Regulation of GABAergic Inhibition by Serotonin Signaling in Prefrontal Cortex

Molecular Mechanisms and Functional Implications

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Abstract

    Serotonergic neurotransmission in prefrontal cortex (PFC) plays a key role in regulating emotion and cognition under normal and pathological conditios. Increasing evidence suggests that serotonin receptors are involved in the complex regulation of GABAergic inhibitory transmission in PFC. Activation of postsynaptic 5-HT$_2$ receptors in PFC pyramidal neurons inhibits GABA$_A$-receptor currents via phosphorylation of GABA$_A$ receptor γ2 subunits by RACK1-anchored PKC. In contrast, activation of postsynaptic 5-HT$_4$ receptors produces an activity-dependent bi-directional regulation of GABA-evoked currents in PFC pyramidal neurons, which is mediated through phosphorylation of GABA$_A$-receptor β subunits by anchored PKA. On the presynaptic side, GABAergic inhibition is regulated by 5-HT through the activation of 5-HT$_2$, 5-HT$_1$, and 5-HT$_3$ receptors on GABAergic intereneurons. These data provide a molecular and cellular mechanism for serotonin to dynamically regulate synaptic transmission and neuronal excitability in the PFC network, which may underlie the actions of many antidepressant and antipsychotic drugs.

    Index Entries: Serotonin receptors; prefrontal cortex; GABA$_A$ receptors; inhibitory synaptic transmission; PKC; RACK1; PKA; AKAP; modulation.

Introduction

   The serotonergic system plays a key role in regulating emotional processes and cognitive

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behaviors in the central nervous system (CNS) *(1–3)*. Dysfunction of serotonergic neurotransmission has long been implicated inthe pathogenesis of neuropsychiatric disorders including schizophrenia, depression, and anxiety *(4–8)*. The new generation of antipsychotic agents, which constitutes a major advance in the treatment of schizophrenia, has high affin-

ity for serotonin receptors *(9–12)*. Drugs effective in treating depression also act mainly on the serotonergic system *(13,14)*.

The serotonergic system has a broad anatomical distribution in the CNS, mediating widespread effects in central neurons. One of the main target structures for the serotonergic system is prefrontal cortex (PFC), a collection of cortical areas in the most anterior portion of the frontal lobes. The anatomical organization of primate PFC can be divided into three major subregions: 1) dorsolateral, 2) orbitofrontal, and 3) medial PFC. In rats the prelimbic, infralimbic, and ventral anterior cingulate cortex represent the major subdivisions of PFC *(15)*. Based on their patterns of neural connectivity, these regions are thought to be related functionally to PFC in the primate *(16–18)*. The entire PFC is highly innervated by serotonergic projections from the brain stem raphe nuclei and dopaminergic projections from the ventral tegmental area *(19,20)*. The processed and integrated information is transmitted from PFC to distributed neural networks including cortex, thalamus, hypothalamus, brain stem, basal ganglia, and limbic areas. All subregions of PFC are robustly interconnected, suggesting that they participate in concert in central executive functions. The PFC has long been associated with high-level, "executive" processes needed for complicated goal-directed behavior *(21)*. Functional studies in primates emphasize that PFC is particularly critical for a form of short-term information storage described as "working memory" *(22)*. The firing of some PFC neurons increases throughout the delay period of a delayed-response task when information must be "held" momentarily and used subsequently to guide correct responses *(22)*. The excitability and tuning of PFC neurons are subject to modulatory influences by serotonin and many other neurotransmitters *(23)*. The damage of PFC in humans results in disturbances in a variety of functions, such as attention, memory, response selection, and planning.

PFC is composed of two major neuronal populations: 1) glutamatergic pyramidal principal neurons and 2) GABAergic interneurons. Local GABAergic neurons form numerous synapses on pyramidal projection neurons *(24)*, exerting strong inhibitory control on the excitatory output of PFC. One important function of the GABAergic inhibition in PFC is to control the timing of neuronal activity during cognitive operations and thereby shape the temporal flow of information *(25)*. Serotonergic projections target both types of PFC neurons in a synaptic and nonsynaptic manner *(26)*. Recent evidence shows that serotonin (5-HT) neurotransmission is predominantly paracrine, raising the possibility that 5-HT can act on receptors that are distant from its release site *(27)*. Specific changes of the PFC serotonin system and PFC neuronal activity have been found in patients with neuropsychiatric disorders *(4,5,7,28–31)*. It suggests that in PFC, serotonin plays a crucial and unique role in neural computation associated with the execution of complex tasks involved in cognition and emotion.

The pleiotropic functions of serotonin are afforded by the concerted actions of multiple serotonin receptor subtypes. The 5-HT receptors are composed of several families that can be grouped on the basis of conserved structures and signaling mechanisms *(32)*. The 5-HT$_1$ class of receptor (5-HT$_{1A, 1B, 1D, 1E, 1F}$) couples to G$_{i/o}$ proteins to inhibit adenylate cyclase. The 5-HT$_2$ class of receptor (5-HT$_{2A,2B,2C}$) couples to G$_q$ proteins to stimulate the turnover of phospholipids. The 5-HT$_4$ class of receptor (5-HT$_{4, 6, 7}$) couples to G$_s$ proteins to stimulate adenylate cyclase. The 5-HT$_5$ class of receptor (5-HT$_{5A, 5E}$) has unknown coupling. In addition, the 5-HT$_3$ receptor is a ligand-gated channel. Serotonin can have both inhibitory and excitatory functions in neuronal networks through the coupling of different 5-HT receptors to distinct ion channels *(33)*. Such a mechanism allows serotonin to simultaneously remodel excitability in a functionally appropriate manner in a wide variety of cell types and neuronal circuits.

Mice lacking different serotonin receptors exhibit diverse phenotypes including epilepsy syndrome *(34)*, increased impulsive aggression

(35), elevated anxiety, and antidepressant-like response *(36)*. Given the critical involvement of GABA$_A$ receptor-mediated inhibitory synaptic transmission in epilepsy, anxiety, and depression *(37)* the phenotypes in 5-HT receptor knockout mice may be correlated to changes in GABAergic transmission of PFC neurons. Indeed, selective alterations in GABA$_A$ receptors, GABA content and GABAergic interneurons have been discovered in PFC of patients with mental disorders *(30,38–40)*. These lines of evidence suggest that GABA$_A$ receptor is one of the cellular substrates of serotonin in PFC, and dysregulation of GABAergic inhibitory transmission by serotonin in PFC is a major contributing factor in neuropsychiatric diseases *(41)*.

The GABA$_A$ receptor contains an intrinsic chloride channel which is thought to be a heteropentameric structure, assembled by combining homologous subunits from five different classes: α (1–6), β (1–4), γ (1–3), δ and ε (1–2) *(37)*. The subunit composition of GABA$_A$ receptors critically affects the functional properties of GABA$_A$ channels. Multiple PKA and PKC phosphorylation sites have been identified in several GABA$_A$-receptor subunits *(42)*. Protein phosphorylation exerts a powerful regulation of GABA-activated currents in recombinant and native GABA$_A$ channels *(43–46)*. Reviewed here are some data showing how serotonin, by activating different receptor-mediated signaling cascades, regulate GABA$_A$-receptor channels and GABAergic inhibitory synaptic transmission in PFC neurons.

Regulation of GABA$_A$-Receptor Channels and GABAergic Synaptic Transmission by 5-HT$_2$ Signaling

Multiple G-protein-coupled 5-HT receptors have been identified in PFC pyramidal neurons *(47)*, among which 5-HT$_{2A}$ is one of the most prominent subtypes expressed in almost all (approx 90%) of these cells *(48)*. Application of 5-HT$_2$-class agonists produces a reduction of GABA$_A$ currents in most of PFC pyramidal neurons, and this effect is blocked by 5-HT$_2$ antagonists, confirming the mediation by 5-HT$_2$ receptors *(48)*. Postsynaptic 5-HT$_{2A}$ receptors are concentrated in apical dendrites proximal to the soma in PFC pyramidal neurons *(49)*, while GABA$_A$ receptors exhibit a compartmentalized distribution on postsynaptic domains of GABAergic synapses on the soma and proximal dendrites *(50)*. The overlap in the expression pattern between 5-HT$_{2A}$ and GABA$_A$ receptors suggests that they may be co-localized at some synapses in PFC pyramidal neurons. Such spatial organization would enable direct serotonergic modulation of local responses to inhibitory input, which offers a dynamic regulatory mechanism of synaptic integration, hence neuronal output.

Several lines of electrophysiological evidence indicate that the 5-HT$_2$ modulation of GABA$_A$-receptor currents is through a mechanism involving stimulation of phospholipid hydrolysis and activation of PKC *(48)*. Moreover, in vitro kinase assays show that treatment of PFC slices with a 5-HT$_2$ receptor agonist significantly increases the kinase activity of PKC towards GABA$_A$-receptor γ2 subunit, but not the β2 subunit over its already high basal level of phosphorylation *(48)*. These results suggest that activation of 5-HT$_2$ receptors in PFC can enhance the PKC catalytic activity, and potentiate PKC phosphorylation of GABA$_A$ receptors in a subunit-specific manner. Consistent with previous studies on PKC regulation of recombinant GABA$_A$ receptors *(45,51)*, the data have revealed that increased PKC phosphorylation of Ser-327 in γ2 subunit by 5-HT$_2$ signaling may play a major role in mediating the serotonergic modulation of GABA$_A$ currents in PFC neurons *(48)*.

The broad substrate selectivity of PKC makes it essential to control the specificity of this enzyme. Emerging evidence has suggested that one important mechanism for signaling enzymes to achieve fidelity and efficacy of signal transduction is through association with anchoring proteins that target to specific sub-
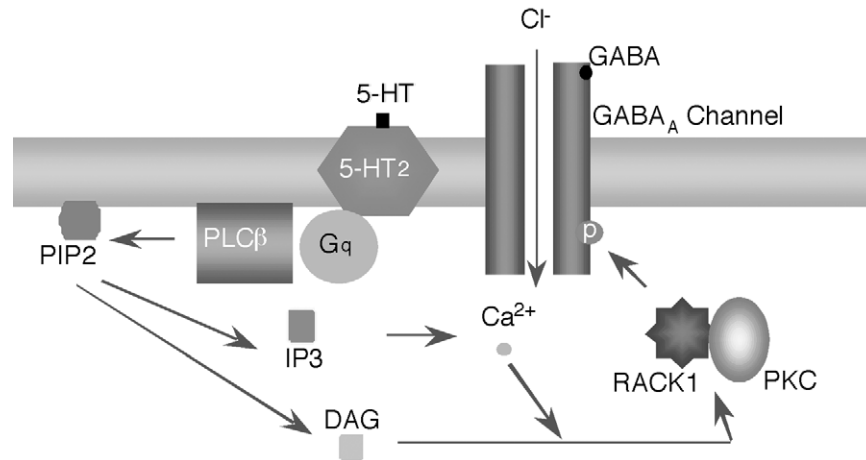
Fig. 1. A diagram illustrating the signal transduction cascade underlying 5-HT$_2$ regulation of GABA$_A$-receptor channels in PFC pyramidal neurons. Activation of 5-HT$_2$ receptors stimulates phospholipase C β isoform (PLCβ), leading to the release of ionsitol-1,4,5-triphosphate (IP$_3$) and diacylglycerol (DAG) through the hydrolysis of membrane phosphoinositol lipids. Activated PKC is targeted to GABA$_A$ receptors by the association with its anchoring protein RACK1, leading to the phosphorylation of GABA$_A$R subunits and reduction of GABA$_A$R-mediated currents.

cellular localization *(52–54)*. The compartmentalization and functions of PKC isoforms are also critically dependent on anchoring proteins *(55–57)*. RACK1, one member of the RACK family proteins, binds only activated PKC isoforms in a selective manner *(58)*. It facilitates the membrane translocation of PKC and PKC phosphorylation of specific substrates *(59–61)*. RACK1 is found to strongly associate with neuronal GABA$_A$ receptors in in vitro assays *(62)*, suggesting that RACK1 may be responsible for targeting PKC to GABA$_A$-receptor channels and allowing PKC to effectively phosphorylate these receptors. Consistent with this, when the PKC-RACK1 complex is disrupted by a PKC-anchoring inhibitory peptide derived from RACK1, which may lead to the removal of PKC from the proximity of GABA$_A$ receptors, the ability of 5-HT$_2$ agonists to inhibit GABA$_A$ currents is eliminated *(48)*. In contrast, disrupting the binding of PKC to another anchoring protein AKAP79 *(63)* does not affect the 5-HT$_2$ regulation of GABA$_A$ currents. These results suggest that targeting of activated PKC to GABA$_A$ receptors via RACK1 plays a critical role in the regulation of GABA$_A$

currents by the 5HT$_2$/PKC signaling pathway. Thus, activation of 5-HT$_2$ receptors causes a reduction of GABA$_A$-receptor currents through RACK1-anchored PKC (*see* Fig. 1), which suggests that serotonin signaling may suppress GABAergic inhibition in PFC circuits.

Recordings in frontal cortical slices have shown that an application of 5-HT through activation of 5-HT$_{2A}$ receptors induces a large, desensitizing enhancement of spontaneous inhibitory postsynaptic currents (sIPSCs), but decreases evoked eIPSCs in pyramidal neurons *(64)*. The increase in both frequency and amplitude of sIPSCs and its TTX sensitivity suggest that 5-HT$_{2A}$ receptors can enhance GABAergic inhibitory inputs to PFC pyramidal neurons. One potential reason is that the activation of 5-HT$_{2A}$ receptors on cortical GABAergic interneurons leads to an increase in their excitability *(64)*, possibly by inhibiting an inward rectifying potassium conductance *(65,66)* or by activating a cation nonselective current *(67,68)* resulting in the enhanced GABA transmission. On the other hand, the 5-HT reduction of eIPSC amplitude, but not the paired-pulse depression *(64)*, suggests that 5-

HT can also decrease the postsynaptic response to GABA in pyramidal neurons, possibly through a 5-HT$_2$-mediated intracellular signaling cascade *(48)*. This 5-HT$_2$-induced suppression of postsynaptic GABA$_A$-receptors may provide a negative feedback mechanism for 5-HT$_2$ regulation of GABAergic inhibition in PFC circuits.

Regulation of GABA$_A$-Receptor Channels by 5-HT$_4$ Signaling

In addition to 5-HT$_{2A}$ receptors, another serotonin receptor subtype that is enriched in PFC pyramidal neurons is 5-HT$_4$ *(69)*. About 60% of these cells express 5-HT$_4$ receptors *(48)*. Emerging evidence contends for a significant role of 5-HT$_4$ receptors in anxiolysis and cognition *(70)*. For example, the anxiolytic effect of diazepam (an enhancer of GABA response) is inhibited by application of 5-HT$_4$-receptor antagonists, particularly when the serotonergic tone is high *(71)*. Activation of 5-HT$_4$ receptors exerts ameliorative effects on spatial memory tests and reverses cognitive-performance deficits induced by cholinergic hypofunction *(70)*. In addition, marked reduction in 5-HT$_4$-receptor expression has been found in hippocampus and frontal cortex of patients with Alzheimer's disease *(72)*, consistent with the potential role of 5-HT$_4$ receptors in cognition. These findings suggest that 5-HT$_4$ receptors may play an important role in regulating ion channel activity, therefore affecting synaptic plasticity and neuronal excitability, which is fundamental in learning and memory. Previous studies have found that 5-HT$_4$-receptor activation exerts an excitatory impact on neurons through regulating different voltage-dependent ion channels *(73–76)*. A recent study *(77)* has identified the GABA$_A$-receptor channel as a molecular target of 5-HT$_4$ receptors in PFC.

Application of 5-HT$_4$-receptor agonists causes a bi-directional modulation of GABA$_A$ currents in PFC pyramidal neurons, with enhancement in some cells and reduction in others. The dual effects are mediated by PKA, because application of PKA activators mimics the dual effects of 5-HT$_4$ agonists on GABA$_A$ currents, while PKA inhibitors block the bi-directional regulation of GABA$_A$ currents by 5-HT$_4$ agonists *(77)*. Due to the broad substrate selectivity of PKA, subcellular targeting through association with anchoring proteins (AKAPs) has been effected as an important mechanism by which PKA achieves specificity in signal transduction *(78–80)*. Dialysis of the neurons with a peptide that can specifically disrupt the interaction between AKAPs and PKA *(78)* significantly attenuates the dual effects of 5-HT$_4$ agonist on GABA$_A$ currents *(77)*, suggesting that the 5-HT$_4$ modulation of GABA$_A$-receptor channels is a highly localized event that requires the fraction of PKA anchored on AKAPs.

Both PKA-induced enhancement and reduction of GABA$_A$ currents have been reported in recombinant systems and native neurons *(43,81)*. The dual effects of PKA seem to be attributable to phosphorylation of different GABA$_A$-receptor β subunits, since enhancement occurs when β3 subunit-containing receptors are phosphorylated by PKA on both S408 and S409, whereas reduction occurs when β1 subunit-containing receptors are solely phosphorylated on S409 *(82)*. However, single cell RT-PCR profiling results show that both β1 and β3 subunits are co-expressed in PFC pyramidal neurons *(77)*, suggesting that reasons other than the differential expression of β subunits may underlie the dual effects of 5-HT$_4$ agonists on GABA$_A$ currents. A set of experiments suggests that the bi-directional modulation of 5-HT$_2$ receptors on GABA$_A$ currents is dependent on the basal phosphorylation states of GABA$_A$ receptors *(77)*. In the same neuron, manipulations that decrease PKA activation levels switch the effect of 5-HT$_4$ receptors on GABA$_A$ currents from reduction to enhancement, while manipulations that increase PKA activation levels switch the effect of 5-HT$_4$ from enhancement to reduction. The molecular mechanism
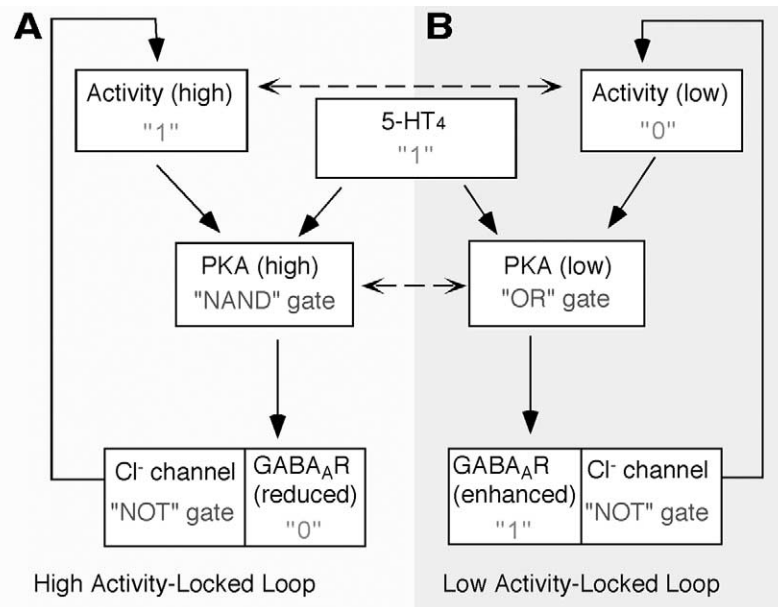
Fig. 2. A neural computational module for the 5-HT$_4$ actions in PFC pyramidal neurons. For those cells at the "high activity" state, the concomitant activation of PKA to a high level by 5-HT$_4$ receptors and high neuronal activity leads to a reduction of postsynaptic GABA$_A$-receptor functions ("NAND" gate), and a reduction of GABA-mediated inhibitory synaptic transmission, which results in the neurons being locked at the "high activity" state. However, for those cells at the "low activity" state, the concomitant activation of PKA to a low level by 5-HT$_4$ receptors and low neuronal activity leads to an enhancement of postsynaptic GABA$_A$-receptor functions ("OR" gate), and an enhancement of GABA-mediated inhibitory synaptic transmission, which results in the neurons being locked at the "low activity" state. "0": logic 0 state; "1": logic 1 state; "NAND" gate: a gate performing the "NOT AND" function, i.e., the output is "0" only when two inputs are both "1";"OR" gate: a gate performing the logic that the output is "1" as long as there is at least one input at "1";"NOT" gate: a gate performing the logic that the output is "1" when the input is "0" and vice versa. (Reproduced with permission from ref. *77*.)

underlying the changes in the polarity of 5-HT$_4$ effects remains to be determined. Differential PKA phosphorylation of different β subunits of GABA$_A$ receptors provides one potential mechanism. At low basal activation levels, PKA may preferentially phosphorylate β3 subunits first in response to a 5-HT$_4$ agonist. This leads to the current enhancement. On the contrary, at high basal activation levels, PKA may cause the phosphorylation of both β1 and β3 subunits in response to the activation of 5-HT$_4$ receptors. The dominant effect of β1 phosphorylation over β3 phosphorylation leads to the current reduction.

How do different PFC pyramidal neurons have varied basal PKA activation levels under physiological conditions? The role of neuronal activity was further tested *(77)*. Several lines of evidence indicate that the direction of 5-HT$_4$ regulation is likely to be activity-dependent. First, only reduction of GABA$_A$ currents by a 5-HT$_4$ agonist is observed in neurons isolated from PFC slices with high neuronal activity. Secondly, the 5-HT$_4$–mediated enhancement of the amplitude of GABAergic sIPSCs in PFC pyramidal neurons in slices is converted to a reduction after KCl-induced elevation of excitability. Furthermore, it has been found

that the PKA activation level is increased when neuronal activity is enhanced by membrane depolarization in the PFC network. Thus, the direction of 5-HT$_4$ modulation of GABA$_A$-receptor channels is dependent on the intracellular PKA activation level, which is determined by neuronal activity. Increased neuronal activity can switch the effect of 5-HT$_4$ receptors on GABA$_A$ channels from enhancement to reduction.

Based on the experimental data, a model which illustrates the potential roles that 5-HT$_4$ receptors may play in neural computation in PFC pyramidal neurons has been proposed *(77)*. The activity-dependent bi-directional regulation of GABAergic signaling by 5-HT$_4$ receptors could potentially form a neural computational module namely, "Activity-Locked Loop (ALL)" (*see* Fig. 2), which could be simplified to resemble digital circuits controlled by logic gates. This ALL module would allow serotonin, by acting on 5-HT$_4$ receptors, to use key molecules (e.g. PKA) within a single neuron to perform complex neural computation in maintaining the activity state of a neuron. Non-physiological, activity-uncoupled perturbations of this module at different levels could disrupt the normal reinforcement of this ALL module, leading to altered output that could have dramatic behavioral consequences. This activity-dependent bi-directional modulation of GABA$_A$ receptors by 5-HT$_4$ receptors provides a novel mechanism for neuromodulators to have either excitatory or inhibitory functions under different physiological conditions *(33,83)*. This mechanism ensures that the modulation is flexible, accurate, and dynamic.

Regulation of GABAergic Transmission by 5-HT$_1$ Receptors

The 5-HT$_{1A}$ receptor is highly concentrated in the limbic system, especially in prefrontal cortex and hippocampus *(84,48)*, as well as in the raphe. Postsynaptic 5-HT$_{1A}$ receptors are found only in the dendritic compartment and associated with dendritic spines *(85)*. Growing attention is being directed towards developing pharmacological agents that target 5-HT$_{1A}$ receptors for the treatment of schizophrenia, anxiety, depression, and cognition disorders *(86–88)*. Increased prefrontal 5-HT$_{1A}$-receptor density has been found in schizophrenia patients *(28)*. 5-HT$_{1A}$-receptor deficient mice show consistently elevated anxiety alongside antidepressant-like response *(36)*. Activation of 5-HT$_{1A}$ receptors has detrimental effects on working memory, and 5-HT$_{1A}$ antagonists ameliorate the cognitive impairment *(89)*.

A potential mechanism underlying these actions of 5-HT$_{1A}$ receptors is the change of synaptic transmission and neuronal activity through regulation of ion channels. Previous electrophysiological studies have shown that 5-HT$_{1A}$ receptors can induce a membrane hyperpolarization by activating the inwardly rectifying potassium channels *(90,91)*. 5-HT$_{1A}$ receptors are also capable of inhibiting multiple classes of calcium channels *(92,93)*. Application of 5-HT$_{1A}$-receptor agonists has no effect on postsynaptic GABA$_A$-receptor currents in pyramidal neurons of PFC and hippocampus *(48,94)*. However, hippocampal slice recordings show that activation of 5-HT$_{1A}$ receptors reduces polysynaptic fast- and slow-inhibitory postsynaptic potentials in CAI projection neurons *(94)*. Presynaptic mechanisms involving the inhibition of GABAergic interneurons have been proposed for the 5-HT$_{1A}$–mediated reduction of polysynaptic inhibition *(94)*. Whether 5-HT$_{1A}$ receptors elicit a similar effect on GABAergic inhibitory synaptic transmission in PFC slices remains to be examined.

Another member of the 5-HT$_1$ subfamily, 5-HT$_{1B}$, is expressed in about 60% of PFC pyramidal neurons *(48)*, and localized predominantly on axon terminals *(95)*. Intracellular recordings show that activation of 5-HT$_{1B}$ receptors in cingulate cortex depresses GABAergic inhibitory postsynaptic potentials evoked by stimulation to the subcortical white matter via a presynaptic mechanism *(66)*. These data suggest that activation of presynap-

tic 5-HT$_{1B}$-receptors reduces the release of GABA at synapses onto PFC pyramidal neurons, and at synapses onto local feed-forward inhibitory interneurons. Similarly, the GABA$_A$-receptor-mediated synaptic input originating from the striatum to substantia nigra pars reticulata neurons is also attenuated by presynaptic 5-HT$_{1B}$ receptors *(96)*. The mechanisms underlying 5-HT$_{1B}$-mediated inhibition of GABAergic transmission are unclear.

Regulation of GABAergic Transmission by 5-HT$_3$ Receptors

Unlike other G protein-coupled serotonin receptors, 5-HT$_3$ receptors are ion channels on plasma membrane *(97,98)*. An inward current can be induced by 5-HT$_3$-receptor activation in a variety of neurons *(99,100)*. The current is mediated by a cationic influx, activated rapidly upon application of serotonin and desensitized upon prolonged exposure to serotonin. Immunohistochemical studies indicated that 5-HT$_3$ receptors are expressed only in a subset of GABAergic neurons in neocortex and hippocampus *(101)*. Activation of 5-HT$_3$ receptors in these inhibitory interneurons evokes a large and rapidly desensitizing inward current and induces a transient enhancement of sIPSCs in these brain regions *(64,102)*. Though the functional significance of this selective expression of 5-HT$_3$ receptors is unclear, it is certain that a short pulse of 5-HT can directly excite fast-spiking GABAergic interneurons, leading to a transient increase in GABAergic synaptic activity onto targets innervated by these interneurons. The enhanced inhibition may underlie the 5-HT$_3$-mediated suppression of the firing rate of PFC neurons *(103)*.

Conclusion

The serotonergic system is involved in the regulation of diverse psychophysiological processes including mood, aggression, percep-

tion, memory, and anxiety. To mediate this large array of brain functions, a complex set of serotonin receptors has evolved. Alterations of 5-HT-receptor activity have been shown to occur in many psychiatric diseases. A number of effective psychopharmacological agents for these disorders have been developed which either specifically alter brain levels of serotonin or bind to 5-HT-receptor subtypes. To search for new therapeutic agents with higher specificity and efficacy, but lower side effects and toxicity, we need to know more about the physiological functions of serotonin in critical brain regions such as the prefrontal cortex. Genetic and behavior studies have provided important knowledge about the actions of serotonin at the systematic level *(34–36)*. The cellular and molecular mechanisms underlying serotonin functions are lacking. By combining electrophysiological, pharmacological, biochemical, and molecular analyses, the data reviewed here demonstrates that by activating different receptors, serotonin plays an important role in regulating GABAergic inhibition in PFC. The information gained from these studies should provide an important basis for understanding physiological functions of serotonergic system in the complex cortical neurocircuits under normal and patahological conditions.

From the postsynaptic loci, 5-HT$_2$ receptors on PFC pyramidal neurons inhibit GABA$_A$-channel activity *(48)*, while 5-HT$_4$ receptors exert an activity-dependent bi-directional regulation of GABA$_A$ channel activity *(77)*. With the convergence of both 5-HT$_2$/PKC and 5-HT$_4$/PKA pathways on the same target—postsynaptic GABA$_A$ receptors, serotonin can exert a complex regulation of the GABAergic inhibitory transmission in PFC. For those pyramidal neurons that express 5-HT$_2$, but not 5-HT$_4$ receptors, 5-HT may induce a reduction of postsynaptic GABA$_A$-receptor function just through the 5-HT/PKC-mediated cascade. For those pyramidal neurons that express both 5-HT$_2$ and 5-HT$_4$ receptors, the 5-HT effect on postsynaptic GABA$_A$ receptors could be complicated. The 5-HT$_2$/PKC-induced change of

$GABA_A$ receptor γ2 or β2 subunit phosphorylation would compete with the $5\text{-}HT_4$/PKA-induced change of $GABA_A$ receptor β1 or β3 subunit phosphorylation, and therefore determine the final effect of 5-HT on the channel function. To determine which of the two modifications plays the dominant role may depend on a variety of factors, such as the relative abundance and subcellular localization of $5\text{-}HT_2$ vs $5\text{-}HT_4$ receptors, basal levels of phosphorylated $GABA_A$ receptors by constitutively active PKC vs PKA, efficacy in altering $GABA_A$-receptor phosphorylation by the 5-HT receptor-activated PKC/RACk1 vs PKA/AKAP complex, and varied activity states of different PFC pyramidal neurons.

In addition to the postsynaptic modulation of $GABA_A$ receptors by 5-HT in pyramidal neurons, the effect of 5-HT on the excitability of GABAergic interneurons and on the release probability at GABAergic terminals also plays a key role in determining the impact of 5-HT on inhibitory transmission. On one hand, 5-HT could enhance GABAergic transmission by directly exciting GABAergic interneurons via the activation of $5\text{-}HT_{2A}$ or $5\text{-}HT_3$ receptors *(64)*. However, 5-HT could also suppress GABAergic transmission via the activation of presynaptic $5\text{-}HT_{1A}$ or $5\text{-}HT_{1B}$ receptors *(94,66)*. To establish which of the two effects plays the dominant role may depend on the expression and properties (e.g., binding affinity and agonist-induced desensitization) of these 5-HT receptors, as well as their coupling to intrinsic ion channels *(64–68)* or release machinery. It has been shown that serotonin at the crayfish neuromuscular junction increases the number of vesicles available for transmitter release, by either an increase in the number of vesicles at each release site, or an activation of previously nonsecreting or silent synapses *(104)*. Further studies need to be explored in order to examine whether serotonin in central neurons also regulate synaptic vesicle recycling, and what receptors are mediating this presynaptic effect of serotonin on synaptic transmission.

With so many different receptors acting at both pre- and postsynaptic sites, it is hard to predict what will happen to PFC neuronal activity in response to serotonin. In vivo studies have demonstrated that the predominant effect of 5-HT on cortical pyramidal neurons is an inhibition of spontaneous spiking *(105,106)*. The presynaptic increase of GABA transmission mediated by $5\text{-}HT_2$ and $5\text{-}HT_3$ receptors *(64)*, together with the enhancement of postsynaptic responses to GABA by $5\text{-}HT_4$ receptors in neurons with "low activity" *(77)*, could be one of the underlying mechanisms for the inhibitory actions of 5-HT. However, in vitro, studies suggest that 5-HT induces depolarization and action potential firing in pyramidal neurons *(66,67)*. The decrease of postsynaptic responses to GABA by $5\text{-}HT_2$ receptors *(48)* and by $5\text{-}HT_4$ receptors in neurons with "high activity" *(77)*, as well as the presynaptic suppression of GABA transmission mediated by $5\text{-}HT_{1A}$ and $5\text{-}HT_{1B}$ receptors, could be one of the originating mechanisms for the excitatory actions of 5-HT. Indeed, serotonergic regulation of glutamatergic transmission *(107,108)* could be another important mechanism to regulate PFC neuronal activity. The complex cellular expression and subcellular locations of different 5-HT receptors, their coupling to different intracellular signaling components, and their impact on multiple targets make it too simplistic to catagorize the actions of 5-HT receptors as excitatory vs inhibitory, or cooperative vs antagonistic. However, integration of all the information should help us to understand how serotonin, by acting on various receptor subtypes, modulates the electrophysiological properties of PFC neurons in a coordinated fashion to achieve their precise functions within neuronal networks. Elucidation of signaling components critically engaged in serotonergic actions could provide the potential targets for novel pharmacological agents in the treatment of neuropsychiatric disorders.

Acknowledgment

References

1. Buhot M. C. (1997) Serotonin receptors in cognitive behaviors. *Curr. Opin. Neurobiol.* **7,** 243–254.

2. Davidson R. J., Putnam K. M., and Larson C. L. (2000) Dysfunction in the neural circuitry of emotion regulation—a possible prelude to violence. *Science* **289,** 591–594.

3. Manji H. K., Drevets W. C., and Charney D. S. (2001) The cellular neurobiology of depression. *Nat. Med.* **7,** 541–547.

4. Breier A. (1995) Serotonin, schizophrenia and antipsychotic drug action. *Schizophr. Res.* **14,** 187–202.

5. Dubovsky S. L. and Thomas M. (1995) Serotonergic mechanisms and current and future psychiatric practice. *J. Clin. Psychiatry* **56 (Suppl. 2),** 38–48.

6. Abi-Dargham A., Laruelle M., Aghajanian G. K., Charney D., and Krystal J. (1997) The role of serotonin in the pathophysiology and treatment of schizophrenia. *J. Neuropsychiatry Clin. Neurosci.* **9,** 1–17.

7. Stockmeier C. A. (1997) Neurobiology of serotonin in depression and suicide. *Ann. NY Acad. Sci.* **836,** 220–232.

8. Deakin J. F. (1998) The role of serotonin in panic, anxiety and depression. *Int. Clin. Psychopharmacol.* **13 (Suppl. 4),** S1–5.

9. Meltzer H. Y. (1995) Role of serotonin in the action of atypical antipsychotic drugs. *Clin. Neurosci.* **3,** 64–75.

10. Busatto G. F. and Kerwin R. W. (1997) Perspectives on the role of serotonergic mechanisms in the pharmacology of schizophrenia. *J. Psychopharmacol.* **11,** 3–12.

11. Kapur S. and Remington G. (1996) Serotonin-dopamine interaction and its relevance to schizophrenia. *Am. J. Psychiatry* **153,** 466–476.

12. Lieberman J. A., Mailman R. B., Duncan G., Sikich L., Chakos M., Nichols D. E., and Kraus J. E. (1998) Serotonergic basis of antipsychotic drug effects in schizophrenia. *Biol. Psychiatry* **44,** 1099–1117.

13. Fuxe K., Ogren S. O., Agnati L. F., Benfenati F., Fredholm B., Andersson K., Zini I., and Eneroth P. (1983) Chronic antidepressant treatment and central 5-HT synapses. *Neuropharmacology* **22,** 389–400.

14. Deakin J. F. (1988) 5HT2 receptors, depression and anxiety. *Pharmacol. Biochem. Behav.* **29,** 819–820.

15. Groenewegen H. J. (1988) Organization of the afferent connections of the mediodorsal thalamic nucleus in the rat, related to the mediodorsal-prefrontal topography. *Neuroscience* **24,** 379–431.

16. Kolb B. (1984) Functions of the frontal cortex of the rat: a comparative review. *Brain Res. Rev.* **8,** 65–98.

17. Uylings H. B. M. and van Eden C. G. (1990) Qualitative and quantitative comparison of the prefrontal cortex in rat and in primates, including humans. *Prog. Brain Res.* **85,** 31–61.

18. Conde F., Marie-Lepoivre E., Audinat E., and Crepel F. (1995) Afferent connections of the medial frontal cortex of the rat. II. Cortical and subcortical afferents. *J. Comp. Neurol.* **352,** 567–593.

19. Berger B. S., Trottier C., Verney P., Gaspar P., and Alvarez C. (1988) Regional and laminar distribution of the dopamine and serotonin innervation in the macaque cerebral cortex: A radioautographic study. *J. Comp. Neurol.* **273,** 99–119.

20. Lewis D. A., Campbell M. J., Foote S. L., and Morrison J. H. (1986) The monoaminergic innervation of primate neocortex. *Hum. Neurobiol.* **5,** 181–186.

21. Miller E. K. (1999) The prefrontal cortex: complex neural properties for complex behavior. *Neuron* **22,** 15–17.

22. Goldman-Rakic P. S. (1995) Cellular basis of working memory. *Neuron* **14,** 477–485.

23. Goldman-Rakic P. S. (1999) The "psychic" neuron of the cerebral cortex. *Ann. NY Acad. Sci.* **868,** 13–26.

24. Somogyi P., Kisvardy Z. F., Martin K. A. C., and Whitteridge D. (1983) Synaptic connections of morphologically identified and physiologically characterized basket cells in the striate cortex of the cat. *Neuroscience* **10,** 261–294.

25. Constantinidis C., Williams G. V., and Goldman-Rakic P. S. (2002) A role for inhibition in shaping the temporal flow of information in prefrontal cortex. *Nature Neuroscience* **5,** 175–180.

26. Smiley J. F. and Goldman-Rakic P. S. (1996) Serotonergic axons in monkey prefrontal cerebral cortex synapse predominantly on interneurons as demonstrated by serial section electron microscopy. *J. Comp. Neurol.* **367,** 431–434.

27. Bunin M. A. and Wightman, R. M. (1999) Paracrine neurotransmission in the CNS: involvement of 5-HT. *Trends Neurosci.* **22,** 377–382.

28. Sumiyoshi T., Stockmeier C. A., Overholser J. C., Dilley G. E., Meltzer H. Y. (1996) Serotonin 1A receptors are increased in postmortem prefrontal cortex in schizophrenia. *Brain Res* **708,** 209–214.

29. Gurevich E. V. and Joyce J. N. (1997) Alterations in the cortical serotonergic system in schizophrenia: a postmortem study. *Biol. Psychiatry* **42,** 529–545.

30. Dean B., Hussain T., Hayes W., Scarr E., Kitsoulis S., Hill C., Opeskin K., and Copolov D. L. (1999) Changes in serotonin2A and GABA(A) receptors in schizophrenia: studies on the human dorsolateral prefrontal cortex. *J. Neurochem.* **72,** 1593–1599.

31. Meyer J. H., Kapur S., Houle S., DaSilva J., Owczarek B., Brown G. M., Wilson A. A., and Kennedy S. H. (1999) Prefrontal cortex 5-HT2 receptors in depression: an [18F]setoperone PET imaging study. *Am. J. Psychiatry* **156,** 1029–1034.

32. Martin G. R., Eglen R. M., Hamblin M. W., Hoyer D., and Yocca F. (1998) The structure and signalling properties of 5-HT receptors: an endless diversity? *Trends Pharmacol. Sci.* **19,** 2–4.

33. Andrade R. (1998) Regulation of membrane excitability in the central nervous system by serotonin receptor subtypes. *Ann. NY Acad. Sci.* **861,** 190–203.

34. Tecott L. H., Sun L. M., Akana S. F., Strack A. M., Lowenstein D. H., Dallman M. F., and Julius D. (1995) Eating disorder and epilepsy in mice lacking 5-HT2c serotonin receptors. *Nature* **374,** 542–546.

35. Saudou F., Amara D. A., Dierich A., LeMeur M., Ramboz S., Segu L., Buhot M. C., and Hen R. (1994) Enhanced aggressive behavior in mice lacking 5-HT1B receptor. *Science* **265,** 1875–1878.

36. Heisler L. K., Chu H. M., Brennan T. J., Danao J. A., Bajwa P., Parsons L. H., and Tecott L. H. (1998) Elevated anxiety and antidepressant-like responses in serotonin 5-HT1A receptor mutant mice. *Proc. Natl. Acad. Sci. USA* **95,** 15,049–15,054.

37. Macdonald R. L. and Olsen R. W. (1994) GABAA receptor channels. *Annu. Rev. Neurosci.* **17,** 569–602.

38. Benes, F. M., Vincent, S. L., Marie, A., and Khan Y. (1996) Up-regulation of GABAA receptor binding on neurons of the prefrontal cortex in schizophrenic subjects. *Neuroscience* **75,** 1021–1031.

39. Ohnuma T., Augood S. J., Arai H., McKenna P. J., and Emson P. C. (1999) Measurement of GABAergic parameters in the prefrontal cortex in schizophrenia: focus on GABA content, GABA(A) receptor alpha-1 subunit messenger RNA and human GABA transporter-1 (HGAT-1) messenger RNA expression. *Neuroscience* **93,** 441–448.

40. Lewis D. A. (2000) GABAergic local circuit neurons and prefrontal cortical dysfunction in schizophrenia. *Brain Res. Brain Res. Rev.* **31,** 270–276.

41. Dean B. (2001) A predicted cortical serotonergic/cholinergic/GABAergic interface as a site of pathology in schizophrenia. *Clin. Exp. Pharmacol. Physiol.* **28,** 74–78.

42. Moss S. J., Doherty C. A., and Huganir R. L. (1992a) Identification of the cAMP-dependent protein kinase and protein kinase C phosphorylation sites within the major intracellular domains of the beta 1, gamma 2S, and gamma 2L subunits of the gamma-aminobutyric acid type A receptor. *J. Biol. Chem.* **267,** 14,470–14,476.

43. Moss S. J., Smart T. G., Blackstone C. D., and Huganir R. L. (1992b) Functional modulation of GABA$_A$ receptors by cAMP-dependent protein phosphorylation. *Science* **257,** 661–665.

44. Porter N. M., Twyman R. E., Uhler M. D., and Macdonald R. L. (1990) Cyclic AMP-dependent protein kinase decreases GABAA receptor current in mouse spinal neurons. *Neuron* **5,** 789–796.

45. Krishek B. J., Xie X., Blackstone C., Huganir R. L., Moss S. J., and Smart T. G. (1994) Regulation of GABA(A) receptor function by protein kinase C phosphorylation. *Neuron* **12,** 1081–1095.

46. Yan Z. and Surmeier D. J. (1997) D5 dopamine receptors enhance Zn2+–sensitive GABA(A) currents in striatal cholinergic interneurons through a PKA/PP1 cascade. *Neuron* **19,** 1115–1126.

47. Goldman-Rakic P. S., Lidow M. S., and Gallager D. W. (1990) Overlap of dopaminergic, adrenergic, and serotoninergic receptors and complementarity of their subtypes in primate prefrontal cortex. *J. Neurosci.* **10,** 2125–2138.

48. Feng J., Cai X., Zhao J. H., and Yan Z. (2001) Serotonin Receptors Modulate GABA$_A$ Receptor Channels through Activation of Anchored Protein Kinase C in Prefrontal Cortical Neurons. *J. Neurosci.* **21,** 6502–6511.

49. Jakab R. L. and Goldman-Rakic P. S. (1998) 5-hydroxytryptamine2A serotonin receptors in the primate cerebral cortex: possible site of action of hallucinogenic and antipsychotic drugs in pyramidal cell apical dendrites. *Proc. Natl. Acad. Sci. USA* **95,** 735–740.

50. Nusser Z., Sieghart W., Benke D., Fritschy J. M., and Somogyi P. (1996) Differential synaptic localization of two major gamma-aminobutyric acid type A receptor alpha subunits on hippocampal pyramidal cells. *Proc. Natl. Acad. Sci. USA* **93,** 11,939–11,944.

51. Kellenberger S., Malherbe P., and Sigel E. (1992) Function of the alpha 1 beta 2 gamma 2S gamma-aminobutyric acid type A receptor is modulated by protein kinase C via multiple phosphorylation sites. *J. Biol. Chem.* **267,** 25,660–25,663.

52. Pawson T. and Scott J. D. (1997) Signaling through scaffold, anchoring, and adapter proteins. *Science* **278,** 2075–2080.

53. Yan Z., Hsieh-Wilson L., Feng J., Tomizawa K., Allen P. B., Fienberg A. A., Nairn A. C., and Greengard P. (1999) Protein phosphatase 1 modulation of neostriatal AMPA channels: regulation by DARPP-32 and spinophilin. *Nat. Neurosci* **2,** 13–17.

54. Feng J., Yan Z., Ferreira A. B., Tomizawa K., Liauw J. A., Zhou M., Allen P. B., Ouimet C. C., and Greengard P. (2000) Regulation of the Formation and Function of Dendritic Spines by Spinophilin. *Proc. Natl. Acad. Sci. USA* **97,** 9287–9292.

55. Tanaka C. and Nishizuka Y. (1994) The protein kinase C family for neuronal signaling. *Annu. Rev. Neurosci.* **17,** 551–557.

56. Mochly-Rosen D. (1995) Compartmentalization of protein kinases by intracellular receptor proteins: a theme in signal transduction. *Science* **268,** 247–251.

57. Mochly-Rosen D. and Gordon A. S. (1998) Anchoring proteins for protein kinase C: a means for isozyme selectivity. *FASEB J.* **12,** 35–42.

58. Mochyl-Rosen D., Khaner H., and Lopez J. (1991) Identification of intracellular receptor proteins for activated protein kinase *C. Proc. Natl. Acad. Sci. USA* **88,** 3997–4000.

59. Ron D., Chen C. H., Caldwell J., Jamieson L., Orr E., and Mochly-Rosen D. (1994) Cloning of an intracellular receptor for protein kinase C: a homolog of the beta subunit of G proteins. *Proc. Natl. Acad. Sci. USA* **91,** 839–843.

60. Ron D., Luo J., and Mochly-Rosen D. (1995) C2 region-derived peptides inhibit translocation and function of beta protein kinase C in vivo. *J. Biol. Chem.* **270,** 24,180–24,187.

61. Battani F., Pascale A., Paoletti R., and Govoni S. (1997) The role of anchoring protein RACK1 in PKC activation in the aging rat brain. *Trends Neurosci.* **20,** 410–415.

62. Brandon N. J., Uren J. M., Kittler J. T., Wang H., Olsen R., Parker P. J., and Moss S. J. (1999) Subunit-specific association of protein kinase C and the receptor for activated C kinase with GABA type A receptors. *J. Neurosci.* **19,** 9228–9234.

63. Klauck T. M., Faux M. C., Labudda K., Langeberg L. K., Jaken S., and Scott J. D. (1996) Coordination of three signaling enzymes by AKAP79, a mammalian scaffold protein. *Science* **271,** 1589–1592.

64. Zhou F. M. and Hablitz J. J. (1999) Activation of serotonin receptors modulates synaptic transmission in rat cerebral cortex. *J. Neurophysiol.* **82,** 2989–2999.

65. North R. A. and Uchimura N. (1989) 5-Hydroxytryptamine acts at 5-HT2 receptors to decrease potassium conductance in rat nucleus accumbens neurones. *J. Physiol.* **417,** 1–12.

66. Tanaka E. and North R. A. (1993) Actions of 5-hydroxytryptamine on neurons of the rat cignulate cortex. *J. Neurophysiol.* **69,** 1749–1757.

67. Araneda R. and Andrade R. A. (1991) 5-HT2 and 5-HT1A receptors mediate opposing responses on membrane excitability in rat association cortex. *Neuroscience* **40,** 399–412.

68. Haj-Dahmane S. and Andrade R. (1996) Muscarinic activation of a voltage-dependent cation nonselective current in rat association cortex. *J. Neurosci.* **16,** 3848–3861.

69. Domenech T., Beleta J., Fernandez A. G., Gristwood R. W., Cruz Sanchez F., Tolosa E., and Palacios J. M. (1994) Identification and charac-

terization of serotonin 5-HT4 receptor binding sites in human brain: comparison with other mammalian species. *Brain Res. Mol. Brain Res.* **21,** 176–180.

70. Eglen R. M., Wong E. H. F., Dumuis A., and Bockaert J. (1995) Central 5-HT$_4$ receptors. *TIPS* **16,** 391–398.

71. Costall B. and Naylor R. J. (1993) The pharmacology of the 5-HT4 receptor. *Int. Clin. Psychopharmacol.* **8 (Suppl 2),** 11–18.

72. Reynolds G. P., Mason S. L., Meldrum A., De Keczer S., Parnes H., Eglen R. M., and Wong E. H. (1995) 5-Hydroxytryptamine (5-HT)4 receptors in post mortem human brain tissue: distribution, pharmacology and effects of neurodegenerative diseases. *Br. J. Pharmacol.* **114,** 993–998.

73. Torres G. E., Chaput Y., and Andrade R. (1995) Cyclic AMP and protein kinase A mediate 5-hydroxytryptamine type 4 receptor regulation of calcium-activated potassium current in adult hippocampal neurons. *Mol. Pharmacol.* **47,** 191–197.

74. Bobker D. H. and Williams J. T. (1989) Serotonin augments the cationic current I$_h$ in central neurons. *Neuron* **2,** 1535–1540.

75. Cardenas C. G., Del Mar L. P., Cooper B. Y., and Scroggs R. S. (1997) 5HT4 receptors couple positively to tetrodotoxin-insensitive sodium channels in a subpopulation of capsaicin-sensitive rat sensory neurons. *J. Neurosci.* **17,** 7181–7189.

76. Ansanay H., Dumuis A., Sebben M., Bockaert J., and Fagni L. (1995) cAMP-dependent, long-lasting inhibition of a K+ current in mammalian neurons. *Proc. Natl. Acad. Sci. USA* **92,** 6635–6639.

77. Cai X., Flores-Hernandez J., Feng J., and Yan Z. (2002) Activity-dependent Bi-directional Regulation of GABA$_A$ Receptor Channels by Serotonin 5-HT$_4$ Receptors in Pyramidal Neurons of the Prefrontal Cortex. *J. Physiology* (London) (in press, published online on March 15, 2002).

78. Rosenmund C., Carr D. W., Bergeson S. E., Nilaver G., Scott J. D., and Westbrook G. L. (1994) Anchoring of protein kinase A is required for modulation of AMPA/kainate receptors on hippocampal neurons. *Nature* **368,** 853–856.

79. Gao T., Yatani A., Dell'Acqua M. L., Sako H., Green S. A., Dascal N., Scott J. D., and Hosey M. M. (1997) cAMP-dependent regulation of cardiac L-type Ca^{2+} channels requires membrane targeting of PKA and phosphorylation of channel subunits. *Neuron* **19,** 185–196.

80. Colledge M. and Scott J. D. (1999) AKAPs: from structure to function. *Trends Cell Biol.* **9,** 216–221.

81. Kapur J. and Macdonald R. L. (1996) Cyclic AMP-dependent protein kinase enhances hippocampal denate granule cell GABAA receptor currents. *J. Neurophysiol.* **76,** 2626–2634.

82. McDonald B. J., Amato A., Connolly C. N., Benke D., Moss S. J., and Smart T. G. (1998) Adjacent phosphorylation sites on GABAA receptor beta subunits determine regulation by cAMP-dependent protein kinase. *Nat. Neurosci.* **1,** 23–28.

83. Nicola S. M., Surmeier J., and Malenka R. C. (2000) Dopaminergic modulation of neuronal excitability in the striatum and nucleus accumbens. *Annu. Rev. Neurosci.* **23,** 185–215.

84. Kia H. K., Miquel M. C., Brisorgueil M. J., Daval G., Riad M., El Mestikawy S., Hamon M., and Verge D. (1996a) Immunocytochemical localization of serotonin1A receptors in the rat central nervous system. *J. Comp. Neurol.* **365,** 289–305.

85. Kia H. K., Brisorgueil M. J., Hamon M., Calas A., and Verge D. (1996b) Ultrastructural localization of 5-hydroxytryptamine1A receptors in the rat brain. *J. Neurosci. Res.* **46,** 697–708.

86. Schreiber R., and De Vry J. (1993) 5-HT1A receptor ligands in animal models of anxiety, impulsivity and depression: multiple mechanisms of action? *Prog. Neuropsychopharmacol. Biol. Psychiatry* **17,** 87–104.

87. Olivier B., Soudijn W., and van Wijngaarden I. (1999) The 5-HT1A receptor and its ligands: structure and function. *Prog. Drug. Res.* **52,** 103–165.

88. Bantick R. A., Deakin J. F., and Grasby P. M. (2001) The 5-HT1A receptor in schizophrenia: a promising target for novel atypical neuroleptics? *J. Psychopharmacol.* **15,** 37–46.

89. Carli M., Luschi R., Garofalo P., and Samanin R. (1995) 8-OH-DPAT impairs spatial but not visual learning in a water maze by stimulating 5-HT1A receptors in the hippocampus. *Behav. Brain Res.* **67,** 67–74.

90. Andrade R., Malenka R. C., and Nicoll R. A. (1986) A G protein couples serotonin and GABAB receptors to the same channels in hippocampus. *Science* **234,** 1261–1265.

91. Colino A. and Halliwell J. V. (1987) Differential modulation of three separate K-conductances

in hippocampal CA1 neurons by serotonin. *Nature* **328,** 73–77.

92. Penington N. J. and Kelly J. S. (1990) Serotonin receptor activation reduces calcium current in an acutely dissociated adult central neuron. *Neuron* **4,** 751–758.

93. Foehring R. C. (1996) Serotonin modulates N- and P-type calcium currents in neocortical pyramidal neurons via a membrane-delimited pathway. *J. Neurophysiol.* **75,** 648–659.

94. Schmitz D., Empson R. M., and Heinemann U. (1995) Serotonin reduces inhibition via 5-HT1A receptors in area CA1 of rat hippocampal slices in vitro. *J. Neurosci.* **15,** 7217–7225.

95. Boschert U., Amara D. A., Segu L., and Hen R. (1994) The mouse 5-hydroxytryptamine1B receptor is localized predominantly on axon terminals. *Neuroscience* **58,** 167–182.

96. Standford I. M. and Lacey M. G. (1996) Differential actions of serotonin, mediated by 5-HT1B and 5-HT2C receptors, on GABA-mediated synaptic input to rat substantia nigra pars reticulata neurons in vitro. *J Neurosci.* **16,** 7566–7573.

97. Derkach V., Surprenant A., and North R. A. (1989) 5-HT3 receptors are membrane ion channels. *Nature* **339,** 706–790.

98. Jackson M. B. and Yakel J. L. (1995) The 5-HT3 receptor channel. *Annu. Rev. Physiol.* **57,** 447–468.

99. McMohan L. L. and Kauer J. A. (1997) Hippocampal interneurons are excited via serotonin-gated ion channels. *J. Neurophysiol.* **78,** 2493–2502.

100. Yang J., Mathie A., and Hille B. (1992) 5-HT3 receptor channels in dissociated rat superior cervical ganglion neurons. *J. Physiol.* **448,** 237–256.

101. Morales M. and Bloom F. E. (1997) The 5-HT3 receptor is present in different subpopulations of GABAergic neurons in the rat telencephalon. *J. Neurosci.* **17,** 3157–3167.

102. Ropert N. and Guy N. (1991) Serotonin facilitates GABAergic transmission in the CAI region of rat hippocampus in vitro. *J. Physiol.* **441,** 121–136.

103. Ashby C. R. Jr, Minabe Y., Edwards E., and Wang R. Y. (1991) 5-HT3-like receptors in the rat medial prefrontal cortex: an electrophysiological study. *Brain Res.* **550,** 181–191.

104. Wang C. and Zucker R. S. (1998) Regulation of synaptic vesicle recycling by calcium and serotonin. *Neuron* **21,** 155–167.

105. Jacobs B. L. and Azmitia E. C. (1992) Structure and function of the brain serotonin system. *Physiol. Rev.* **72,** 165–229.

106. Reader T. A. and Jasper H. H. (1984) Interactions between monoamines and other transmitters in cerebral cortex, in *Monoamine Innervation of Cerebral Cortex* (edited by L. Descarries, T. R. Reader, and H. H. Jasper). Alan R. Liss New York, NY, pp. 195–225.

107. Aghajanian G. K. and Marek G. J. (2000) Serotonin model of schizophrenia: emerging role of glutamate mechanisms. *Brain Res. Brain Res. Rev.* **31,** 302–312.

108. Martin-Ruiz R., Puig M. V., Celada P., Shapiro D. A., Roth B. L., Mengod G., and Artigas F. (2001) Control of serotonergic function in medial prefrontal cortex by serotonin-2A receptors through a glutamate-dependent mechanism. *J. Neurosci.* **21,** 9856–9866.