GABAergic Deficits in Schizophrenia: Evidence and Implications

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Abstract

Many biochemical theories have attempted to explain the pathophysiology of schizophrenia. The dopamine hypothesis is one of oldest and most popular biochemical theories, but it is too simplistic to account for all aspects of the illness. Other neurotransmitter systems, including glutamate, serotonin and γ-aminobutyric acid (GABA), have been implicated in schizophrenia. The role of the GABA system in schizophrenia has been investigated, and research to date suggests that GABAergic deficits contribute to both the pathophysiology and symptomatology of schizophrenia. In this review GABA neurotransmission in the central nervous system (CNS) is described, evidence for cortical and hippocampal GABAergic deficits in schizophrenia is presented, and inferences about how these deficits may contribute to schizophrenia symptomatology are made.

Key words: schizophrenia, GABA, chandelier cells, basket cells, prefrontal cortex, hippocampus.

GABA Neurotransmission In The Central Nervous System

In order to understand the consequences of γ-aminobutyric acid (GABA) abnormalities in schizophrenia, it is necessary to understand the normal structure and function of GABAergic neurons. There are many different types of GABAergic neurons, which are widely distributed throughout the CNS, including the cortex and hippocampus. The structure and function of GABAergic, nonpyramidal neurons is reviewed below.

GABAergic interneurons (i.e. nonpyramidal neurons that modulate the activity of pyramidal/principal cells) can be categorized according to the types of synaptic contacts they form with other neurons. Basket cells, which play a role in the pathology of schizophrenia [1], form axo-somatic contacts with their target neurons and are the most common type of interneurons in the CNS. They are called “basket” cells because the terminal boutons of these neurons form a basket-like arrangement around a large proportion of pyramidal neuronal cell bodies. The axon terminals of basket cells are positioned at proximal portions of apical dendrites of pyramidal neurons. Since input at the level of the cell body alters membrane potential to a greater extent than input at distal points along the dendritic tree, the inhibitory effects of basket cells are very potent. As basket cells are directly innervated by thalamic neurons, they control the amount of pyramidal neuronal activity of pyramidal/principal cells) can be categorized according to the types of synaptic contacts they form with other neurons. Basket cells, which play a role in the pathology of schizophrenia [1], form axo-somatic contacts with their target neurons and are the most common type of interneurons in the CNS. They are called “basket” cells because the terminal boutons of these neurons form a basket-like arrangement around a large proportion of pyramidal neuronal cell bodies. The axon terminals of basket cells are positioned at proximal portions of apical dendrites of pyramidal neurons. Since input at the level of the cell body alters membrane potential to a greater extent than input at distal points along the dendritic tree, the inhibitory effects of basket cells are very potent. As basket cells are directly innervated by thalamic neurons, they control the amount of pyramidal neuronal activity of pyramidal neurones [3]. Since action potentials (APs) are generated at the AIS, chandelier cells can short-circuit AP propagation, and therefore play a major role in regulating the output activity of pyramidal neurons [3]. Thus, these neurons are involved in modulating stimulus-response properties of their target neurons [4]. Unlike basket cells, chandelier cells are not directly contacted by thalamic afferent fibers. GABAergic neurons can also be classified according to the type of calcium binding protein (CBP) they express. During neurodegeneration calcium homeostasis is altered. In particular, intracellular calcium levels rise, leading to detrimental effects that ultimately result in cell death. CBPs are neuroprotective because they can bind to intracellular calcium, and restore impaired calcium homeostasis that occurs during cellular degeneration. Basket cells and chandelier cells both express the CBP parvalbumin (PVB), which is not expressed in any other types of GABAergic neurons. Some also express calbindin (CB), but not exclusively since double bouquet cells also express this CBP. In contrast to the CBPs calretinin (CR) and CB, which are expressed in double bouquet neurons, PVB is expressed late during development (i.e. 3-6 months after birth, after the formation of synaptic contacts) [5]. Furthermore, there is little change in CR-expressing cells during infancy, but substantial CB- and PVB-containing cell reorganization occurs during this time period [5]. PVB-containing neurons are fast spiking: they can fire repeatedly when activated, have short AP durations and after-hyperpolarizations, have relatively negative resting potentials, relatively low input threshold, and are connected to each other via excitatory electrical synapses (gap-junctions) and inhibitory chemical synapses. PVB-containing neurons are innervated by CR-containing GABAergic neurons, via electric and chemical synaptic contacts. GABAergic innervations can be intrinsic.
or extrinsic, originating from either other cortical areas or from other areas of the brain, respectively [1].

The function of most inhibitory interneurons is to mediate feedback or feed-forward inhibition of pyramidal/principal neurons, and thus stabilize the activity of these neurons. In feedback inhibition, a pyramidal neuron, which is activated by excitatory inputs from another source, activates inhibitory interneurons, which in turn inhibit principal neurons, including the one that activated them. In feed-forward inhibition, interneurons are activated by an afferent, excitatory input from another source, which also activates a downstream target. In turn, the activated interneurons inhibit downstream targets of the neuron that activated them. An example of feed-forward inhibition occurs in the hippocampus which is composed of 3 sectors (CA1, CA2 and CA3). Information proceeds from the CA3 sector to the CA1 sector of the. Pyramidal cells of the CA3 sector of the hippocampus, which project downstream to other pyramidal cells in the CA1 sector, also project to GABAergic interneurons in the CA1 sector. These interneurons in turn inhibit pyramidal cells in the CA1 sector [1].

There are two types of GABA receptors: GABA_A and GABA_B. GABA_A receptors are transmembrane ligand-gated ion channels that are permeable to chloride. When GABA binds to a GABA_A receptor, the transmembrane anion channel opens, allowing chloride to enter and usually causing hyperpolarization/inhibition of the cell [6]. GABA_A receptors are composed of five subunits (eg. 2 α subunits, 2 β subunits and 1 γ subunit, where each subunit exists in multiple isoforms). GABA_A receptors in different brain regions are composed of different subunits, and subunit composition determines the physiological and pharmacological properties of the receptor [7]. For example, activation and deactivation rates of GABA_A receptor channels are dependent on the type of α subunit isoform expressed, where α2 subunits appear to have higher affinity for GABA, with faster activation and slower deactivation times than the more common α1 subunits [8]. This is of interest because α2 subunits are prominently localized on AISs of pyramidal neurons, the postsynaptic targets of chandelier cells. Thus the presence of α2 on AISs allows chandelier cells to more potently regulate the output activity of pyramidal neurons [8]. GABA_A receptors also have binding sites for compounds other than GABA, including benzodiazepines, barbiturates, and neuroactive steroids, which allosterically modulate GABA_A receptors in the presence of GABA, and in some cases directly induce GABA_A receptor channel activation [6].

GABA_A receptors are G-protein coupled receptors that are present on postsynaptic membranes, and mediate slow inhibitory postsynaptic potentials by opening potassium channels. GABA_A receptors are also localized on postsynaptic and presynaptic neuron terminals, in which case they inhibit neurotransmitter release by closing calcium channels [6].

Reuptake is the primary mode of GABA signal termination. There are at least four different GABA transporters (GAT-1, GAT-2, GAT-3 and BGT-1) that are responsible for GABA re-uptake from the synaptic cleft [9]. Cells expressing GATs, including GABAergic and non-GABAergic neurons, as well as glia, can take up GABA, and thus terminate GABA effects [10].

### Presynaptic GABA Neurotransmission Changes In Schizophrenia

Roberts [11] was the first to postulate that a defect in the GABA system may play a role in the pathology of schizophrenia. An overview of postmortem studies providing evidence for GABAergic deficits in schizophrenia follows.

The rate limiting GABA-synthesizing enzyme glutamic acid decarboxylase (GAD), which exists in both 65 kD (GAD_65) and 67 kD (GAD_67) forms, catalyzes the conversion of glutamate into GABA, and is a marker of GABAergic neurons [7]. Multiple postmortem studies have found low GABA levels in the amygdala of patients with schizophrenia, when compared to healthy individuals [12]. Perry and coworkers [13] also found decreased GABA concentrations in the nucleus accumbens (NAC) and the thalamus of patients with schizophrenia. Reduced GABA concentrations may reflect altered GAD activity, which is also reduced in patients with schizophrenia, particularly in the NAc, amygdala, hippocampus, and putamen [14]. In recently conducted postmortem studies, Volk and coworkers [15] found decreased GAD_67 messenger RNA (mRNA)-positive interneurons in layers III-V of the prefrontal cortex (PFC), while Heckers and coworkers [16] found decreased GAD_65 mRNA, and to a lesser extent decreased GAD_67 mRNA expression in the sectors CA2/3 of the hippocampus, and in the dentate gyrus in patients with schizophrenia. Decreased GAD mRNA expression is consistent with findings of reduced GAD activity and GABA concentrations in schizophrenia.

In contrast to the findings of other researchers, Gluck and coworkers [17] found that GAD activity was increased in the dorsolateral PFC in patients with schizophrenia. This inconsistent finding may be the result of different brain tissue collection and/or the brains analyzed in this study. Alternatively increased GAD activity may be a compensatory upregulation of GABA synthesis in response to reduced GABA levels described above.

Findings from the following studies strongly suggest that reduced GAD mRNA expression and activity is not the result of antipsychotic treatment, but rather is a part of the disease process. It has been found that haloperidol has no effect on GAD_65 mRNA expression in layers III-V of the PFC in monkeys [15], and chronic haloperidol, sertindole or olanzapine treatment actually increased levels of GAD_65 in the ventral and dorsal striatum, thalamus and entorhinal cortex in rats [18]. Therefore, it appears that antipsychotic treatment has no effect on, or may even reverse, decreased GAD expression seen in schizophrenia. On the other hand, several days of amphetamine treatment, which mimics the hyperdopaminergic state and some symptoms of schizophrenia, decreases extracellular GABA and intracellular GAD_65 levels in the rat NAc [19]. Furthermore chronic administration of quinpirole, a D_2 receptor agonist, decreases GAD_65 mRNA expression in rat dorsal and ventral striatum [20]. It thus appears that administration of compounds that mimic some physiological abnormalities of schizophrenia cause alterations in GAD consistent with those found in schizophrenia. Therefore, evidence to date suggests that reduced GAD mRNA
expression and GAD activity are due to pathological mechanisms rather than to the effects of drugs used in the treatment of schizophrenia.

GABA reuptake also appears to be altered in schizophrenia. Simpson and coworkers [21] found a decrease in GABA uptake sites in the amygdala, hippocampus, and the left side of the temporal cortex in a postmortem study conducted on brains of patients with schizophrenia. Similarly, Volk and coworkers [22] discovered a decrease in GAT-1 mRNA expression in layers I-V of the PFC in patients with schizophrenia, when compared to healthy control subjects. A reduction in GABA reuptake would result in increased GABA concentrations in the synapse, which may be a compensatory mechanism to reduced GABA levels found in schizophrenia patients. The researchers also found that GAT-1 mRNA expression was not altered in monkey PFC following chronic treatment with haloperidol, implying that the decreased GAT-1 mRNA expression and GABA uptake sites are not the result of antipsychotic treatment, but rather are part of the disease process.

Besides altered GABA synthesis and reuptake, GABAergic neuronal cell density is also decreased in schizophrenia, particularly in the cortex and the limbic system. In a postmortem study Benes and coworkers [23] found decreased nonpyramidal neuronal density in layers II-VI of the anterior cingulate cortex and layer II of the PFC in patients with schizophrenia, when compared to healthy controls. More recently, Benes and coworkers [24] found decreased nonpyramidal neuronal density in sector CA2 of the hippocampus. This decreased cell density is apparently not attributable to treatment with antipsychotic medication, since chronic haloperidol administration increases GABA-immunoreactive terminals in the rat medial PFC (as determined by immunohistochemistry using specific antibodies) [25], and patients treated with antipsychotics for the shortest duration of time had the greatest reduction in chandelier axonal terminals and GAD-immunoreactivity in the hippocampus [26].

Chandelier cells and basket cells exhibit the greatest deficiencies in schizophrenia. In a postmortem study, the density of GAT-1 immunoreactive chandelier axonal cartridges were decreased in layers II-IV of the PFC in patients with schizophrenia, when compared to healthy volunteers [27]. Similarly, Hashimoto and coworkers [28] found reduced PVB mRNA expression in layers II and IV of the PFC in patients with schizophrenia. Other PVB neuronal cell deficits in schizophrenia include decreased PVB-immunoreactive neuronal density in layer II of the entorhinal cortex [29] and all regions of the hippocampus [30], and decreased numbers of PVB- and CB-immunoreactive neurons in the PFC with no change in CR-immunoreactivity [29, 31, 32]. Since PVB is expressed exclusively in chandelier and basket cells, it appears that deficits seen in schizophrenia are restricted to these GABAergic cell types. It is unlikely that a reduced CB-immunoreactivity reflects a deficit in double bouquet cells because CL-immunoreactivity, a marker for double bouquet cells, is not altered in schizophrenia. Instead, altered CB-immunoreactivity reflects deficits in chandelier and basket cells because some of these GABAergic neurons express CB.

PVB-containing neuronal cell deficit is a potential marker for schizophrenia with a genetic etiology. PVB-immunoreactive neuronal deficits are most profound in patients who do not have ventricular enlargement [5], which is a structural abnormality primarily found in patients with schizophrenia who do not have a family history of the illness [33]. Future research may ascertain PVB-containing cell loss as a marker to distinguish between genetically inherited and otherwise acquired schizophrenia.

PVB-containing neuronal loss also supports the neurodevelopmental hypothesis of schizophrenia, which postulates that a disruption of brain development early in life underlies late emergence of symptoms during early adulthood [34]. As mentioned earlier, PVB, which is neuroprotective, is expressed late during development. The late expression of PVB can potentially leave a window of vulnerability during early development, prior to the expression of PVB, where a subtle neurotoxic challenge can result in neuronal loss that may later contribute to the emergence of clinical symptoms during adolescence [5].

### Postsynaptic GABA Neurotransmission Changes in Schizophrenia

In schizophrenia there are changes in postsynaptic targets of GABAergic neurons, including pyramidal and nonpyramidal neurons, in response to the presynaptic changes discussed above. Several postmortem studies using bicuculline as a selective GABA antagonist, reveal increased [3H]-muscimol binding to nonpyramidal cells in sector CA3, and to pyramidal cells in sector CA1 of the hippocampal formation [35], in layers II, III, V and VI of the PFC [36], and layers II and III of the anterior cingulate cortex [37] in patients with schizophrenia (compared to healthy controls). Increased muscimol binding implies an upregulation of postsynaptic GABA<sub>B</sub> binding sites. Increased GABA<sub>A</sub> receptor density is a compensatory mechanism for presynaptic GABAergic deficiencies. Upregulation of GABA<sub>A</sub> receptor density reported in these studies was not correlated with increased benzodiazepine binding sites, suggesting that an uncoupling in the regulation of GABA<sub>A</sub> receptor complex components also may also occur in schizophrenia.

In order to study postsynaptic consequences of chandelier cell deficit in schizophrenia, Volk and coworkers [38] examined postmortem brain tissue to detect changes in AISs of pyramidal neurons. They measured the density of α<sub>2</sub> GABA<sub>A</sub> receptor subunit that is predominantly localized at AISs of pyramidal neurons, and found that patients with schizophrenia have increased α<sub>2</sub> subunit density at AISs (α<sub>2</sub>-AIS density), reflecting an increase in GABA<sub>A</sub> receptor density at chandelier cell targets. The researchers also found an inverse relationship between α<sub>2</sub>-AIS density and GAT-1 cartridge density, indicating that presynaptic alterations in chandelier cells are associated with postsynaptic changes in GABA<sub>A</sub> receptors at AISs of pyramidal neurons in schizophrenia. This study further supports the proposal that chandelier cell deficit occurs in schizophrenia.

GABA<sub>A</sub> receptors are also altered in schizophrenia. GABA<sub>A</sub> receptor expression is decreased in layers II, III, and V of entorhinal and layer V of inferior temporal cortices in patients with schizophrenia [39]. Since GABA<sub>A</sub> receptor activation inhibits GABA release, reduced
GABA<sub>A</sub> receptor density may serve to compensate for decreased GABA levels found in postmortem schizophrenic brain tissue.

**Consequences Of GABA Dysfunction In Schizophrenia**

It is thought that individuals with schizophrenia are unable to filter out extraneous information, resulting in an over-inclusive thought pattern and difficulty concentrating [40]. This has led to the speculation that central sensory gating mechanisms are impaired, and that this effect may be the result of GABAergic hypofunction in schizophrenia [1].

Working memory dysfunction is a cognitive deficit in schizophrenia that is associated with prefrontal cortical abnormalities. Working memory allows an individual to retain an image based on recent sensory information, to integrate the image with cognitive and affective associations and to use the image to plan subsequent behavior [6]. Information processing, including the input of relevant sensory information and the formation of appropriate behavioral responses, is dependent on long-range connections between the PFC, the thalamus, and posterior cortical areas [41]. Pyramidal cells of the PFC receive sensory input from the thalamus, and play a critical role in processing the information prior sending it to appropriate brain regions.

Patients with schizophrenia perform poorly on tasks that require proper working memory function, such as the Wisconsin Card Sorting task [42], and fail to show normal activation of the dorsolateral PFC when performing this task [43]. Since prefrontal cortical inhibitory (GABAergic) interneurons play a role in regulating both the input and output of pyramidal neurons in the PFC, decreased GABAergic tone can increase pyramidal neuronal activity, resulting in working memory dysfunction.

Memory and affect dysfunction, reflecting altered circuitry of the hippocampus, are also symptoms of schizophrenia. Pyramidal neurons of the CA1 sector of the hippocampus receive sensory information from the entorhinal cortex via two inputs: a direct pathway and an indirect pathway through the dentate gyrus and the CA2/3 [44]. The hippocampus plays a crucial role in regulating both memory and affect by comparing the two inputs from the entorhinal cortex and sending appropriate information to the cortex and limbic structures, respectively [44]. GABAergic neurons in the CA1 sector of the hippocampus play an important role in feed-forward inhibition of hippocampal pyramidal neurons in the CA1 sector [1]. GABAergic hypofunction can result in over-stimulation of the cortical and limbic structures by CA1 pyramidal neurons, resulting in altered memory and affect regulation, respectively.

**Conclusion**

Postmortem studies conducted on patients with schizophrenia reveal that GABAergic hypofunction in the PFC and the hippocampus may be an important contributor to the pathophysiology and symptomatology of the illness. Both GABAergic neuronal density and GABA synthesis are decreased in schizophrenia. Compensatory up-regulation of GABA<sub>A</sub> receptors, down-regulation of GABA<sub>B</sub> receptors, and decreased GABA uptake is also evident. Poor memory and affect regulation, and altered working memory function are likely clinical manifestations of GABAergic hypofunction.

**References**


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