Mathematical Models of Neuromodulation and Implications for Neurology and Psychiatry

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Introduction

Mathematical models can test hypotheses of brain function in health and disease and can be used to investigate how different systems in the brain affect each other. We are particularly interested in how the electrophysiology affects the pharmacology and how the pharmacology affects the electrophysiology of the brain. Thus, many of the questions we address are on the interface between the electrophysiological and pharmacological views of the brain.

The human brain contains approximately 10^{11} neurons, each of which makes on the average several thousand connections to other neurons [66]. It is natural and comforting to think of the brain as a computational device, much like the computers that we build and understand. In this electrophysiological view of the brain, the neurons are the elementary devices and the way they are connected determines the functioning of the brain. But the brain is a much more flexible, adaptive, and complicated organ than this point of view suggests. The brain is made up of cells and there are several times as many glial cells as there are neurons. Glial cells protect neurons from oxidative stress by helping them synthesize glutathione [64], and astrocytes store glucose as glycogen [9]. Neurons synthesize and release more than 50 different kinds of neurotransmitters and myriad different receptor types allow neurons to influence each other's electrophysiology by volume transmission in which cells in

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one nucleus change the local biochemistry in a distant nucleus. This is the pharmacological view of the brain.

This is just the beginning of the full complexity of the problem. The functioning of neurons and glial cells is affected by an individual's genotype and dynamic changes of gene expression levels, on short and long time scales. These dynamic changes are influenced by the endocrine system, because the brain is an endocrine organ and is influenced by other endocrine organs like the gonads and the adrenal glands. And, although we think of the brain as producing behavior, in fact our behavior influences the electrophysiology, the pharmacology, and endocrine status of the brain, and therefore the gene expression levels. This is true both in the short term and in the long term. Individuals who exercise in their 30 and 40 s are 30 % less likely to get Parkinson's disease [3, 29] and the progression of Parkinson's symptoms is slower in those who exercise [38]. Thus the functioning of an individual brain depends on the history of environmental inputs and behavior throughout the individual's lifetime. And, we haven't even mentioned the complicated and changing anatomy, by which we mean the morphology of individual cell types, the connection patterns of neurons, and the proprioceptive feedback to the brain from the body [31].

Mathematical models are an important tool for understanding complicated biological systems. A model gives voice to our assumptions about how something works. Every biological experiment or psychology experiment is designed within the context of a conceptual model and its results cause us to confirm, reject, or alter that model. Conceptual models are always incomplete because biological systems are very complex and incompletely understood. Moreover, and as a purely practical matter, experiments tend to be guided by small conceptual models of only a very small part of a system, with the assumption (or hope) that the remaining details and context do not matter or can be adequately controlled.

Mathematical models are formal statements of conceptual models. Like conceptual models, they are typically incomplete and tend to simplify some details of the system. But what they do have, which experimental systems do not, is that they are completely explicit about what is in the model, and what is not. Having a completely defined system has the virtue of allowing one to test whether the assumptions and structure of the model are sufficient to explain the observed, or desired, results.

The Scientific Problem—Volume Transmission

The Electrophysiological View of the Brain

It is natural for us to think of the brain as a large computational device that processes information analogously to a computer. In this view, which we like to call the electrophysiological point of view, the basic elements are the neurons that receive inputs from other neurons and, via action potentials, send information to other neurons. There are then two fundamental classes of biological (and mathematical) questions.

How do individual neurons receive and process their inputs and decide when to fire? How do connected sets of neurons perform information processing functions that individual neurons cannot do? The electrophysiological point of view is natural for two reasons. First, we have had great success in building computational machines and we understand completely how they work. If brains are like our computational devices then we can use computational algorithms as metaphors and examples of what must be going on in the brain. Secondly, the electrophysiological point of view fits well with our modern scientific method of trying to understand complex behavior in the large as merely the interaction of many fundamental parts (the neurons) whose behavior we understand very well. The electrophysiological point of view is perfect for mathematical analysis and computation. One need not deal with the messy details of cell biology, the existence of more than 50 identified neurotransmitters, changing gene expression levels, the influence of the endocrine system, or the fact that neurons come in a bewildering variety of morphological and physiological types. All of these things appear, if they appear at all, as parameters in models of neurons, or as parameters in local or global network simulations. In particular, the chemistry of neurotransmitters themselves is not very important, since their only role is to help the electrophysiological brain transmit information from one neuron to the next.

The Pharmacological View of the Brain

There is a different point of view that we call the pharmacological view of the brain. It has been known for a long time that not all neurons are engaged in the one-to-one transfer of information to other neurons [39]. Instead, groups of neurons that have the same neurotransmitter can project densely to a distant volume (a nucleus or part of a nucleus) in the brain and when they fire they increase the concentration of the neurotransmitter in the extracellular space in the distant volume. This increased concentration modulates neural transmission in the distant region by binding to receptors on the cells in the target region. This kind of neural activity is called **volume transmission**. It is also called **neuromodulation** because the effect of the neurotransmitter is not one-to-one neural transmission but instead the modulation of other transmitters that are involved in one-to-one transmission. Two examples of volume transmission are the dopaminergic projection to the striatum in the basal ganglia from cells of the substantia nigra pars compacta and the serotonergic projection to the striatum from the dorsal raphe nucleus (DRN); these are discussed in more detail below. The locus coeruleus projects widely throughout the brain and spinal cord, with thin varicose norepinephrine (NE) axonal networks of low to moderate densities [45]. Projections of NE neurons from the locus coeruleus to the cortex play an important role in initiating and maintaining wakefulness [112].

There are many pieces of evidence that suggest that volume transmission plays a fundamental role in the functioning of the brain. Dopamine (DA) has been linked to fundamental brain functions such as motivation, pleasure, cognition, memory, learning, and fine motor control, as well as social phobia, Tourette's syndrome,

Parkinson's disease, schizophrenia, and attention deficit hyperactivity disorder [39]. In most experiments it is the concentration of dopamine in a particular nucleus that is important. Similarly, serotonin (5-HT) has been linked to feeding and body weight regulation, aggression and suicidality, social hierarchies, obsessive compulsive disorder, alcoholism, anxiety disorders, and affective disorders such as depression [39]. Many pharmaceutical and recreational drugs have been shown to act by binding to certain receptors and thus changing the local concentrations of various neurotransmitters in regions of the brain. For example, the immediate effect of selective serotonin reuptake inhibitors (SSRIs) is to inhibit the reuptake of 5-HT after it has been released thus increasing its concentration in the extracellular space in certain brain regions. Adenosine is an important neuromodulator that protects the brain from continuous neuronal activation: adenosine concentrations increase with neuronal activity and in turn inhibit individual neurons while also facilitating a transition into sleep, thereby promoting rest at a systemic level [34, 45]. Caffeine binds to adenosine receptors promotes wakefulness. Cocaine blocks the reuptake of DA, 5-HT, and norepinephrine [39] and has strong psychological effects.

Furthermore, various morphological and physiological features of the brain are consistent with the idea that the purpose of some neurons is to change the local biochemistry at distant regions of the brain. Often the projections are dense in the target volume suggesting that the idea is to change the local concentration at all parts of the target region simultaneously by the same amount. There are more than a dozen subtypes of receptors for 5-HT in the brain [91], suggesting that this great variety allows the concentration of 5-HT to modulate neurons in different ways depending on what receptors they express. As illustrated conceptually in Fig. 1, the 5-HT neurons in the dorsal raphe nucleus (DRN) have very thin unmyelinated axons and release 5-HT from many small varicosities rather than synapses [61], suggesting that their purpose is not one-to-one neural transmission. 5-HT neurons in different parts of the DRN project to many different brain regions that frequently project back, suggesting that the DRN is differentially changing the local biochemistry in many distinct regions [82], in particular the DRN sends a dense projection to the striatum

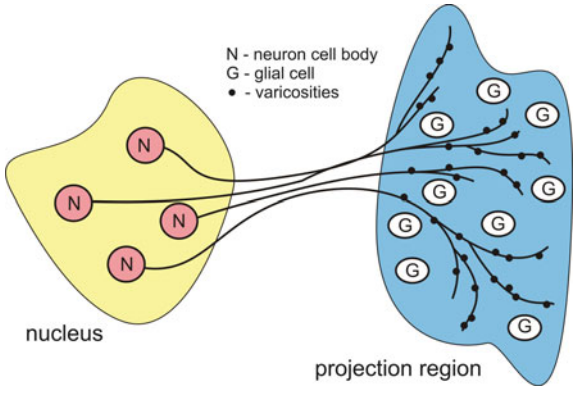

Fig. 1 Volume transmission and axonal varicosities

[106, 111]. The higher the concentration of 5-HT in the striatum, the more DA is released from the DA neurons projecting from the SNc per action potential [19, 22, 37]. Thus the neurotransmitters affect each other. There are also multiple DA receptor types in the striatum [6] that enrich neurotransmitter interactions with important functional consequences for the basal ganglia as we describe below.

Notice that what is important in volume transmission is that groups of neurons project to distant nuclei and change the local biochemistry there. That is, they project changes in biochemistry over long distances [97]. Of course they do this by firing action potentials. But the action potentials do not carry information in the usual sense; their only purpose is to allow the neurons to project biochemistry over long distances. This is the pharmacological view of the brain. To understand the brain one must understand both the electrophysiology and the pharmacology, and how they affect each other. For excellent reviews of volume transmission with a historical perspective and many examples, see [45, 117].

Volume Transmission and Balance in the Basal Ganglia

Here we discuss in more detail the dopaminergic volume transmission in the basal ganglia that motivates much of the computational work described in this chapter.

The basal ganglia (BG) are a group of subcortical nuclei including the striatum, subthalamic nucleus, internal and external globus pallidus, and substantia nigra. Cortical-BG-thalamic circuits are critically involved in many functions including sensorimotor, emotion, cognition [51, 75]. Multiple paths and subcircuits within BG have been identified. In some cases the different circuits perform different functions; for instance the striatum, the input nucleus of the BG, has anatomic and functional subdivisions including sensorimotor and associative. In other cases, pathways may compete, as has been postulated for action selection.

Two of the most studied pathways through the basal ganglia are the direct and indirect pathways through the striatum. The names reflect the fact that the direct pathway proceeds from the striatum directly to either the internal portion of the globus pallidus (GPi) or the Substantia Nigra pars reticulata (SNr), the two output nuclei of the BG. The indirect pathway, on the other hand, also involves a subcircuit that includes the external portion of the globus pallidus (GPe) and the subthalamic nucleus (STN) before reaching the output nuclei. The two pathways have opposing effects on the thalamus: the indirect pathway has an inhibitory effect, while the direct pathway has an excitatory effect [48, 103].

Albin and DeLong [4, 35] proposed that the balance of these opposing pathways is important for healthy function. Dopaminergic cells in the SNc project to the striatum and inhibit medium spiny neurons (MSNs) in the indirect pathway by binding to D2 receptors while the same neurons excite MSNs in the direct pathway by binding to D1 receptors [103]. Albin and DeLong noted that, during PD, as cells in the SNc die, less DA is released in the striatum; the result is that the direct pathway is less excited and the indirect pathway is less inhibited, so the thalamus receives

more inhibition and less excitation. Thus the loss of dopaminergic cells in the SNc has the effect of shifting the balance in favor of the indirect pathway, and they reasoned that the increased inhibitory output from BG to the thalamus might account for some of the motor symptoms of PD, such as bradykinesia and difficulty in initiating movement. This view later lost favor in the face of new experimental observations that appeared to contradict the Albin-DeLong theory. The fact that pallidotomy—lesioning the GPi—alleviates some PD motor symptoms fit well with the theory, but, paradoxically, it emerged that high frequency stimulation of GPi was equally effective therapeutically. The solution to this conundrum seemed to be that the pattern of neuronal firing in the BG was as important for symptoms as the rate of firing, which led some to dismiss the Albin-DeLong theory. Interestingly, as PD progresses, firing patterns in the GPi become bursty and cells become more synchronized [12, 62, 92]. Note that synchronous bursting and pausing result in higher amplitude variation in the GPi output compared to the uncorrelated, irregular firing observed in the healthy GPi and thus constitutes an effectively stronger signal. This observation allows the possibility that the Albin-DeLong theory retains merit but the notion of balance needs to be interpreted more generally, recognizing that not only firing rate but also firing patterns and correlation among cells can contribute to the strength of the signal. With this more general notion of balance, it is again widely hypothesized that many of the motor symptoms of PD are due to an imbalance between the direct and indirect pathways [48, 71, 115].

The projection from the SNc to the striatum is very dense and the evidence is strong that it is the DA concentration in the extracellular space that is important for keeping the balance between the direct and indirect pathways, not one-to-one neural transmission. In particular, DA agonists given to Parkinson's patients are somewhat successful in restoring function [24]. Thus, DA, projected from the SNc, is acting as a neuromodulator of the direct and indirect pathways.

Mathematical Modeling of Volume Transmission

It is clear that understanding the interaction between the electrophysiology of the brain and the pharmacology of the brain is fundamental to brain function in health and disease. Because of the myriad receptor types and the complicated anatomy of the brain, it is unlikely that there are simple recipes that specify how the phamacological-electrophysiological interactions work in different local brain regions and different functional systems. In this chapter, we review some of our investigations into those interactions, concentrating on serotonin and dopamine, especially in the basal ganglia. In section "Computational Methods" we indicate how we go about constructing our mathematical models by sketching our dopamine model. In section "A Serotonin Model", we show three applications of our 5-HT model. We show how the serotonin autoreceptors stabilize extracellular 5-HT in the face of genetic polymorphisms. We investigate how substrate inhibition of the enzymes tyrosine hydroxylase and tryptophan hydroxylase determines how sensitive

the brain concentrations of DA and 5-HT are to the content of meals. And, in section "Homeostasis of Dopamine" we propose a new mechanism of action for selective serotonin reuptake inhibitors. In section "Serotonin and Levodopa" we explain why levodopa is taken up by 5-HT neurons and is used in those neurons to make DA. A mathematical model is used to investigate the consequences for levodopa therapy for Parkinson's disease. Finally, in section "Homeostasis of Dopamine", we investigate various homeostatic mechanisms that stabilize the extracellular concentration of DA in the striatum.

Computational Methods

In 2009, we constructed a mathematical model of dopamine (DA) terminal [15] so that we could study synthesis, release, and reuptake and the homeostatic mechanisms that control the concentration of DA in the extracellular space. We investigated the substrate inhibition of tyrosine hydroxylase (TH) by tyrosine, the consequences of the rapid uptake of extracellular dopamine by the dopamine transporters, and the effects of the autoreceptors on dopaminergic function. The main focus was to understand the regulation and control of synthesis and release and to explicate and interpret experimental findings. We started with a model of a DA terminal because dopamine is known to play an important role in many brain functions. Dopamine affects the sleep-wake cycle, it is critical for goal-directed behaviors and reward learning, and it modulates the control of movement via the basal ganglia. Cognitive processing, such as executive function and other prefrontal cortex activities, are known to involve dopamine. Finally, dopamine contributes to synaptic plasticity in brain regions such as the striatum and the prefrontal cortex.

Dysfunction in various dopaminergic systems is known to be associated with a number of disorders. Reduced dopamine in the prefrontal cortex and disinhibited striatal dopamine release is seen in schizophrenic patients. Loss of dopamine in the striatum is a cause of the loss of motor control seen in Parkinson's patients. Studies have indicated that there is abnormal regulation of dopamine release and reuptake in Tourette's syndrome and dopamine appears to be essential in mediating sexual responses. Furthermore, microdialysis studies have shown that addictive drugs increase extracellular dopamine and brain imaging has shown a correlation between euphoria and psycho-stimulant-induced increases in extracellular dopamine. These consequences of dopamine dysfunction indicate the importance of maintaining dopamine functionality through homeostatic mechanisms that have been attributed to the delicate balance between synthesis, storage, release, metabolism, and reuptake. It is likely that these mechanisms exist both at the level of cell populations and at the level of individual neurons.

A schematic diagram of the mathematical model is given in the Fig. 2 that represents a DA terminal or varicosity. The boxes contain the acronyms of substrates and the ellipses the acronyms of enzymes and transporters. For convenience in the equations below and in the diagram we denote the concnetrations in the

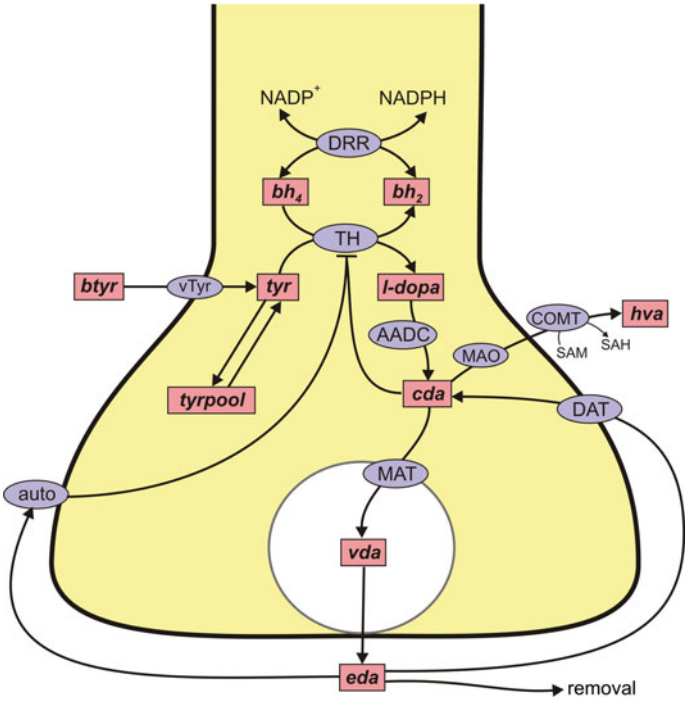

Fig. 2 Dopamine metabolism

mathematical model by lower case letters and we omit the brackets for concentration. Thus, *cda*, *vda*, and *eda* represent the concentrations of cytosolic DA, vesicular DA, and extracellular DA, respectively. Each arrow indicates a biochemical reaction, a transport velocity or an influence. Dopamine is synthesized in the nerve terminal from tyrosine *tyr* which is transported across the blood brain barrier. We include exchange between tyrosine and a tyrosine pool that represents all the other uses and sources of tyrosine in the terminal. Tyrosine is converted into L-3,4-dihydroxyphenylalanine, $l-dopa$, by tyrosine hydroxylase, *TH*, and $l-dopa$ is converted into cytosolic dopamine, *cda*, by aromatic amino acid decarboxylase, *AADC*. *cda* inhibits *TH* and is transported into the vesicular compartment by the monoamine transporter, *MAT*, and vesicular dopamine, *vda*, is released from the vesicular compartment into the extracellular space at a rate proportional to the firing rate of the neuron. In the extracellular space, extracellular dopamine, *eda*, affects the autoreceptors, is taken up into the terminal by the dopamine transporters, *DAT*, and is removed from the system by uptake into glial cells and the blood and by diffusion. Dopamine is also catabolized in the terminal by monoamine oxidase, *MAO*.

The variables in the mathematical model are the concentrations of the 10 boxed substrates. Each differential equation simply reflects mass balance: the rate of change of the concentration of the substrate is the sum of the rates by which it is being

made minus the sum of the rates by which it is lost. So, for example, the differential equation for *cda* is

$$\frac{dcda}{dt} = V_{\mathrm{AADC}}(l - dopa) + V_{\mathrm{DAT}}(eda) - V_{\mathrm{MAO}}(cda) - V_{\mathrm{MAT}}(cda, vda).$$

Each V is a velocity (a rate) and the subscript indicates which rate. These velocities depend on the current concentrations of one or more of the variables, as indicated. So, for example, V_{MAT} depends on both *cda* and *vda* because there is leakage out of the vesicles back into the cytosol. Similarly the differential equation for *eda* is

$$\frac{deda}{dt} = auto(eda)fire(t)(vda) - V_{DAT}(eda) - k(eda).$$

The first term on the right is release of dopamine into the extracellular space, which depends on the current state of the autoreceptors, *auto*(*eda*), the current firing rate, *fire*(*t*), and the vesicular concentration, *vda*. The second term is the uptake back into the terminal cytosol and the third term is removal in the extracellular space by uptake into glial cells and blood vessels.

Determination of the functional forms of the velocities. Each of the velocities, such as V_{TH}, V_{AADC}, or V_{DAT} depends on the current state of one or more of the variables. How do we determine the functional form of the dependence? If we can, we assume simple Michaelis-Menten kinetics. For example,

$$V_{\mathrm{DAT}}(eda) = \frac{V_{max}(eda)}{(K_m + eda)}.$$

In other cases the formula might be much more complicated depending on what is known about about how enzymes or transporters are activated or inhibited by molecules that are or are not its substrates. For example the formula for VTH is:

$$V_{\mathrm{TH}}(tyr, bh4, cda, eda) = \left(\frac{V_{max}(tyr)(bh4)}{(tyr)(bh4) + K_{tyr}(bh4) + K_{tyr}K_{bh4}(1 + \frac{(cda)}{K_{i(cda)}})}\right) \cdot \left(\frac{.56}{1 + \frac{(tyr)}{K_{i(tyr)}}}\right) \cdot \left(\frac{4.5}{8(\frac{eda}{.002024})^4 + 1} + .5\right)$$

The velocity V_{TH} depends on the current concentrations of its substrates, *tyr* and *bh*4, and on *cda* because *cda* inhibits the enzyme TH. The first term on the right is standard Michaelis-Menten kinetics with the additional inhibition by *cda*. The second term expresses the fact that *TH* is inhibited by its own substrate, *tyr*. The last term on the right is the effect of the autoreceptors and that depends on *eda*. Not so much is known about the mechanism of this effect, so we took a functional form that was consistent with in vitro experiments in the literature.

How are the parameters determined? The answer is, alas, with difficulty. There are measurements of K_m values in the literature. Sometimes they vary over two orders of magnitude, which is not surprising because often they are measured in test tubes or in vitro or measured in different cell types under different conditions. There are few V_{max} values in the literature because fluxes are hard to measure, especially in vivo. Typically we adjust the V_{max} values so that the concentrations at steady state in the model are in the ranges measured experimentally. We take the choice of parameters seriously, and we do the best we can.

But if we don't know the exact, correct values of the parameters, is the model right? This question (that we frequently get) is based on two misunderstandings. First, there are no exact, correct values of the parameters. In each of us, the parameters are somewhat different, because of genotype, because of environmental inputs, and because of changing gene expression levels. If we do our job well, we can hope to have a dopamine terminal system for an "average" person, and it is of course very relevant to ask how the behavior of the system depends on the parameters. Secondly, there's no "right" model. Every model is a simplification of a very complicated biological situation. We don't regard our models as fixed objects, or "right" or "wrong", but as growing and changing as we learn more from experiments and from our computations. The purpose of the model is to have a platform for in silico experiments that will increase our understanding of the biology.

Results

A Serotonin Model

We created a similar model for a serotonin (5-HT) terminal or varicosity [16] and have used the DA and 5-HT models to study various questions in brain physiology including depression and Parkinson's disease. In this section we provide three examples.

Homeostatic Effects of 5-HT Autoreceptors

The 5-HT_{1B} autoreceptors on terminals and varicosities provide a kind of end product inhibition for the extracellular 5-HT concentration. When the extracellular concentration rises, the autoreceptors inhibit synthesis (a long term effect) by inhibiting the enzyme TPH (the first step in the 5-HT synthesis pathway) and they also inhibit release of 5-HT from vesicles (a short term effect). Figure 3 shows some of the consequences autoreceptor regulation. In Panel A the firing rate is varied, in Panel B the activity of the serotonin transporter (SERT) is varied, and in Panel C the activity of tryptophan hydroxylase (TPH) is varied. TPH is the rate-limiting enzyme for the synthesis of 5-HT. In Panels B and C, common polymorphisms in the population are

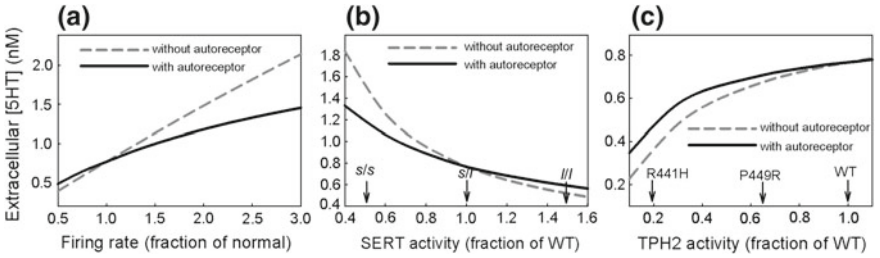

Fig. 3 Homeostatic effects of the 5-HT autoreceptors

shown on the x axis. In each panel, the y axis shows the concentration of 5-HT in the extracellular space at steady state in the model. The black curves show the steady state values in the model with the autoreceptors present and the dashed grey curves show the steady state values when the autoreceptors are turned off. In each case, the variation in extracellular 5-HT is much less in the presence of the autoreceptors. This shows how the autoreceptors buffer the extracellular concentration of 5-HT against changes in firing rate and genetic polymorphisms in two key proteins.

The Effect of Substrate Inhibition of TH and TPH

It is interesting that TH, the key enzyme for creating DA, and TPH, the key enzyme for creating 5-HT both show substrate inhibition. One can see this in the velocity curves in Fig. 4. That is, the substrate of the enzyme inhibits the enzyme itself. Substrate inhibition was emphasized by Haldane in the 1930s [55], but has been regarded as a curiousity although many enzymes exhibit this property. In all the cases that we have examined, substrate inhibition has a biological purpose [94]. In substrate inhibition, instead of the normal Michaelis-Menten shape where the velocity curve

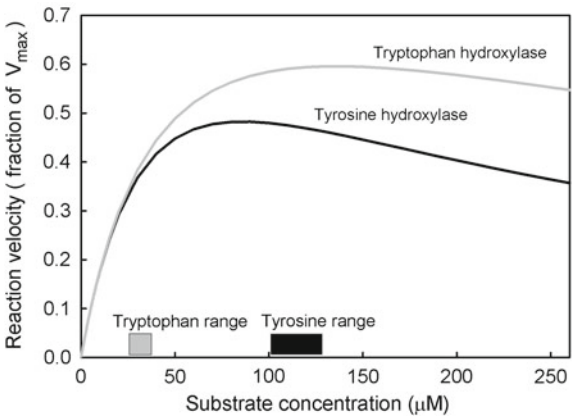

Fig. 4 Substrate inhibition of tyrosine and tryptophan hydroxylase

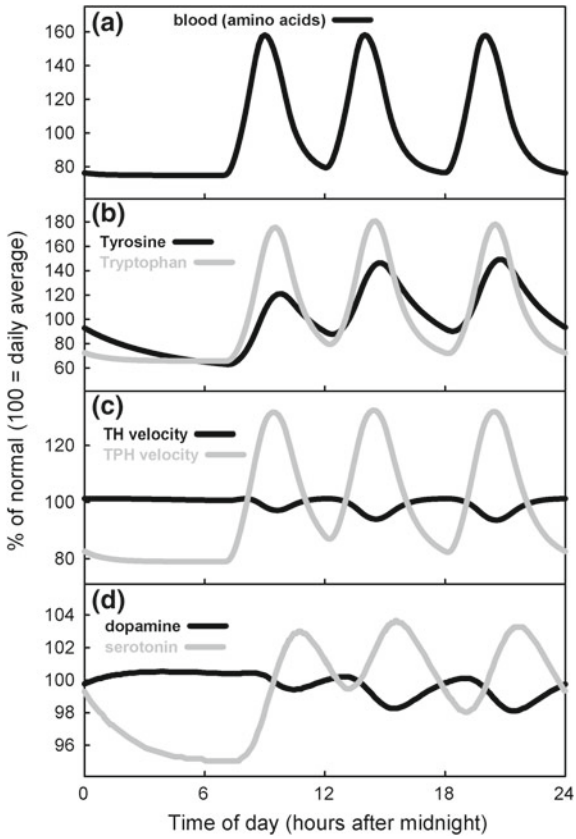

Fig. 5 5-HT and DA changes due to meals

saturates as the concentration of substrate gets large, the curves reach a maximum and then begin to descend as one can see in Fig. 4. The effect is much stronger for TH than for TPH. Does this matter? Well, that is something one can try out using the models. Panel A in Fig. 5 shows (assumed) amino acid curves in the blood for tyrosine and tryptophan due to three daily meals. Panel B shows the tyrosine and tryptophan concentrations in the cytosols of the terminals. Panel C shows the velocities of the TH and TPH reactions. Notice that the TH velocity varies little but the TPH velocity varies a lot. Why is that? The normal fasting concentration of tyrosine in the DA cells is 100–125 μM which puts it in the flat part of the TH velocity curve in Fig. 4, so changes in tyrosine in the cell don't change the synthesis rate of DA very much. In contrast, the synthesis rate of 5-HT varies quite a bit with the changes in blood and cytosolic tryptophan because the fasting concentration of tryptophan in 5-HT cells is on the sharply rising part of the velocity curve. As a consequence, the concentration of 5-HT in the vesicles (Panel 4) and the extracellular space (not shown) varies modestly while the concentration of DA varies very little. In fact, it is known that brain DA is quite insensitive to the protein content of meals [40, 41], but

that the brain content of 5-HT does vary with meals [39] and these simulations show why. For more information on substrate inhibition, see [94]. These simulations show that sometimes the details of the biochemistry, in this case the Michaelis-Menten constants of TH and TPH and the normal concentrations of tyrosine and tryptophan in cells, really do matter.

How Do SSRIs Work?

Depression, which is characterized by feelings of worthlessness and lack of motivation, is a major health problem and has important economic consequences (treatment and lost productivity) as well [21, 49]. The antidepressants used to treat depression are among the most widely prescribed drugs, but unfortunately most have unwanted side effects and limited therapeutic effects for most patients [47, 104]. In fact, none of the drugs are efficacious for a majority of the patients for whom it is prescribed [32, 80, 110]. Most of the commonly used antidepressants are selective serotonin reuptake inhibitors (SSRIs). It is not known what their mechanism of action is, or why they work on some patients and not on others. In this section we discuss these issues in the context of volume transmission.

Early evidence indicated that low 5-HT levels in the brain are linked to depression [39, 101]. 5-HT is synthesized in terminals and varicosities from the amino acid tryptophan and there is evidence that acute tryptophan depletion causes depression in humans [10, 114] and a decrease in 5-HT release in the rat hippocampus [107]. Thus it was natural to use SSRIs as antidepressants. Since SSRIs block the reuptake of 5-HT into the cytosol by the serotonin transporters (SERTs), it was expected that the SSRIs would raise the level of 5-HT in the extracellular space. Figure 6 shows a 5-HT neuron in the DRN sending its axon to a projection region where 5-HT is

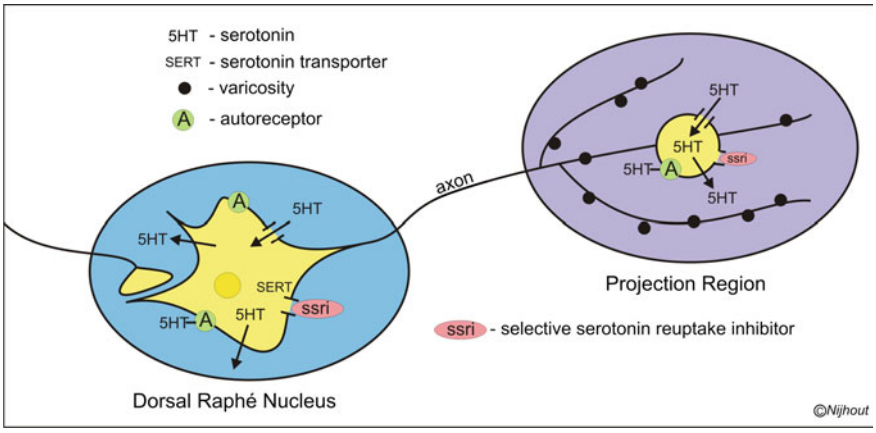

Fig. 6 SSRIs block the serotonin transporters

released from varicosities when the action potential arrives. Since the SSRIs block some of the SERTs, the effects of the SSRIs should be to raise the 5-HT level in the extracellular space in projection region. However, there is a complication. When a DRN neuron fires an action potential, 5-HT is also released from the cell body in the DRN [46]. The released 5-HT binds to 5-HT_{1A} autoreceptors on the cell bodies and the autoreceptors decrease cell firing when stimulated. Thus, when one gives an SSRI one blocks some SERTs in the projection region which should tend to make 5-HT go up there. However, the SSRIs also block SERTs in the DRN and the resulting increase of 5-HT in the extracellular space there will decrease firing and this will tend to *lower* extracellular 5-HT in the projection region. So, what will happen? The answer is that it depends on the balance between these two effects, and so it is not surprising that experimentalists found that 5-HT goes up in some projection regions and down in others and the magnitudes of the changes are dose dependent [1, 8, 57, 77]. Even at the beginning, it was clear that other effects besides the 5-HT concentration in the extracellular space must be important, because changes in concentration will happen in minutes and hours after an SSRI dose, but patients usually feel beneficial effects only after 3–6 weeks of treatment [39].

Attention focused next on the 5-HT_{1A} autoreceptors on the raphe nucleus cell bodies. It was shown in numerous studies (for example, [28]) that giving 5-HT_{1A} antagonists potentiates the SSRI-induced increase of 5-HT in projection regions. Similarly, 5-HT_{1A} knockout mice show increased release in projection regions [70]. Both types of studies confirm the role of the 5-HT_{1A} autoreceptors in decreasing tonic firing of 5-HT neurons in raphe in the presence of SSRIs. Furthermore, a number of studies showed that chronic treatment with SSRIs desensitizes the 5-HT_{1A} autoreceptors [20, 28, 58, 63]. And, thus, one could explain the improvements of patients on the time scale of 3–6 weeks by the slow desensitization of autoreceptors. Consistent with this hypothesis were several studies that showed that 5-HT levels in projection regions are higher after chronic treatment as compared to acute treatment [72, 100, 109]. These studies did not measure e5-HT in projections regions during the full course of chronic SSRI treatment. Unfortunately, when this was done, Smith et al. [105] found that extracellular 5-HT concentrations in neocortex, caudate, and hippocampus of awake monkeys went up initially and then declined somewhat over the course of treatment. Similar findings were found by Anderson et al. [5] who saw an initial quick rise in 5-HT in the cerebral spinal fluid of rhesus monkeys but then a plateau during chronic treatment. Thus, the autoreceptor desensitization hypothesis seems unlikely to explain the delay of beneficial effects of SSRI treatments.

In fact, the mechanisms of action of SSRIs are not understood, nor is it understood why some patients are helped and others not and why different SSRIs have different efficacies. The problem is extremely difficult because one has to understand mechanism and function on 4 different levels, genomic, biochemical, electrophysiological, and behavioral, but changes on each level affect function on the other 3 levels, and this makes the interpretation of experimental and clinical results very difficult. As we indicated above, the release of 5-HT affects dopamine signaling. 5-HT may activate the hypothalamic-pituitary-adrenal axis by stimulating production of corticotropin-

releasing hormone [56] and the endocrine system affects the 5-HT system [18, 105]. This may be the basis of gender differences in depression and response to SSRIs. And, finally, both gene expression and neuronal morphology are changing in time. In this circumstance, it is not surprising that many variables on all 4 levels are correlated with depression or to the efficacy of the SSRIs. All such correlations are candidates for causal mechanisms, so sorting out which mechanisms are causal is extremely difficult.

We used our mathematical model of serotonin varicosities and terminals to propose and investigate a new hypothesis about the action of SSRIs. The serotonin neurons in the DRN fire tonically at about 1 Hz but some of the individual spikes are replaced by short bursts. In a series of pioneering studies [42, 59, 65, 109], Jacobs, Fornal and coworkers studied the relationship between the electrophysiology of the 5-HT system and various behaviors in nonanaesthetized cats. They showed that the firing rate and pattern of some DRN 5-HT neurons differ in active waking, quiet waking, slow-wave sleep, and REM sleep [65]. And Hajos showed that some DRN neurons have bursts and others do not [53, 54]. Thus, it is plausible that the purpose of tonic firing is to maintain a basal 5-HT concentration in projection regions, but that it is bursts that contain incoming sensory input and stimulate specific behavioral responses.

If depression is caused by low tissue levels of 5-HT then vesicular 5-HT must be low in depressed patients since the normal concentrations in the cytosol and extracellular space are extremely low. So we assumed that our model depressed patient had low vesicular 5-HT, about 20–25 % of normal. For example, this could be caused by low tryptophan input. As a result, the model depressed patient had low extracellular 5-HT in projection regions. We modeled chronic treatment by SSRIs by assuming that the SSRIs block 2/3 of the SERTs. As we expected, this does not change extracellular 5-HT in projection regions very much because of the two competing effects discussed above. We then included the result of Benmansour et al. [11] that the expression level of SERTs declines considerably in rats during a 21 day treatment by SSRIs. Here are the results of our modeling. After 21 days, the average levels of 5-HT in projection regions of the model depressed patient did *not* return to normal. However, the response to bursts did return to normal. The intuitive reason behind this is that as the available SERTs decline considerably, reuptake of 5-HT becomes must slower. This has a much greater effect on bursts than on tonic firing because during bursts the extracellular 5-HT in projection regions is still high when the next action potential arrives. Thus, our proposed hypothesis is that it is burst firing that is connected to behavior and that, in depressed patients, the response to burst firing is brought back to normal by SSRIs because after 21 days the number of available SERTs is further depressed. It is interesting that Zoli et al. [116] emphasized that neurons that communicate via one-to-one neural transmission during tonic firing may contribute to volume transmission during burst firing. During bursts, the large amount of neurotransmitter in the extracellular space cannot be taken up in the synapse but spills out into the rest of the extracellular space.

Serotonin and Levodopa

Parkinson's disease has been traditionally thought of as a dopaminergic disease due to the death of dopaminergic cells in the substantia nigra pars compacta (SNc). These dopaminergic cells project to the striatum where low levels of DA cause dysfunction in the motor system. DA does not cross the blood-brain barrier because it doesn't have a carboxyl group and is not recognized as an amino acid. However, its precursor, L-DOPA, still has the carboxyl group and does cross the blood-brain barrier. Thus, the idea of levodopa therapy is to fill the remaining DA terminals in the striatum with L-DOPA so that these terminals will release more DA into the extracellular space when action potentials arrive, compensating for the DA terminal loss caused by cell death in the SNc. However, accumulating evidence implies an important role for the serotonergic system in Parkinson's disease in general and in physiological responses to levodopa therapy. We used a mathematical model [95] to investigate the consequences of levodopa therapy on the serotonergic system and on the pulsatile release of dopamine (DA) from dopaminergic and serotonergic terminals in the striatum.

Levodopa Makes DA in 5-HT Neurons

The key idea is the recognition of the similarities in the synthesis pathways of 5-HT in 5-HT neurons and DA in DA neurons. DA is synthesized from the amino acid tyrosine (tyr) that crosses the blood-brain barrier and is taken up into DA nerve terminals by the L-transporter. In the DA terminal, the enzyme tyrosine hydroxylase (TH) adds an OH group to tyr making levodopa (L-DOPA). We will abbreviate levodopa by L-DOPA and by LD. The amino acid decarboxylase (AADC) cuts off the carboxyl group to make cytosolic DA. The monoamine transporter (MAT) packages DA into vesicles. When the action potential arrives a sequence of events, including Ca^{++} influx, causes some vesicles to move to the boundary of the terminal and release their contents into the extracellular space. The extracellular DA is taken back up into the cytosol by the dopamine transporter (DAT). Extracellular DA also binds to DA autoreceptors (A-DA) that inhibit synthesis and release. This control mechanism stabilizes the concentration of DA. Of course, the actual situation is more complicated, for example, cytosolic DA itself inhibits TH and extracellular DA can be taken up by glial cells.

The situation for 5-HT is remarkably similar. 5-HT is synthesized from the amino acid tryptophan (tryp) that crosses the blood-brain barrier and is taken up into 5-HT nerve terminals by the L-transporter. In the 5-HT terminal, the enzyme tryptophan hydroxylase (TPH) adds an OH group to tryp making 5-HTP. The enzyme amino acid decarboxylase (AADC) cuts off the carboxyl group to make cytosolic 5-HT. The monoamine transporter (MAT) packages 5-HT into vesicles. When the action potential arrives, some vesicles to move to the boundary of the terminal and release their contents into the extracellular space. The extracellular 5-HT is taken back up

into the cytosol by the serotonin transporter (SERT). Extracellular 5-HT also binds to 5-HT autoreceptors (A-5HT) that inhibit synthesis and release.

The main difference between DA neurons and 5-HT neurons is that DA neurons express the enzyme TH and thus make DA, and 5-HT neurons express TPH and thus make 5-HT. As we will explain, this distinction is eliminated in 5-HT neurons when one gives a dose of levodopa.

L-DOPA is taken up into all cells by the L-transporter, just like tyr and tryp. When L-DOPA is taken up into 5-HT terminals, the enzyme AADC cuts off the carboxyl group to make DA, which is then packaged into vesicles by MAT. Thus vesicles in the 5-HT neurons are filled with both 5-HT and DA, and when the action potential arrives, both are released into the extracellular space. There is a large dense projection of 5-HT neurons from the dorsal raphe nucleus (DRN) to the striatum. So, during a dose of levodopa, the 5-HT neurons release a large pulse of DA into the striatum.

All the aspects of this story have been verified experimentally in the last 15 years. Experiments have verified that 5-HT neurons can store and release DA in vivo and in vitro [88]. In levodopa treatment of a hemiparkinsonian rat, striatal extracellular DA decreased substantially when the serotonergic system was lesioned [108]. Glial cells also express AADC and so could contribute to the conversion of LD to DA, but experiments using reserpine to block vesicular packaging showed a great reduction of extracellular DA, suggesting that most of the levodopa-derived DA is released by exocytosis of vesicles rather than by glia, at least at physiological levels of levodopa administration [67]. It has also been shown that 5-HT_{1A} autoreceptor agonists (that decrease DRN firing) and 5-HT_{1B} autoreceptor agonists (that decrease release at 5-HT terminals) both lower extracellular DA in the striatum in a dose-dependent manner after an LD dose [76].

The new understanding of 5-HT neurons in levodopa therapy has helped to explain a serious side effect of levodopa therapy. Within 5 years of chronic LD treatment, many patients experience a variety of complications [84, 85]. For instance, the length of the therapeutic time window in which a given LD dose relieves PD symptoms gradually shortens and approaches the plasma half-life of LD (wearing-off). Rapid variations in efficacy may occur (on-off fluctuations). Another, particularly troubling, complication of chronic LD therapy is the appearance of involuntary movements (levodopa-induced dyskinesia, LID). These complications increase patients disability substantially, pose a therapeutic dilemma, and limit the use of LD.

There is good evidence that large pulses of DA in the striatum are the proximal cause of LID that are seen in long-term dosing [44]. And there is conclusive evidence that these large pulses result from DA release from 5-HT neurons in the striatum. Lesioning the 5-HT system or giving selective serotonin autoreceptor (5-HT_{1A} and 5-HT_{1B}) agonists results in a nearly complete elimination of LID [26].

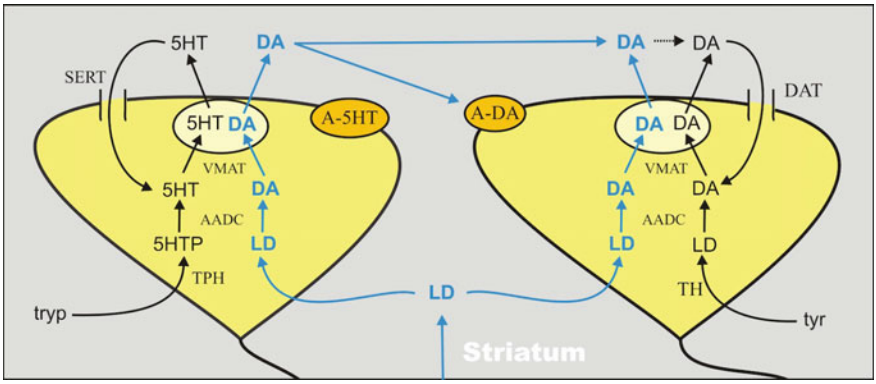

Fig. 7 Levodopa makes DA in 5-HT neurons

Mathematical Modeling

In order to investigate these phenomena, we created a mathematical model that corresponds to Fig. 7 [95]. What we discovered was that the size of these large pulses of DA coming from 5-HT neurons depends critically on the fraction, f, of SNc cells left alive, which is why there are more and more dyskinesias as Parkinson's disease progresses. Here is the intuitive explanation. As long as there are lots of SNc cells alive, there will be lots of DA terminals in the striatum with DATs and DA autoreceptors. The DATs take up a lot of the excess DA that comes from the 5-HT neurons and it is stored in DA terminals, and the DA autoreceptors restrict DA release from the DA terminals when the extracellular DA concentration is high. These effects keep the DA concentration in the extracellular space from going too high. However, as the fraction of SNc cells left alive gets smaller and smaller these two control mechanisms have less and less effect. The DA released from 5-HT neurons causes high pulses of DA in the striatum because it is not taken up quickly by the remaining DATs and it cannot be taken up into 5-HT terminals by the SERTs. It is these high pulses of DA in the striatum that lead to the aforementioned dyskinesias. In addition, the extra DA created by the levodopa dose and the 5-HT neurons is used up much faster because it cannot be stored in the small number of remaining DA terminals. It diffuses away or is taken up by glial cells, thus shortening the period of efficacy of the LD dose.

Figure 8, Panel A, shows model calculations of the time course of extracellular DA in the striatum for different values of f, the fraction of SNc cells left alive. Each curve is labeled with the corresponding value of f. As f declines from 1 (normal) to 0.2 and then 0.1, the level of extracellular DA gets higher because there are fewer DATs to take up the DA released by 5-HT neurons. However, when f is very small ($f = 0.05$ or 0.01), the peaks decline because removal mechanisms such as catabolism, diffusion, and uptake into glial cells become more important. The dashed black horizontal line in (A) represents the level of extracellular DA needed in the striatum for anti-Parkinsonian effects. In Panel B, the two solid curves reproduce the simulations

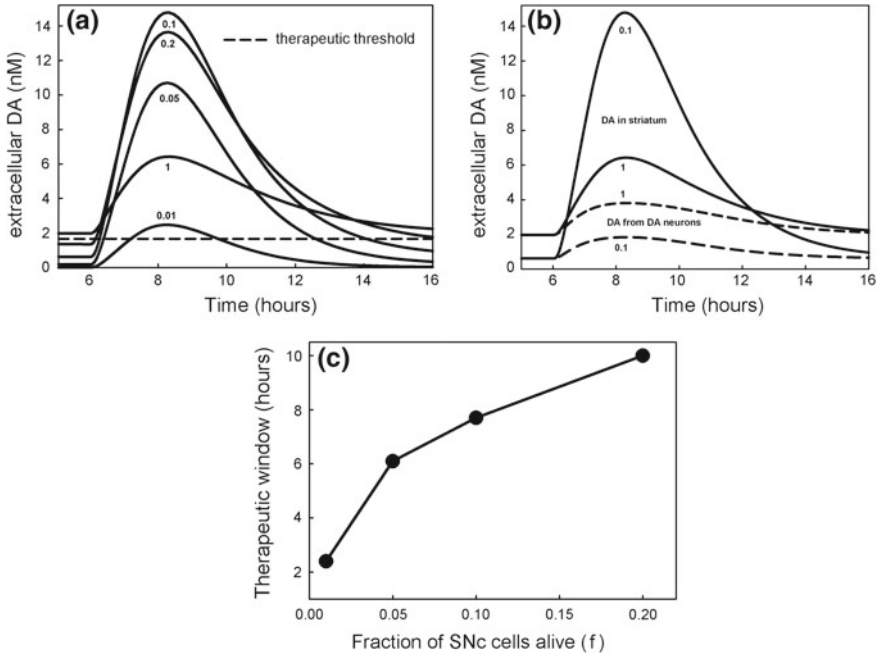

Fig. 8 5-HT effects as SNc cells die

from A for f = 1 and f = 0.1. The corresponding dashed curves in show the amount of extracellular DA in the striatum that comes from the DA neurons. For a normal individual (f = 1) the DA neurons contribute approximately 60 %, but as SNc cells die (f = 0.1) most of the DA comes from the 5-HT neurons.

Panel C shows that the amount of time that extracellular DA stays above the therapeutic level (the dashed black line in (A)) declines as PD progresses and f gets smaller until the therapeutic window becomes approximately 2 h.

How does an LD dose affect the 5-HT system? We will describe verbally what we saw in our modeling; details and figures can be found in [95]. First of all, LD competes with tyrosine and tryptophan for the L-transporter at the blood-brain barrier, so during an LD dose there is less tryptophan in the extracellular space in the brain. The tryptophan and LD compete again to be transported into 5-HT neurons, resulting in lowered tryptophan in 5-HT neurons. TPH turns some tryptophan into 5-HTP, but 5-HTP then has to compete with LD for the enzyme AADC that cuts off carboxyl groups and makes DA and 5-HT. Then, DA and 5-HT compete for the monoamine transporter, MAT, that packages them into vesicles. Thus it is not surprising that the concentration of 5-HT in the vesicles and in the extracellular space goes down approximately 50 % during an LD dose; Fig. 9 shows the extracellular 5-HT curve computed by the model. This drop in extracellular 5-HT is consistent with exper-

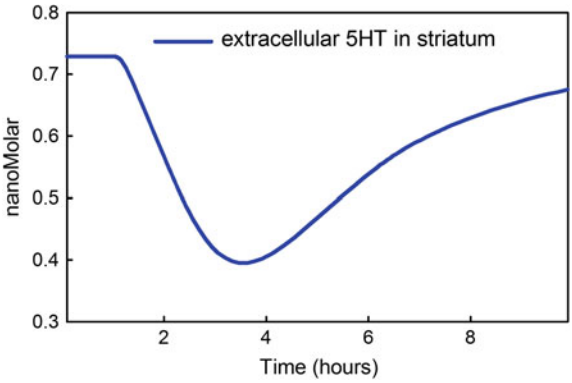

Fig. 9 LD dosing lowers 5-HT in the striatum

imental findings in animals. Extracellular 5-HT was found to decrease 50–90 % in different brain regions [23]. Carta et al. [26] found that tissue 5-HT decreased 48 % in the striatum during an LD dose, and Navailles et al. [86] showed that 5-HT decreases 30 % in the striatum and 53 % in motor cortex after chronic LD dosing. All this can be summed up by saying that dosing with LD turns 5-HT neurons partially into DA neurons, which is good for relieving the symptoms of Parkinson's disease but has the side effect of compromising the 5-HT system in the brain.

This raises the interesting question of whether levodopa doses could be one of the reasons for depression in Parkinson's patients. Decreased serotonergic signaling has been linked to depression [78]. As we pointed out above, acute tryptophan depletion is known to lower 5-HT brain levels in various animals [83], and results in lowered mood in humans [114]. While depression is frequently described as the most common psychiatric complication in PD [74], reported rates vary widely, from 2.7 % to greater than 90 % [98], due to factors including whether both major and minor depression are included and how subjects are selected for inclusion in the study. Moreover, many complicating factors make it difficult to draw conclusions about the possible connections between LD therapy and depression [43, 90, 95]. Nevertheless, the case of LD therapy and its effect of the 5-HT system is a cautionary tale. We create drugs and prescribe them because we expect them to have a specific effect in a specific brain location (in this case more DA in the striatum). However, the drug may have many other effects throughout the brain (in this case by lowering serotonin in all brain nuclei to which the raphe nuclei project).

Homeostasis of Dopamine

Neurotransmitters provide the mechanism by which electrical signals are communicated from one neuron to the next. However, as we discussed above, there is strong evidence that in many cases it is the concentration of a neurotransmitter in the extra-

cellular space of a nucleus that affects electrophysiological neurotransmission by other neurotransmitters. This volume transmission raises several natural questions. What are the mechanisms by which the extracellular concentrations of neurotransmitters are controlled? How do neurotransmitters in the extracellular space affect synaptic transmission by other neurotransmitters? How robust are these mechanisms in the face of polymorphisms in the enzymes affecting synthesis, release, and reuptake of neurotransmitters? How are dysfunctions in these control mechanisms related to neurological and neuropsychiatric diseases? In this section we briefly describe our work on several of these questions.

Passive Stabilization of DA in the Striatum

As discussed above, progressive cell loss from the substantia nigra pars compacta (SNc) is the proximal cause of the symptoms of Parkinson's disease [39]. The dopaminergic cells of the SNc send projections to the striatum where the loss of dopaminergic tone is thought to be the main cause of tremor and other motor symptoms of parkinsonism [25, 33]. An interesting and important feature of the disease is that symptoms do not appear until a very large percentage (75–90 %) of SNc cells have died and therefore this feature has been the focus of much experimental and clinical investigation [2, 118]. Experiments with animal models [13, 17, 36] have shown that although striatal tissue content of dopamine declines more or less proportionally to cell death in the SNc, the extracellular concentration of dopamine in the striatum remains near normal until more than 85 % of SNc neurons have died. This is widely believed to be the reason that symptoms do not appear until very late in the degeneration of the SNc.

What is the basis of this remarkable homeostasis of striatal extracellular DA in the face of progressive cell death in the SNc? Some researchers proposed that the nigrostriatal system adapts to cell death to maintain extracellular DA level by increasing DA synthesis in the living terminals or by sprouting more terminals. But in 2003, Bergstrom and Garris proposed a very simple explanation that they called "passive stabilization" and provided experimental evidence for it [13]. The extracellular concentration of DA depends on the balance between release of DA and reuptake of DA by the dopamine transporters (DATs). If half of the SNc cells die, there will be only half as much release, but there will also be only half as much reuptake, so the concentration of DA in the extracellular space should remain the same.

We used our mathematical model of a DA terminal to investigate the proposal of Bergstrom and Garris [13]. Notice that their hypothesis does not explain why passive stabilization breaks down when f, the fraction of SNc cells left alive, gets small. We believe that passive stabilization breaks down at small f because there is always some removal of DA from the system in the extracellular space by uptake into glial cells and blood vessels or simply diffusion out of the tissue. As the number of DA terminals in the striatum gets smaller, these removal effects get proportionally larger because the reuptake DATs become sparser and sparser. This hypothesis was confirmed and explained by our mathematical modeling. Figure 10 shows the con-

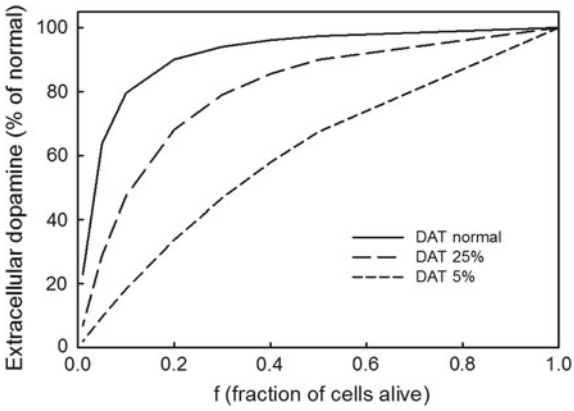

Fig. 10 DA concentration in the striatum as the fraction of SNc cells alive changes

centration of DA in the striatum in the model as a function of f, the fraction of SNc cells left alive. One can see that the passive stabilization effect of Bergstrom and Garris keeps the extracellular DA concentration quite constant until approximately 80 % of the SNc cells have died. As even more cells die the concentration drops to zero because the removal effects dominate more and more. The dashed curves show that the passive stabilization depends on the dopamine transporters.

Homeostasis of DA and Cryptic Genetic Variation

In our 2009 DA model [15] that included synthesis, packaging into vesicles, release, and reuptake via the DATs, we also included the effects of the DA autoreceptors that sense the DA concentration in the extracellular space. When extracellular DA gets higher than normal, the autoreceptors inhibit synthesis and release of DA, and when extracellular DA gets lower than normal this inhibition is withdrawn stimulating synthesis and release. Thus the autoreceptors act to modulate extracellular DA against both long term and short term perturbations such as changes in the supply of tyrosine or changes in firing rate. The mechanisms by which extracellular DA affects synthesis and release via the autoreceptors are mostly unknown and an important topic of current research that involves difficult questions in cell biology. The control of DA in the extracellular space is also affected by other neurotransmitters. For example, there is a dense serotonergic projection to the striatum from the dorsal raphe nucleus (DRN). The released 5-HT binds to 5-HT receptors on DA terminals and increases DA release when the SNc cells fire.

An important field of study in the past 15 years has been to quantify the effects of gene polymorphisms on the proteins that are important for the dopaminergic system, for example, tyrosine hydroxylase (TH) or the dopamine transporter (DAT). Typically, these experiments are done in test tubes or in vitro and the polymorphisms often have large quantitative effects on the function of the proteins. And, it is very tempting to conclude that the polymorphisms are therefore the causes of various

Table 1 Polymorphisms in TH and DAT

Gene	Mutation	Relative activity (%)	Citation
TH	T245P	150	[99]
TH	T283M	24	[99]
TH	T463M	116	[99]
TH	Q381K	15	[69]
DAT	V382A	48	[79]
DAT	VNTR10	75	[79]
DAT	hNET	65	[52]

neurological or neuropsychiatric diseases. Some of these polymorphisms that have large in vitro effects are shown in Table 1.

However, in vivo there are many control mechanisms (we've discussed two above) that buffer the DA concentration in the extracellular space against perturbations in the DA system. We pointed this out already in our 2009 paper [15], but the point is made dramatically by the two dimensional surface taken from [89]. The surface shows the extracellular DA concentration (z-axis) at steady state compared to normal, as a function of the activity of tyrosine hydroxylase and the efficacy of the dopamine transporter computed from our model. In both cases, 1 indicates normal activity for TH and DAT. The large white dot on the surface is wild type, the concentration of extracellular DA when TH and DAT have their normal activities. The smaller white dots on the surface indicate points that correspond to common polymorphisms (homozygotes and heterozygotes) in the human population taken from the table. Notice that all the white dots lie on the flat part of the surface where the polymorphisms cause only very modest changes in extracellular DA despite the fact that they cause large changes in protein activity. This is the effect of the autoreceptors. It is quite amazing that these polymorphisms all lie on the flat part of the surface. Presumably, if they didn't, they would have been selected against and would not be common in the human population. This example shows why one has to be very careful about jumping to physiological conclusions from in vitro experiments. The polymorphisms in Table 1 have large effects on the activity of the proteins but homeostatic mechanisms ensure that the effect on the phenotype (the extracellular DA concentration) is very small.

The surface in Fig. 11 is a perfect example of cryptic genetic variation in which large variation in genes (gene products), that is, TH and DAT, produce very little variation in a phenotypic variable, the extracellular DA concentration. It should be kept in mind that the actual situation is much more complicated than this two-dimensional surface in three-dimensional space would lead one to believe. There are many other variables, both genetic variables (for example a polymorphism in the monoamine transporter) or phenotypic variables (for example the 5-HT concentration, see below) that could affect the shape of this surface. The "real" surface is a high dimensional surface in a high dimensional space. Nevertheless this surface does tell us a lot, and

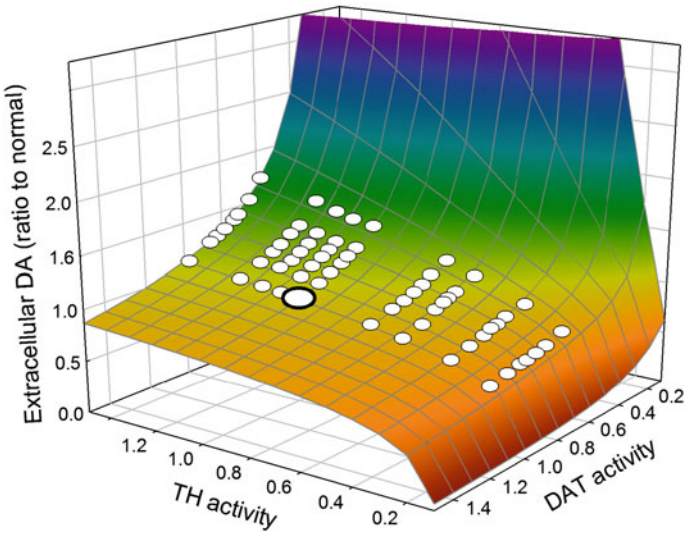

Fig. 11 DA homeostasis in the striatium. On each axis, 1 corresponds to normal

it is interesting to think about the people who have the 15 % mutation in TH. They are the ones to the right sitting at the edge of the cliff where DA drops to zero. Interestingly, these genotypes sometimes show a dystonia, involuntary muscle contractions that affect posture, brought about by low levels of extracellular DA that can be alleviated by levodopa [69]. So, one could say that their position at the edge of the cliff (that is, having the 15 % TH polymorphism) predisposes them to the dystonia. Some of them are pushed over the cliff by other variables not pictured and thus show the dystonia. The job of a precision medicine provider would be to advise a patient with the 15 % TH polymorphism how to flatten the region around where they lie on the surface and thus to avoid being pushed over the cliff by other variables. In our simulations, the region around these individuals gets flatter if one increases the strength of the autoreceptor effect.

Escape from Homeostasis and Neurological and Neuropsychiatric Diseases

In Parkinson's disease, many motor symptoms are caused by very low DA in the striatum of the basal ganglia, which, in turn, is caused by cell death of the dopaminergic neurons in the substantia nigra pars compacta (SNc) [39]. By contrast, the dyskinesias that may result from levodopa therapy are known to be caused by unusually high concentrations of extracellular DA in the striatum [44]. The chorea of Huntington's disease is also associated with high concentrations of extracellular DA in the striatum, that in turn may be caused by degeneration of inhibitory projections from

the striatum to the SNc [30]. There is a hypothesis that hyperactivity of dopamine transmission underlies many of the symptoms of schizophrenia [68]. The fact that amphetamines, cocaine, and other drugs that increase levels of extracellular DA cause similar symptoms to schizophrenia supports this hypothesis. These three diseases illustrate the idea that when one departs from the homeostatic region, either above or below, disease processes may occur.

Homeostasis does not mean that a system is rigid. In means that an important biological variable (in this case the extracellular DA concentration in the striatum) is kept within a narrow range by a host of homeostatic mechanisms. Note that not all variables are homeostatic; on the contrary, some variables change dramatically so that other variables can remain homeostatic [89]. Each of the homeostatic mechanisms works well only over a certain range of biological variation. If inputs or other biological parameters leave this range then the biological variable is no longer homeostatic and departs from its normal range and neurological and neuropsychiatric symptoms appear.

Does 5-HT Stabilize DA in the Striatum?

The 5-HT neurons in the dorsal raphe nucleus (DRN) send a dense projection to the basal ganglia, in particular to the striatum and the substantia nigra pars compacta [111]. We have discussed above that the projection to the striatum plays an important role in levodopa therapy for Parkinson's disease because much of the newly created DA during a dose comes from 5-HT terminals (see section "Serotonin and Levodopa"). The 5-HT projection to the striatum is an example of volume transmission. The released 5-HT binds to receptors on DA terminals, such as the ones from the SNc, and increases the release of DA in the striatum when the DA neurons fire [19, 22, 37]. Thus, increased firing in the DRN causes increased release of 5-HT in the striatum, which in turn causes inhibition of the indirect pathway and excitation of the direct pathway from the cortex through the striatum to the thalamus; see Fig. 12. This circuitry is even more complicated because there are excitatory projections from the thalamus to the cortex [102]. And the DRN sends projections to many regions in the brain and most of those regions project back to the DRN [82]. One of those returning projections is an inhibitory projection from the medial prefrontal cortex [27] pictured in Fig. 12.

Here is a plausible mechanism by which 5-HT release from DRN neurons could partially compensate for cell loss in the SNc. When SNc cells die, then some inhibition of the indirect pathway is withdrawn and some excitation of the direct pathway is withdrawn. Since the indirect pathway inhibits the thalamus and the direct pathway excites the thalamus, the effect of cell loss in the SNc is greater net inhibition of the thalamus. Since projections from the thalamus excite cortical neurons there will be less stimulation of the cortex. But the inhibitory projections from the medial prefrontal cortex will fire less, removing inhibition from the DRN. Thus the DRN will fire more, which will increase the release of DA from DA terminals in the striatum partially compensating for cell loss in the SNc. Thus the "purpose" of the 5-HT

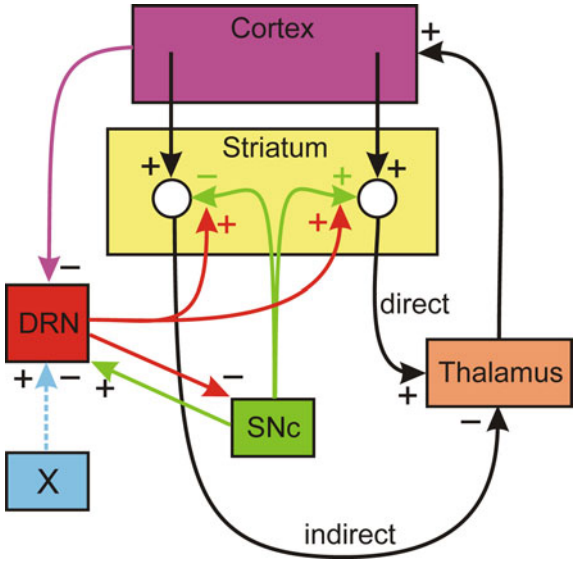

Fig. 12 A 5-HT circuit that could stabilize DA in the striatum

projection from the DRN to the striatum might be to stabilize the balance between the direct and indirect pathways against cell loss in the SNc. That this idea would work is supported by a simple mathematical model [96], but not enough is known about the details of projections from the thalamus to the cortex and from the cortex to the DRN to be sure of the anatomy.

"Take Home" Message for Neurologists, Psychiatrists

The complicated electrophysiological, pharmacological, and anatomical structure of the brain makes the design and delivery of drugs to achieve specific ends a very daunting challenge. There are several difficulties that are implicit in the examples that we've given in this chapter, but it is useful to make them explicit.

1. The brain contains many homeostatic mechanisms (in the electrophysiology, the pharmacology, the endocrinology, etc.) that tend to act to counter the intent of specific interventions. For example, as discussed in section "How Do SSRIs Work?", the original idea of SSRI development was to block the reuptake of 5-HT in projection regions and thus raise the 5-HT concentration in the extracellular space of the projection region. However, the SSRI will also block reuptake of the 5-HT released in the DRN when 5-HT cells fire. The released 5-HT stimulates the 5-HT$_{1A}$ autoreceptors on DRN cell bodies lowering the firing rate of the DRN 5-HT neurons, and this would tend to *lower* the 5-HT concentrations in projection regions.

2. A drug will not only affect the cells that you want it to affect, but, may also affect many other cells in the brain. A perfect example is the use of levodopa for Parkinson's patients where the intent is to increase the production of DA in the remaining SNc neurons. But levodopa is taken up by all cells of the body and in 5-HT varicosities and cell bodies it is used to manufacture and store DA in 5-HT neurons. This not only causes the large pulses of DA in the striatum that have been implicated in the development of dyskinesias, but also severely impairs the 5-HT system during the dose.
3. As emphasized by Fuxe et al. [45], brain cells express a myriad of different receptors. Often receptor populations are at locations distant from the endogenous sources of neurotransmitter and in fact may never be reached by endogenous transmitter [116]. When one gives an exogenous drug, one may stimulate receptors that are not normally stimulated under physiological conditions, and therefore the consequences are difficult to predict.
4. The brain is not homogeneous, receptors are not spread out evenly, and local consequences in one nucleus can differentially project to other brain regions. For example, suppose that one designs an antagonist for the 5-HT_{1A} receptors on the cell bodies of 5-HT neurons with the intent of raising DRN cell firing and the release of 5-HT in the striatum. Then, depending on the strength of projections, the 5-HT concentration will likely change differentially in all regions to which the DRN projects. Moreover, a number of studies have identified roles for 5-HT$_{1A}$ receptors in processes such as thermoregulation, immune function, and memory [91], where side effects might be anticipated.

The examples that we have given all involve volume transmission. Mathematical modeling of volume transmission can help us to understand the differential effects of drugs in local brain regions as well as side effects caused by projections to other regions. And, thus, mathematical modeling is an important tool for a better and more rational design of drug interventions.

There is another way in which the study of volume transmission can help us understand the brain. There are biophysical models of individual neurons and models of small and large networks of neurons. On the other hand, there are top-down models of behavior created by cognitive scientists in which different brain regions or nuclei are treated as black boxes and one studies how the regions influence each other and cause behavior. Connecting the models at these two very different levels is a difficult but important problem in brain science. Here the study of volume transmission can help because volume transmission operates at intermediate levels between these two types of models [50, 81, 87]. For example, studies of cholinergic modulation of neural circuits (volume transmission) have helped bridge understanding from the cellular effects of acetylcholine to its role in cognitive performance [87].

"Take Home" Message for Computationalists

Most of the mathematical modeling that has been done in computational and mathematical neuroscience addresses the electrophysiological view of the brain. All mathematical models are simplifications of the real biology, of course, and a natural simplification is that the brain consists of its electrical circuitry. If so, one should study the fundamental unit of the circuitry, the individual neuron, how neurons influence each other, and the behavior of small, medium, and large networks of interacting neurons. Because of the sheer size of the circuitry and the biological variation in individual neurons and connection patterns, these problems have been a fertile source of interesting biological and mathematical questions since the time of Hodgkin and Huxley [60]. All along it was understood that neurons are cells and that they are influenced by glia, local biochemistry, diet, the endocrine system, behavior, and changing gene expression levels, but it was hoped that those other details could be ignored because function arises mainly from the electrical circuitry. If function arises from the coordination of all those systems, then understanding function in the brain is a daunting challenge, indeed.

We believe, and we have been making the case in this chapter, that volume transmission is an important new area for computational and mathematical modelers who study the brain. By volume transmission, a local nucleus, for example the DRN or the SNc, can change the local biochemistry in the extracellular space in a distant nucleus. And, as we have indicated, there are 5-HT receptors on DA neurons that change the amount of DA released in the striatum, when the concentration of 5-HT goes up. So the different volume transmission systems are not independent, but affect each other. Unraveling these long distance biochemical networks and their interactions will be fundamental to understanding the brain in health and disease. In addition, the study of volume transmission raises interesting mathematical questions. We mention three such questions here.

If the purpose of a projection is to keep the neurotransmitter in the extracellular space within close upper and lower bounds, how precise does the placement of varicosities or terminals have to be to accomplish the goal? This is an interesting mathematical question because the neurons are firing stochastically, and the varicosities and terminals are both the sources of the neurotransmitter and the sinks into which it is absorbed. What is the spatial dependence in the extracellular space of the long term (average) neurotransmitter concentration? A natural first assumption would be to assume that the glial cells do not take up neurotransmitter, so in this case it is just a question of release, diffusion, and reuptake in the tortuous extracellular space. However, glial cells do take up neurotransmitters, and this means that the boundaries of the extracellular space are weakly absorbing. We have some preliminary results on these questions [73].

Secondly, though it has been known for years that autoreceptors play an important role in controlling the extracellular concentrations of neurotransmitters, not so much is known about the intracellular mechanisms involved in inhibiting synthesis and release or in the strengths of the inhibitions or the ranges over which they

operate. In most of our models, we suppose that the inhibition of synthesis and release depends on the current extracellular concentration of the neurotransmitter. However, recent experimental and computational evidence [113] shows that autoreceptor effects are long-lived. The autoreceptor effect can last 30–60 s after the concentration in the extracellular space has returned to normal. Thus models will have to take into account the dynamics of autoreceptor effects inside of cells.

How should volume transmission and pharmacology appear in electrophysiological network models? For firing rate models, the firing rate of a neural population might depend on the concentration of a neurotransmitter released from a presynaptic population. Behn and Booth [7] used such an approach in modeling the rat sleep-wake regulatory network. Thus they were able to simulate experiments in which neurotransmitter agonists and antagonists were microinjected into the locus coeruleus to see the effects on the structure of the sleep-wake cycle. In conductance-based models, such as those based on the Hodgkin-Huxley formalism, modulation can be accounted for through effects on model parameters [50]. Researchers often have found bifurcations in synaptic conductance parameters that dramatically change the dynamics of the system. On inspection, these conductance strengths would depend on local neurotransmitter concentrations through volume transmission. For example, in [14], the transition from normal to pathological (parkinsonian) neural activity may be due in part to an increased level of inhibition from the striatum; as discussed above, this is expected to result from decreased volume transmission of DA from the SNc. This is just one example of a neural circuit that would exhibit very different dynamics at different concentration levels of a neurotransmitter. This possibility shows that there will be very interesting dynamical systems questions in the interactions between the volume transmission network and local electrophysiological systems.

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References

1. Adell, A., Celada, P., Abella, M.T., Artigasa, F.: Origin and functional role of the extracellular serotonin in the midbrain raphe nuclei. Brain Res Rev **39**, 154–180 (2002)
2. Agid, Y.: Parkinson's disease: pathophysiology. Lancet **337**, 1321–1324 (1991)
3. Ahlskog, J.E.: Does vigorous exercise have a neuroprotective effect in Parkinson disease? Neurology **77**, 288–294 (2011)
4. Albin, R.L., Young, A.B., Penney, J.B.: The functional anatomy of basal ganglia disorders. Trends Neurosci. **12**, 366–375 (1989)
5. Anderson, G.M., Barr, C.S., Lindell, S., Durham, A.C., Shifrovich, I., Higley, J.D.: Time course of the effects of the serotonin-selective reuptake inhibitor sertraline on central and peripheral serotonin neurochemistry in the rhesus monkey. Psychopharmacology **178**, 339–346 (2005)

6. Beaulieu, J.M., Gainetdinov, R.R.: The physiology, signaling and pharmacology of dopamine receptors. Pharmacological Reviews **63**(1), 182–217 (2011)
7. Behn, C.D., Booth, V.: Simulating microinjection experiments in a novel model of the rat sleep-wake regulatory network. J Neurophysiol **103**, 1937–1953 (2010)
8. Bel, N., Artigas, F.: Fluoxetine preferentially increases extracellular 5-hydroxytryptamine in the raphe nuclei: an in vivo microdialysis study. Eur. J. Pharmacol. **229**, 101–103 (1992)
9. Belanger, M., Allaman, I., Magistretti, P.J.: Brain energy metabolism: focus on astrocyte-neuron metabolic coooperation. Cell Metabolism **14**, 724–738 (2011)
10. Benkelfat, C., Ellenbogen, M.A., Dean, P., Palmour, R.M., Young, S.N.: Mood-lowering effect of tryptophan depletion. Arch. Gen. Psych. **51**, 687–697 (1994)
11. Benmansour, S., Owens, W.A., Cecchi, M., Morilak, D., Frazer, A.: Serotonin clearance in vivo is altered to a greater extent by antidepressant-induced downregulation of the serotonin transporter than by acute blockade of the transporter. J. Neurosci. **22**(15), 6766–6772 (2002)
12. Bergman, H., Wichmann, T., Karmon, B., DeLong, M.: The primate subthalamic nucleus. II. neuronal activity in the MPTP model of parkinsonism. J Neurophysiol **72**, 507–520 (1994)
13. Bergstrom, B., Garris, P.: "Passive stabilization" of striatal extracellular dopamine across the lesion spectrum encompassing the presymptomatic phase of Parkinson's disease: a voltammetric study in the 6-OHDA-lesioned rat. J. Neurochem. **87**, 1224–1236 (2003)
14. Best, J., Park, C., Terman, D., Wilson, C.: Transitions between irregular and rhythmic firing patterns in an excitatory-inhibitory neuronal network. J Comp Neurosci **23**, 217–235 (2007)
15. Best, J.A., Nijhout, H.F., Reed, M.C.: Homeostatic mechanisms in dopamine synthesis and release: a mathematical model. Theor Biol Med Model **6**, 21 (2009)
16. Best, J.A., Nijhout, H.F., Reed, M.C.: Serotonin synthesis, release and reuptake in terminals: a mathematical model. Theor Biol Med Model **7**, 34– (2010)
17. Bezard, E., Dovero, S., C, C.P., Ravenscroft, P., Chalon, S., Guilloteau, D., Crossman, A.R., Bioulac, B., Brotchie, J.M., Gross, C.E.: Relationship between the appearance of symptoms and the level of nigrostriatal degeneration in a progressive 1-methyl-4-phenyl-1,2,3,6tetrahydropyridine-lesioned macaque model of Parkinson's disease. J Neurosci **21**, 6853–6861 (2001)
18. Birzniece, V., Johansson, I.M., Wang, M.D., Secki, J.R., Backstrom, T., Olsson, T.: Serotonin 5-HT1A receptor mRNA expression in dorsal hippocampus and raphe nuclei after gonadal hormone manipulation in female rats. Neuroendocrinology **74**(2), 135–142 (2001)
19. Blandina, P., Goldfarb, J., Craddock-Royal, B., Green, J.P.: Release of endogenous dopamine by stimulation of 5-hydroxytryptamine3 receptors in rat striatum. J. Pharmacol. Exper. Therap. **251**, 803–809 (1989)
20. Blier, P., de Montigny, C., Chaput, Y.: Modifications of the serotonin system by antidepressant treatment: implications for the therapeutic response in major depression. J. Clin. Psychoharmaco1. **7**, 24S–35S (1987)
21. Bloom, D.E., Cafiero, E.T., Jané-Llopis, E., Abrahams-Gessel, S., Bloom, L.R., Fathima, S., Feigl, A.B., Gaziano, T., Mowafi, M., Pandya, A., Prettner, K., Rosenberg, L., Seligman, B., Stein, A.Z., Weinstein, C.: The global economic burden of noncommunicable diseases. Harvard School of Public Health, World Economic Forum (2011)
22. Bonhomme, N., Duerwaerdère, P., Moal, M., Spampinato, U.: Evidence for 5-HT4 receptor subtype involvement in the enhancement of striatal dopamine release induced by serotonin: a microdialysis study in the halothane-anesthetized rat. Neuropharmacology **34**, 269–279 (1995)
23. Borah, A., Mohanakumar, K.P.: Long-term L-DOPA treatmeant causes indiscriminate increase in dopamine levels at the cost of serotonin synthesis in discrete brain regions of rats. Cell. Mol. Neurobiol. **27**, 985–996 (2007)
24. Brooks, D.J.: Dopamine agonists: their role in the treatment of Parkinson's disease. J. Neurol. Neurosurg Psychiatry **68**, 685–689 (2000)
25. Carlsson, A.: Perspectives on the discovery of central monoaminergic neurotransmission. Annu. Rev. Neurosci. **10**, 19–40 (1987)

26. Carta, M., Carlsson, T., Kirik, D., Björklund, A.: Dopamine released from 5-HT terminals is the cause of L-DOPA-induced dyskinesia in parkinsonian rats. Brain **130**, 1819–1833 (2007)
27. Celada, P., Puig, M.V., Casanovas, J.M., Guillazo, G., Artigas, F.: Control of dorsal raphe serotonergic neurons by the medial prefrontal cortex: Involvement of serotonin-1A, $GABA_A$, and glutamate receptors. J. Neurosci **15**, 9917–9929 (2001)
28. Chaput, Y., Blier, P., de Montigny, C.: *In vivo* electrophysiological evidence for the regulatory role of autoreceptors on serotonergic terminals. J. Neurosci **6**(10), 2796–2801 (1986)
29. Chen, H., Zhang, S.M., Schwarzschild, M.A., Hernan, M.A., Ascherio, A.: Physical activity and the risk of Parkinson disease. Neurology **64**, 664–669 (2005)
30. Chen, J., Wang, E., Cepeda, C., Levine, M.: Dopamine imbalance in Huntington's disease: a mechanism for the lack of behavioral flexibility. Front. Neurosci. **7** (2013)
31. Chiel, H.J., Beer, R.D.: The brain has a body: adaptive behavior emerges from interactions of nervous system, body, and environment. Trends Neuroscience **20**, 553–557 (1997)
32. Cipriani, A., Furukawa, T., Salanti, G., Geddes, J., Higgins, J., Churchill, R., Watanabe, N., Nakagawa, A., Omori, I., McGuire, H., Tansella, M., Barbui, C.: Comparative efficacy and acceptability of 12 new-generation antidepressants: a multiple-treatments meta-analysis. Lancet **373**, 746–758 (2009)
33. Cooper, J., Bloom, F., Roth, R.: The Biochemical Basis of Neuropharmacology. Oxford U. Press, New York, NY (2003)
34. Cunha, R.A.: Different cellular sources and different roles of adenosine: A_1 receptor-mediated inhibition through astrocytic-driven volume transmission and synapse-restricted A_{2A} receptor-mediated facilitation of plasticity. Neurochem. Int. **52**, 65–72 (2008)
35. DeLong, M.: Primate models of movement disorders of basal ganglia origin. TINS **13**, 281–285 (1990)
36. Dentresangle, C., Cavorsin, M.L., Savasta, M., Leviel, V.: Increased extracellular DA and normal evoked DA release in the rat striatum after a partial lesion of the substantia nigra. Brain Res **893**(178–185) (2001)
37. Deurwaerdère, P., Bonhomme, N., Lucas, G., Moal, M., Spampinato, U.: Serotonin enhances striatal overflow in vivo through dopamine uptake sites. J. neurochem. **66**, 210–215 (1996)
38. Duncan, R.P., M.Earhart, G.: Randomized controlled trial of community-based dancing to modify disease progression in Parkinson disease. Neurorehabilitation and Neural Repair **26**(2), 132–143 (2012)
39. Feldman, R., Meyer, J., Quenzer, L.: Principles of Neuropharmacology. Sinauer Associates, Inc, Sunderland, MA. (1997)
40. Fernstrom, J.: Role of precursor availability in control of monoamine biosynthesis in brain. Physiol. Rev. **63**, 484–546 (1983)
41. Fernstrom, J., Wurtman, R.D.: Brain serotonin content: physiological dependence on plasma tryptophan levels. Science **173**, 149–152 (1971)
42. Fornal, C., Litto, W., Metzler, C.: Single-unit responses of serotonergic dorsal raphe neurons to 5-ht1a agonist and antagonist drug administration in behaving cats. J. Pharmacol. Exper. Therap. **270**, 1345–1358 (1994)
43. Frisina, P.G., Haroutunian, V., Libow, L.S.: The neuropathological basis for depression in Parkinson's disease. Parkinsonism Relat Disord **15**(2), 144–148 (2009)
44. de la Fuente-Fernandez, R., Lu, J.Q., Sossi, V., Jivan, S., Schulzer, M., Holden, J.E., Lee, C.S., Ruth, T.J., Calne, D.B., Stoessl, A.J.: Biochemical variations in the synaptic level of dopamine precede motor fluctuations in Parkinson's disease: PET evidence of increased dopamine turnover. Annals of Neurology **49**(3), 298–303 (2001)
45. Fuxe, K., Dahlstrom, A.B., Jonsson, G., Marcellino, D., Guescini, M., Dam, M., Manger, P., Agnati, L.: The discovery of central monoamine neurons gave volume transmission to the wired brain. Prog. Neurobiol. **90**, 82–100 (2010)
46. Gartside, S.E., Umbers, V., Hajos, M., Sharp, T.: Interaction between a selective 5-HT1A receptor antagonist and an SSRI in vivo: effects on 5-HT cell firing and extracellular 5-HT. Br. J. Pharmacol. **115**, 1064–1070 (1995)

47. Gelenberg, A., Chesen, C.: How fast are antidepressants? J. Clin. Psychiatry **61**, 712–721 (2000)
48. Gerfen, C.R., Surmeier, D.J.: Modulation of striatal projection systems by dopamine. Annu. Rev. Neurosci. **34**, 441–466 (2011)
49. Gonzalez, O., Berry, J., McKight-Eily, L., Strine, T., Edwards, K., Kroft, J.: Current depression among adults - United States, 2006, 2008. Center for Disease Control and Prevention (2010)
50. Graupner, M., Gutkin, B.: Modeling nicotinic neuromodulation from global functional and network levels to nAChR based mechanisms. Acta Pharmacologica Sinica **30**, 681–693 (2009)
51. Haber, S.N., Calzavara, R.: The cortico-basal gangia integrative network: the role of the thalamus. Brain Research Bulletin **78**, 69–74 (2009)
52. Hahn, M., Mazei-Robison, M., Blakely, R.: Single nucleotide polymorphisms in the human norepinephrine transporter gene affect expression, trafficking, antidepressant interaction, and protein kinase C regulation. Mol. Pharmacol. **68**, 457–466 (2005)
53. Hajos, M., Gartside, S.E., Villa, A.E.P., Sharp, T.: Evidence for a repetitive (burst) firing pattern in a sub-population of 5-hydroxytryptamine neurons in the dorsal and median raphe nuclei of the rat. Neuroscience **69**, 189–197 (1995)
54. Hajos, M., Sharp, T.: Burst-firing activity of presumed 5-ht neurones of the rat dorsal raphe nucleus: electrophysiological analysis by antidromic stimulation. Brain Res. **740**, 162–168 (1996)
55. Haldane, J.: Enzymes. Longmans, Green and Co, New York (1930)
56. Heisler, L.K., Pronchuk, N., Nonogaki, K., Zhou, L., Raber, J., Tung, L., Yeo, G.S.H., O'Rahilly, S., Colmers, W.F., Elmquist, J.K., Tecott, L.H.: Serotonin activates the hypothalamic–pituitary–adrenal axis via serotonin 2C receptor stimulation. J. Neurosci. **27**(26), 6956–6964 (2007)
57. Hervás, I., Artigas, F.: Effect of fluoxetine on extracellular 5-hydroxytryptamine in rat brain. role of 5-HT autoreceptors. Eur. J. Pharmacol. **358**, 9–18 (1998)
58. Hervás, I., Vilaró, M.T., Romero, L., Scorza, M., Mengod, G., Artigas, F.: Desensitization of 5-HT_{1A} autoreceptors by a low chronic fluoxetine dose. effect of the concurrent administration of WAY-100635. Neuropsychopharmacology **24**, 11–20 (2001)
59. Heyn, J., Steinfels, G., Jacobs, B.: Activity of serotonin-containing neurons in the nucleus raphe pallidus of freely moving cats. Brain Res. **251**, 259–276 (1982)
60. Hodgkin, A.L., Huxley, A.F.: A quantitative description of membrane current and its application to conduction and excitation in nerve. The Journal of Physiology **117**(4), 500–544. (1952)
61. Hornung, J.P.: The human raphe nuclei and the serotonergic system. J. Chem. Neuroanat. **26**, 331–343 (2003)
62. Hurtado, J., Gray, C., Tamas, L., Sigvardt, K.: Dynamics of tremor-related oscillations in the human globus pallidus: a single case study. Proc. Nat. Acad. Sci. **96**, 1674–1679 (1999)
63. Invernizzi, R., Bramante, M., Samanin, R.: Citalopram's ability to increase the extracellular concentrations of serotonin in the dorsal raphe prevents the drug's effect in the frontal cortex. Brain Res. **260**, 322–324 (1992)
64. Iwata-Ichikawa, E., Kondo, Y., Miyazaki, I., Asanuma, M., Ogawa, N.: Glial cells protect neurons against oxidative stress via transcriptional up-regulation of the glutathione synthesis. J Neurochem **72**(6), 2334–2344 (1999)
65. Jacobs, B.L., Fornal, C.A.: 5-ht and motor control: a hypothesis. TINS **16**, 346–352 (1993)
66. Kandel, E., Schwartz, J., Jessell, T., Siegelbaum, S., Hudspeth, A.: Principles of Neural Science, 5th edn. McGraw-Hill Education/Medical (2012)
67. Kannari, K., Tanaka, H., Maeda, T., Tomiyama, M., Suda, T., Matsunaga, M.: Reserpine pretreatment prevents increases in extracellular striatal dopamine following L-DOPA administration in rats with nigrostriatal denervation. J Neurochem **74**, 263–269 (2000)
68. Kegeles, L., Abi-Dargham, A., Frankle, W., Gil, R., Cooper, T., Slifstein, M., Hwang, D.R., Huang, Y., Haber, S., Laruelle, M.: Increased synaptic dopamine function in associative regions of the striatum in schizophrenia. Arch. Gen Psychiat. **67**, 231–239 (2010)

69. Knappskog, P., Flatmark, T., Mallet, J., Lüdecke, B., Bartholomé, K.: Recessively inherited L-DOPA-responsive dystonia caused by a point mutation (Q381K) in the tyrosine hydroxylase gene. Hum. Mol. Genet. **4**, 1209–1212 (1995)
70. Knobelman, D.A., Hen, R., Lucki, I.: Genetic regulation of extracellular serotonin by 5-hydroxytryptamine$_{1A}$ and 5-hydroxytryptamine$_{1B}$ autoreceptors in different brain regions of the mouse. J. Pharmacol. Exp. Ther. **298**, 1083–1091 (2001)
71. Kravitz, A.V., Freeze, B.S., Parker, P.R.L., Kay, K., Thwin, M.T., Deisseroth, K., Kreitzer, A.C.: Regulation of parkinsonian motor behaviors by optogenetic control of basal ganglia circuitry. Nature letters **466**, 622–626 (2010)
72. Kreiss, D.S., Lucki, I.: Effects of acute and repeated administration of antidepressant drugs on extracellular levels of 5-HT measured in vivo. J. Pharmacol. Exper. Therap. **274**, 866–876 (1995)
73. Lawley, S., Best, J., Reed, M.: Neurotransmitter concentrations in the presence of neural switching in one dimension. AIM's J. **(under revision)** (2016)
74. Lemke, M.R.: Depressive symptoms in Parkinson's disease. European Journal of Neurology **15**(Suppl. 1), 21–25 (2008)
75. Lincoln, C.M., Bello, J.A., Lui, Y.W.: Decoding the deep gray: A review of the anatomy, function, and imaging patterns affecting the basal ganglia. Neurographics **2**, 92–102 (2012)
76. Lindgren, H.S., Andersson, D.R., Lagerkvist, S., Nissbrandt, H., Cenci, M.A.: L-DOPA-induced dopamine efflux in the striatum and the substantia nigra in a rat model of Parkinson's disease: temporal and quantitative relationship to the expression of dyskinesia. J. Neurochem. **112**, 1465–1476 (2010)
77. Malagie, I., Trillat, A.C., Jacquot, C., Gardier, A.M.: Effects of acute fluoxetine on extracellular serotonin levels in the raphe: an in vivo microdialysis study. Eur. J. Pharmacol. **286**, 213–217 (1995)
78. Mann, J.J.: Role of the serotonergic system in the pathogenesis of major depression and suicidal behavior. Neuropsychopharmacology **21**(2S), 99S–105S (1999)
79. Miller, G., Madras, B.: Polymorphisms in the 3'-untranslated region of the human and monkey dopamine transporter genes effect reporter gene expression. Mol. Psychiat. **7**, 44–55 (2002)
80. Moncrieff, J., Kirsch, I.: Efficacy of antidepressants in adults. Brit. Med. J. **331**, 155 (2005)
81. Montague, P.R., Dolan, R.J., Friston, K.J., Dayan, P.: Compuational psychiatry. Trends in Cognitive Sciences **16**(1), 72–80 (2012)
82. Monti, J.M.: The structure of the dorsal raphe nucleus and its relevance to the regulation of sleep and wakefulness. Sleep Med. Rev. **14**, 307–317 (2010)
83. Moore, P., Landolt, H.P., Seifritz, E., Clark, C., Bhatti, T., Kelsoe, J., Rapaport, M., Gillim, C.: Clinical and physiological consequences of rapid tryptophan depletion. Neuropsychopharmacology **23**(6), 601–622 (2000)
84. Mouradian, M.M., Juncos, J.L., Fabbrini, G., Chase, T.N.: Motor fluctutations in Parkinson's disease: pathogenetic and therapeutic studies. Annals of Neurology **22**, 475–479 (1987)
85. Mouradian, M.M., Juncos, J.L., Fabbrini, G., Schlegel, J., J.Bartko, J., Chase, T.N.: Motor fluctuations in Parkinson's disease: Central pathophysiological mechanisms, part ii. Annals of Neurology **24**, 372–378 (1988)
86. Navailles, S., Bioulac, B., Gross, C., Deurwaerdère, P.D.: Chronic L-DOPA therapy alters central serotonergic function and L-DOPA-induced dopamine release in a region-dependent manner in a rat model of Parkinson's disease. Neurobiol. Dis. **41**, 585–590 (2011)
87. Newman, E.L., Gupta, K., Climer, J.R., Monaghan, C.K., Hasselmo, M.E.: Cholinergic modulation of cognitive processing: insights drawn from computational models. Frontiers in Behavioral Neuroscience **6**(24), 1–19 (2012)
88. Nicholson, S.L., Brotchie, J.M.: 5-hydroxytryptamine (5-HT, serotonin) and Parkinson's disease - opportunities for novel therapeutics to reduce problems of levodopa therapy. European Journal of Neurology **9**(Suppl. 3), 1–6 (2002)
89. Nijhout, H.F., Best, J., Reed, M.: Escape from homeostasis. Mathematical Biosciences **257**, 104–110 (2014)

90. Pålhagen, S.E., Carlsson, M., Curman, E., Wålinder, J., Granérus, A.K.: Depressive illness in Parkinson's disease – indication of a more advanced and widespread neurodegenerative process? Acta Neurol Scand **117**, 295–304 (2008)
91. Raymond, J.R., Mukhin, Y., Gelasco, A., Turner, J., Collinsworth, G., Gettys, T., Grewal, J., Garnovskaya, M.N.: Multiplicity of mechanisms of serotonin receptor signal transduction. Pharmaco. Therap. **92**, 179–212 (2001)
92. Raz, A., Vaadia, E., Bergman, H.: Firing patterns and correlations of spontaneous discharge of pallidal neurons in the normal and tremulous 1-methyl-4-phenyl-1,2,3,6 tetra-hydropyridine vervet model of parkinsonism. J Neurosci. **20**, 8559–8571 (2000)
93. Reed, M., Best, J., Nijhout, H.: Passive and active stabilization of dopamine in the striatum. BioScience Hypotheses **2**, 240–244 (2009)
94. Reed, M., Lieb, A., Nijhout, H.: The biological significance of substrate inhibition: a mechanism with diverse functions. BioEssays **32**, 422–429 (2010)
95. Reed, M., Nijhout, H.F., Best, J.: Mathematical insights into the effects of levodopa. Frontiers Integrative Neuroscience **6**, 1–24 (2012)
96. Reed, M., Nijhout, H.F., Best, J.: Computational studies of the role of serotonin in the basal ganglia. Frontiers Integrative Neuroscience **7**, 1–8 (2013)
97. Reed, M., Nijhout, H.F., Best, J.: Projecting biochemistry over long distances. Math. Model. Nat. Phenom. **9**(1), 133–138 (2014)
98. Reijnders, J.S.A.M., Ehrt, U., Weber, W.E.J., Aarsland, D., Leentjens, A.F.G.: A systematic review of prevalence studies of depression in Parkinson's disease. Movement Disorders **23**(2), 183–189 (2008)
99. Royo, M., Daubner, S., Fitzpatrick, P.: Effects of mutations in tyrosine hydroxylase associated with progressive dystonia on the activity and stability of the protein. Proteins **58**, 14–21 (2005)
100. Rutter, J.J., Gundlah, C., Auerbach, S.B.: Increase in extracellular serotonin produced by uptake inhibitors is enhanced after chronic treatment with fluoxetine. Neurosci. Lett. **171**, 183–186 (1994)
101. Schildkraut, J.J.: The catecholamine hypothesis of affective disorders: a review of supporting evidence. Amer. J. Psych. **122**, 509–522 (1965)
102. Shepherd, G.M. (ed.): The Synaptic Organization of the Brain, 5th edn. Oxford U. Press, (2004)
103. Smith, Bevan, M., Shink, E., Bolam, J.P.: Microcircuitry of the direct and indirect pathways of the basal ganglia. Neuroscience **86**, 353–387 (1998)
104. Smith, D., Dempster, C., Glanville, J., Freemantle, N., Anderson, I.: Comparative efficacy and acceptability of 12 new-generation antidepressants: a multiple-treatments meta-analysis. Brit. J. Physchiatry **180**, 396– (2002)
105. Smith, T., Kuczenski, R., George-Friedman, K., Malley, J.D., Foote, S.L.: In vivo microdialysis assessment of extracellular serotonin and dopamine levels in awake monkeys during sustained fluoxetine administration. Synapse **38**, 460–470 (2000)
106. Soghomonian, J.J., Doucet, G., Descarries, L.: Serotonin innervation in adult rat neostriatum i. quantified regional distribution. Brain Research **425**, 85–100 (1987)
107. Stancampiano, R., Melis, F., Sarais, L., Cocco, S., Cugusi, C., Fadda, F.: Acute administration of a tryptophan-free amino acid mixture decreases 5-HT release in rat hippocampus in vivo. Am. J. Physiol. **272**, R991–R994 (1997)
108. Tanaka, H., Kannari, K., Maeda, T., Tomiyama, M., Suda, T., Matsunaga, M.: Role of serotonergic neurons in L-DOPA- derived extracellular dopamine in the striatum of 6-OHDA-lesioned rats. NeuroReport **10**, 631–634 (1999)
109. Tanda, G., Frau, R., Chiara, G.D.: Chronic desipramine and fluoxetine differentially affect extracellular dopamine in the rat pre-frontal cortex. Psychopharmacology **127**, 83–87 (1996)
110. Turner, E., Rosenthal, R.: Efficacy of antidepressants. Brit. Med. J. **336**, 516–517 (2008)
111. Vertes, R.P.: A PHA-L analysis of ascending projections of the dorsal raphe nucleus in the rat. J. Comp. Neurol. **313**, 643–668 (1991)
112. W., B.C., E., S.B., A., E.R.: Noradrenergic modulation of wakefulness/arousal. Sleep Med. Rev. **16**(2), 187–197 (2012)

113. Wood, K.M., Zeqja, A., Nijhout, H.F., Reed, M.C., Best, J.A., Hashemi, P.: Voltammetric and mathematical evidence for dual transport mediation of serotonin clearance in vivo. J. Neurochem. **130**, 351–359 (2014)
114. Young, S.N., Smith, S.E., Pihl, R., Ervin, F.R.: Tryptophan depletion causes a rapid lowering of mood in normal males. Psychopharmacology **87**, 173–177 (1985)
115. Zold, C.L., Kasanetz, F., Pomata, P.E., Belluscio, M.A., Escande, M.V., Galinanes, G.L., Riquelme, L.A., Murer, M.G.: Striatal gating through up states and oscillations in the basal ganglia: Implications for Parkinson's disease. J. Physiol-Paris **106**, 40–46 (2012)
116. Zoli, M., Jansson, A., Syková, E., Agnati, L., Fuxe, K.: Volume transmission in the CNS and its relevance for neuropsychopharmacology. Trends in Pharmacological Sciences **20**, 142–150 (1999)
117. Zoli, M., Torri, C., Ferrari, R., Jansson, A., Zini, I., Fuxe, K., Agnati, L.: The emergence of the volume transmission concept. Brain Res. Rev. **28**, 136–147 (1998)
118. Zygmond, M., Abercrombie, E.D., Berger, T.W., Grace, A.A., Stricker, E.M.: Compensation after lesions of central dopaminergic neurons: some clinical and basic implications. TINS **13**, 290–296 (1990)