological treatments and inhibited by specific lesions. A unilateral ibotenic acid lesion into the nucleus basalis, but not into the striatum, produced a strong decrease in extracellular ACh levels in the ipsilateral cortex (47). Unilateral decortication with the excitotoxin kainic acid, which selectively damages local neurons while sparing

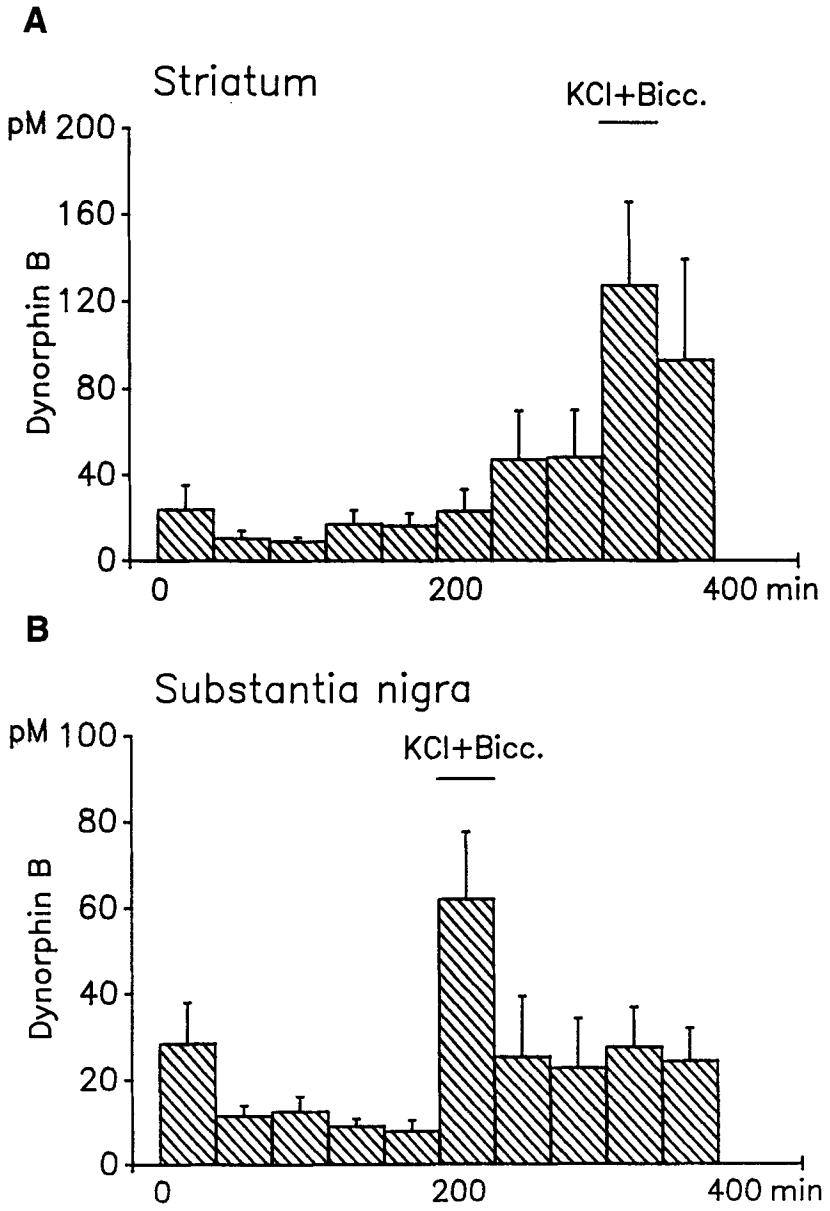

Fig. 1. Extracellular dynorphin B **(A, B)** and GABA **(C, D)** levels (means ± SEM) simultaneously monitored in striatum (A, C) and substantia nigra (B, D) of "normal" rats ($n = 6$) by in vivo microdialysis. A 4- and a 2-mm microdialysis probe (CMA 12; CMA/Microdialysis AB, Stockholm, Sweden) was simultaneously implanted into the left striatum and the left substantia nigra, respectively, in halothane-anesthetized rats. Microdialysis probes were perfused with a modified CSF solution.

afferents and fibers of passage, also produced a decrease in cortical ACh levels. This decrease was lower than that produced after a nucleus basalis lesion, in agreement with neuroanatomical evidence showing that, in the cortex, ACh mainly constitutes an extrinsic system *(48)*. Furthermore, we found that nucleus basalis lesions affected cortical and striatal DA levels as well, probably reflecting indirect functional interactions *(47)*.

Experimental Models for Neurodegenerative Diseases

The 6-Hydroxy-Dopamine (60HDA) Model

The 60HDA model, developed by Urban Ungerstedt at the Karolinska Institute, Stockholm *(26)*,

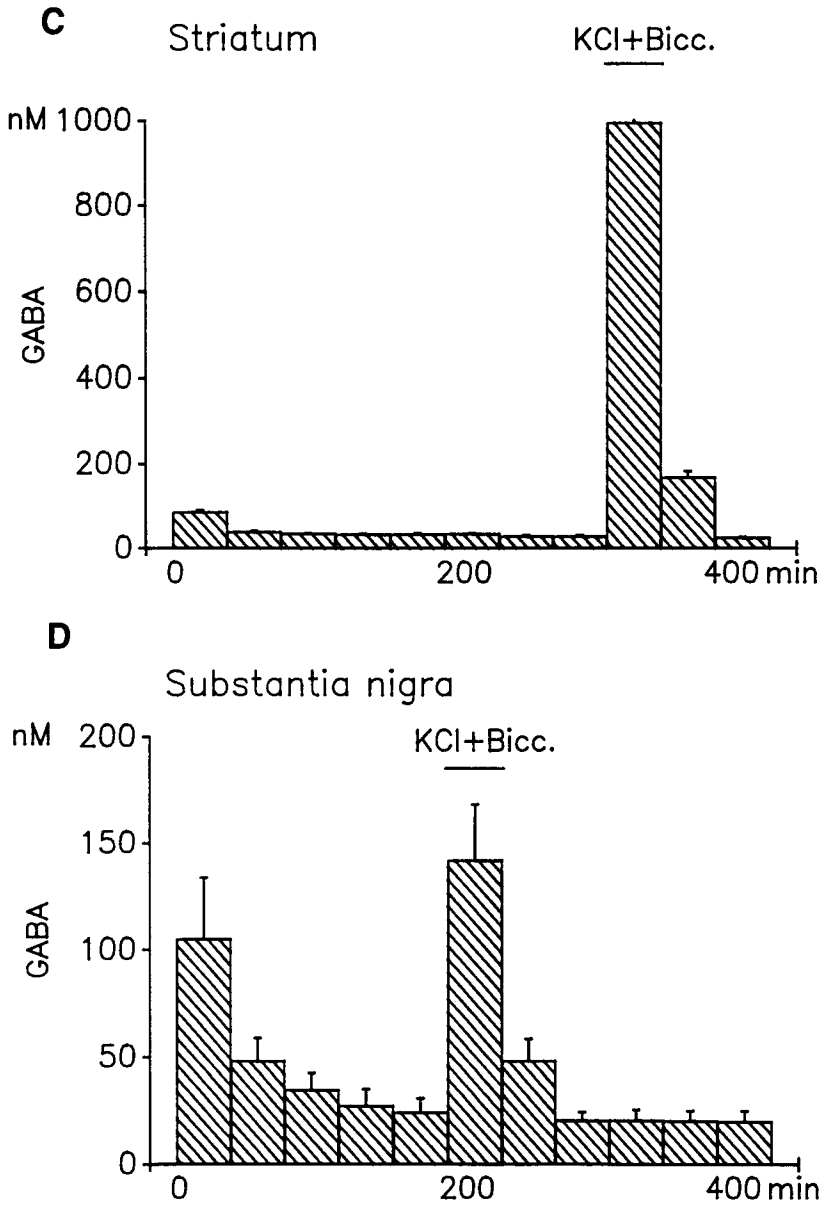

Fig. 1. *(continued)* At the 200–240-min period following microdialysis implantation, 100 m*M* KCl plus 100 μ*M* biccuculine were included in the perfusion medium of the left substantia nigra. At the 320–360-min period, KCl plus biccuculine were included in the ipsilateral striatum. GABA levels were detected by HPLC-EC. Dynorphin B levels were measured in the same perfusion samples by using a highly sensitive RIA method.

constitutes an experimental prototype for studying Parkinson's disease. 60HDA is unilaterally injected into the medial forebrain bundle (MFB) in order to produce an extensive DA deafferentation of the ipsilateral telencephalon and therefore a biased behavior *(11,12)*. This model allows the analysis of new therapeutic strategies, e.g., treatments with selective DA agonists and with trophic factors *(1,12,49–56; see also* ref. *15)*.

Unilateral Lesion of the Basal Ganglia by Transections at the Mesodiencephalic Junction

This model, developed by Agnati et al. *(57)*, can be used to study the effects of partial lesions on DA pathways and that of treatments with endogenous and exogenous neurotrophic factors. We have found in this model that chronic treatment with

nicotine administered by subcutaneously implanted Alzet minipumps counteracts the decrease in extracellular neostriatal DA induced by the transection, supporting the idea that chronic nicotine may protect against degeneration of central DA neurons *(58,59)*.

Models for Dementia of Alzheimer Type

We have proposed two models to study dementia of Alzheimer type, one based on cortical infarcts induced by devascularizing lesions *(46,60,61; see also* ref. *1)* producing retrograde degeneration of neurons in the nucleus basalis, and another where lesions are directly performed into the nucleus basalis *(47)*. The effects of NGF and GM1 treatments on neural repairing have been studied in these models. We have found that chronic administration of GM1 or NGF could reverse biochemical and morphological changes induced by these lesions *(46,60,61)*.

In vivo microdialysis has been used to study the effects of the aziridinium ion of ethylcholine mustard (AF64A), a powerful alkylating agent that binds to high-affinity choline uptake sites, on intrinsic and extrinsic neuronal systems of rat neostriatum. This neurotoxin has been proposed as a selective toxin for cholinergic neurons, and thus, perhaps, a useful tool in the development of animal models of Alzheimer's disease and senile dementia of the Alzheimer type *(62)*. We compared the effect of AF64A to that of ibotenic acid on striatal ACh, GABA, DA, glutamate, and aspartate levels *(63)*. We concluded that, although AF64A is a potent neurotoxin for intrinsic neuronal systems, it appears, like ibotenic acid, to have similar effects on local cholinergic and GABAergic neuronal systems in the striatum.

Pharmacological Treatments

Mechanisms of Actions of Antiparkinsonian Drugs

Most of the antiparkinsonian drugs are DA agonists. Therefore, we have studied the selectivity of several DA agonists on different receptor populations and proposed the idea that receptor multiplicity may constitute a mechanism by which the actions of DA are gated by different neuronal pathways *(15)*. Studies with rotational behavior *(11–14)*, microdialysis *(14–19,40,64)*, and recently, studies with *in situ* hybridization histochemistry combined with fluorogold tracing technique *(10)* support this hypothesis *(65,66)*. We have found that, in the striatum, DA exerts an inhibitory modulation on GABA neurons via D-2 receptors, whereas D-1 stimulation exerts a stimulatory modulation on GABA neurons *(19)*.

Looking for new antiparkinsonian therapies, we have found that caffeine shares some of the properties of DA agonists. Thus, the methylxanthines, caffeine, theophylline, and theobromide produce rotational behavior in 60HDA lesioned rats, a behavior partially inhibited by DA antagonists *(67,68)*. Although we have studied several hypotheses, we still lack conclusive results to support a single mechanism for explaining the effects produced by methylxanthines in 60HDA lesioned rats *(67–73)*.

Treatment with Endogenous and Exogenous Trophic Factors

The idea that degenerative processes may be delayed or even reversed by exogenously administered trophic factors, such as NGF and GM1, is now accepted. NGF is synthesized within target tissues of some peripheral and central neurons and can act on specific receptors *(74)*. NGF, in turn, is internalized and retrogradely transported to cell bodies *(75)*. Thus, NGF can be used as a pharmacological tool to induce nerve growth and repair.

At the beginning of the 1980s, we studied the promotion of phenotypical transformation of chromaffin cell grafts by NGF. We were the first to report in vivo studies *(50,76,77)* showing that NGF could induce changes in chromaffin cells grafted into a DA deafferented striatum, transforming their endocrine-like into a neuron-like feature. It was found that the changes were associated with the reversing of symptoms of experimentally induced parkinsonism. We also showed the effects of NGF treatments on extracellular ACh, DA, and adenosine levels in the cortex and striatum of rats with unilateral devascularizing cortical lesions *(61)*.

Another neurotrophic factor, the monosialoganglioside GM1, has also been tested for the promotion of nerve growth and repair *(78)*. GM1 can prevent retrograde changes in nucleus basalis produced by cortical lesions *(79)* and might also stimulate the activity of cortical cholineacetyltransferase in regions adjacent to the lesions. We have extended these studies by analyzing the effects of decortication and treatments with GM1 on cortical and striatal ACh, catecholamines, and adenosine lev-

els measured with microdialysis (*46*). A novel administration route for neurotrophic factors, i.e., microencapsulation into human serum albumin microspheres, which can then be topically applied onto damage regions, has also been studied (*60*). Such treatments with GM1 promote (1) recovery of retrograde morphological changes produced by devascularization and (2) a parallel increase in cortical ACh release. Although these results appear to be promising, the possibility that excessive trophic stimulation might lead to aberrant connections in addition to, or instead of functionally reparative ones has to be carefully investigated.

Perinatal Asphyctic Lesions

The Experimental Model

We have developed a noninvasive animal model for studying the short- and long-term consequences of hypoxic-ischemic lesions in rats, similar to those produced under labor in clinical situations (*80*). We found that perinatal asphyxia for a period longer than 22 min, in a water bath at 37°C, led to 100% mortality within the first 20-min period following delivery. However, when the uterus containing the pups was kept 22 min in a 30°C water bath, 100% of the pups recovered respiratory function following tactile oral stimulation and were accepted by the surrogate mothers (*81*, Loidl et al., in preparation). The protective effect of hypothermia at 30°C even allows for a 47–48-min asphyctic period. When asphyxia was induced in a water bath at 15°C, 100% survival could be extended to 101 min.

Short-Term Effects of Perinatal Asphyxia

Several parameters are acutely or chronically recorded by direct observation or by in vivo microdialysis. Following asphyxia, pups are subcutaneously implanted with 4-mm microdialysis probes in the dorsal region, while kept on a heating pad (Fig. 2). Thus, subcutaneous levels of amino acids (glutamate, aspartate), and metabolism products (lactate, pyruvate, and ascorbate) are monitored during the 40–60- and 60–80-min periods after removal from the uterus in asphyctic and controls pups.

Effects of Delivering by Hysterectomy

Pups delivered from uterus horns removed by hysterectomy from rats at the final day of gestation started regular breathing (respiratory frequency 60/min) almost immediately after the delivering was completed. These control pups showed a pink-colored skin and intensive vocalization and motility. They were accepted by surrogate mothers after an 80-min observation period. When the pups were accepted, they grew up in a similar manner to that of normally delivered rats (at least during a 1-mo period of observation). Subcutaneous glutamate levels were ≈2 μ*M*; aspartate ≈0.4 μ*M*; lactate ≈1 m*M*; and pyruvate ≈60 μ*M*.

Short Asphyctic Exposure

Following a 5–6-min asphyctic period, induced in a water bath at 37 or 30°C, all the pups started breathing shortly after delivery. Their behavior was similar to that observed in the control animals. Following 5–6 min of asphyxia at 37°C, glutamate levels were ≈10 μ*M*; aspartate ≈1.2 μ*M*; lactate ≈2 m*M*; and pyruvate ≈60 μ*M*. At 30°C glutamate levels were ≈6 μ*M*; aspartate ≈2 μ*M*; lactate ≈1 m*M*; and pyruvate ≈40 μ*M*. Following 15–16 min asphyctic periods, induced in a water bath at 37 or 30°C, all the pups survived, without differences in color of the skin and respiratory frequency, as compared to the control animals. A slight decrease in spontaneous motility was observed, but all the pups were, however, accepted by the surrogate mothers. At 37°C, glutamate levels were ≈7 μ*M*; aspartate ≈1 μ*M*; lactate ≈2 m*M*; and pyruvate ≈60 μ*M*. At 30°C glutamate levels were ≈3 μ*M*; aspartate ≈0.5 μ*M*; lactate ≈2 m*M*; and pyruvate ≈60 μ*M*.

Intermediate Asphyctic Exposure

Following a 19–20-min asphyctic period at 37°C, the pups had to be intensively stimulated to start breathing. The surviving pups remained akinetic for a long period after delivery, showed a significant decrease in respiratory frequency (≈20/min), which was accompanied by gasping and showed a pink/pale skin coloration. Approx 30% of the pups died shortly after delivery. In contrast, all the pups survived following a 19–20-min asphyctic period at 30°C. Initial gasping, a slight decrease in respiratory frequency (≈40/min), and in motility could be observed. The color of the skin was similar to that in control pups. All the surviving pups were accepted by the surrogate mothers. At 37°C, glutamate levels were ≈5 μ*M*; aspartate ≈0.5 μ*M*; lactate ≈2 m*M*; and pyruvate ≈60 μ*M*. At 30°C glutamate levels were ≈6 μ*M*; aspartate ≈1 μ*M*; lactate ≈2 m*M*; and pyruvate ≈40 μ*M*.

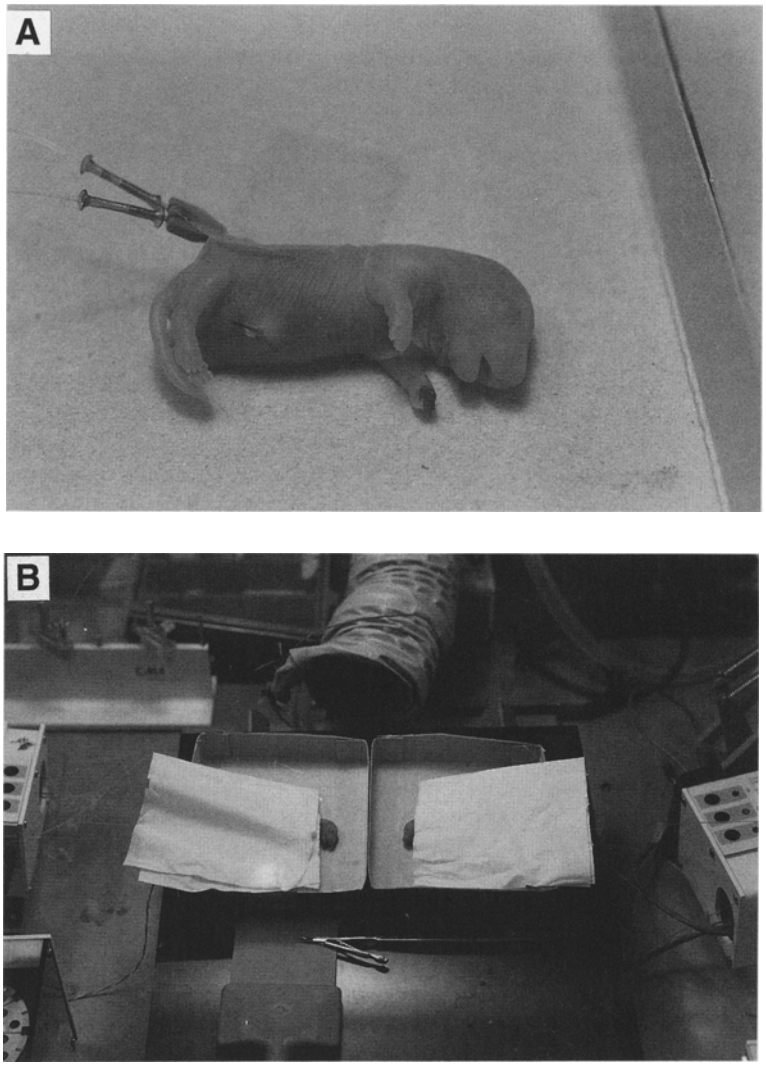

Fig. 2. **(A)** Pup recovering from a 19–20-min asphyctic period, at 37°C. A 4-mm microdialysis probe was subcutaneously implanted into the dorsal region in order to monitor peripheral glutamate, aspartate, lactate, and pyruvate in 20-min (40 μL) perfusion samples. Two fraction samples were taken before the pup was given to surrogate mothers. The experimental conditions under which the pups are normally microdialysed are shown in **(B)**.

Long Asphyctic Exposure

The rate of survival rapidly decreased following prolonged asphyctic periods at 37°C (>20 min), and as a whole, the physiological condition of the surviving pups deteriorated (increased gasping, decreased respiratory frequency, lack of vocalization, akinesia, and pale skin). No pups survived following asphyctic periods longer than 22 min. In contrast, at 30°C, all the pups survived up to a 30–31-min asphyctic period, although some signs of physiological impairment (presence of gasping, decrease in respiratory frequency and motility, and pale skin) could be observed. At this temperature, 40% survival could be observed following a 47–48-min asphyctic period. All the surviving pups showed gasping, a decrease in respiratory frequency (≈10/min), akinesia, and pale skin. No survival was observed following asphyctic periods longer than 48 min. Following a 21–22-min asphyctic period at 37°C, glutamate levels were ≈4 μ*M*; aspartate ≈0.4 μ*M*; lactate ≈2 m*M*; and pyruvate ≈90 μ*M*. At 30°C glutamate levels were ≈9 μ*M*; aspartate ≈1 μ*M*; lactate ≈2 m*M*; and pyruvate ≈40 μ*M*.

At 15°C, 100% survival was observed up to 101 min of asphyxia. Following a 50–51-min asphyctic

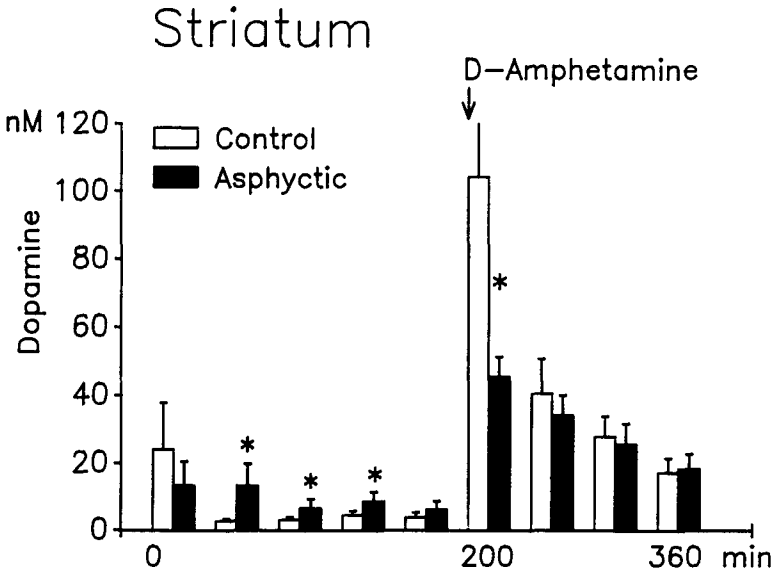

Fig. 3. Effect of perinatal 19–20-min asphyxia on extracellular striatal dopamine levels measured in samples collected by in vivo microdialysis 1 mo after delivery. A 4-mm microdialysis probe was implanted into the left striatum of halothane-anesthetized asphyctic ($n = 7$) and control ($n = 6$) rats. Microdialysis probes were perfused with a modified CSF solution. At the 200-min period following microdialysis implantation, a dose of D-amphetamine (2 mg/kg sc) was administered. Dopamine was detected in 20 µL samples using a highly sensitive HPLC-EC. Vertical lines show SEM. * = $p < 0.05$ for the two-tailed test (Mann-Whitney U-test, corrected by the Bonferroni's procedure).

period, gasping was observed in 50% of the pups, the respiratory frequency was ≈40/min, vocalization and motility were decreased, and the skin was pink/pale. Following a 100–101-min asphyctic period at 15°C, gasping was observed in all the pups, and the respiratory frequency was 10/min. The pups were akinetic and pale, and no vocalization was observed.

Long-Term Effects of Perinatal Asphyxia

We have completed a series of experiments in which the pups were exposed to asphyxia and, approx 1–6 mo later, implanted with two microdialysis probes, one into the striatum and another one into the substantia nigra. Monoamines and amino acids were monitored under basal and D-amphetamine-stimulated conditions.

Significant changes in monoamines and GABA were observed in rats exposed to subsevere and severe asphyxia, but nonsignificant effects were observed in glutamate and aspartate levels. Striatal extracellular DA levels were significantly increased in animals exposed to subsevere (19–20 min, at 37°C) asphyxia (approximately twofold), although the effect of D-amphetamine on extracellular DA levels was significantly decreased (≈50% compared to controls) (Fig. 3). Extracellular GABA levels in the substantia nigra were decreased by ≈50%. In animals with severe asphyxia (>20 min, at 37°C), a decrease in striatal DA levels was observed under basal (≈70%) and under D-amphetamine stimulation (≈50% compared to controls) *(86)*. The increase in basal DA levels in subsevere asphyxia could be in agreement with a histochemical study *(80)* showing that under the same conditions, asphyxia produced an increase in the number of tyrosine hydroxylase-immunoreactive (TH-IR) nerve cell bodies, which was considered to be sign of proliferation of dopaminergic neurons. A cause for this increased number of nigral DA cell bodies was suggested to be a deficit in the GABAergic striato-nigral feedback, which would set the nigrostriatal DA neurons in a hyperactive state. In agreement with this is the present finding that there is a decrease in nigral GABA levels and a decrease in the effect of D-amphetamine. Further studies combining microdialysis, immunohistochemistry, and quantitative histofluorometry are now in progress.

The present results show that perinatal asphyxia leads to death or to long-term neuronal deficits, affecting such systems as the nigrostriatal dopamine pathway. The extent of the damage appears to

depend directly on the length of the asphyctic period, as well as on the general metabolic condition under which asphyxia is induced. Thus, it is striking that decreasing the temperature from 37 to 30 or 15°C resulted in a significant increase in survival. This finding is in agreement with experimental studies demonstrating that low brain temperature protects brain neurons in rats subjected to transient forebrain ischemia *(82,83)*, an effect probably resulting from reduction in brain energy demands and a consequent decrease in the rate of ATP depletion *(84)*.

Summary

The interactions among different nuclei of the basal ganglia have been studied with in vivo microdialysis, in normal and lesioned rats. DA, ACh, glutamate, aspartate, GABA, adenosine, and neuropeptides have been simultaneously monitored and assayed with highly sensitive HPLC and RIA methods. Rotational behavior has also been recorded, together or in parallel with microdialysis.

The modulation of striatal DA release by striato-nigral and cortico-striatal pathways has been studied. It has been found that striato-nigral GABAergic and dynorphinergic pathways exert a negative feedback on striatal DA, whereas tachykininergic pathways exert a positive feedback on striatal DA. Cortical stimulation produces an increase in striatal DA release, probably via glutamatergic receptors of the kainate type. The interaction between cortex and striatum is, however, complicated by the fact that there are ipsilateral and contralateral cortical inputs, utilizing glutamate and CCK as transmitter signals.

Several experimental models have been developed to study the pathophysiology and therapy of neurodegenerative disorders of the basal ganglia. Different pathways of the basal ganglia are destroyed by intracerebral injections of selective toxins or surgical knife cuts. Thereafter, various drugs, including endogenous and exogenous trophic factors, are tested to reverse the effects induced by lesions.

A novel animal model to study the short- and long-term consequences of perinatal asphyctic lesions is presented. It has been found that perinatal asphyxia leads to death or to long-term neuronal deficits. The extent of the damage appears to depend on the length of the asphyctic period, as well as on the general metabolic condition under which asphyxia is induced. Hypothermia appears to be a powerful treatment to increase survival following severe asphyxia. The presented model is largely noninvasive on the pups, and it mimics the condition produced under labor in clinical situations. Therefore, it appears to be useful as a model for studying treatments to ameliorate the deleterious effects induced by hypoxic-ischemic lesions.

Acknowledgments

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