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The role of glutamate receptors in antipsychotic drug action

Review Article

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Summary. It has recently been postulated that disturbances in glutamatergic neurotransmission may contribute to the pathophysiology of schizophrenia. Therefore the aim of the present study was to evaluate the role of glutamate NMDA and group II metabotropic receptors in the antipsychotic drug action. To this aim the influence of some well-known neuroleptics on cortical NMDA receptors was examined. Furthermore, their behavioral effects were compared with those of the novel agonist of group II glutamate metabotropic receptors, LY 354740, in some animal models of schizophrenic deficits. We found that long-term administration of the typical neuroleptic haloperidol and the atypical one clozapine increased the number of NMDA receptors labelled with [^{3}H]CGP 39653 in different cortical areas. Long-, but not short-term, treatment with haloperidol and raclopride diminished the deficit of prepulse inhibition produced by phencyclidine, which is a model of sensorimotor gating deficit in schizophrenia. In contrast, neither short- nor long-term treatment with clozapine influenced the phencyclidine effect in that model. Acute treatment with LY 354740 reversed neither (1) the deficit of prepulse inhibition produced by phencyclidine or apomorphine, nor (2) the impairment in a delayed alternation task induced by MK-801, which is commonly used to model the frontal lobe deficits associated with schizophrenia. The present study suggests that an increase in the density of cortical NMDA receptors may be important to a longterm neuroleptic therapy. Conversely, the results do not support the role of group II metabotropic glutamate receptors in the antipsychotic drug action.

Keywords: Amino acids – NMDA receptors – Group II metabotropic glutamate receptors – Cerebral cortex – Neuroleptics – Prepulse inhibition – Delayed alternation task

Introduction

It is commonly accepted that the dopaminergic hyperactivity in subcortical structures underlies symptoms of schizophrenia. This concept is based mainly on the fact that the majority of known neuroleptics block dopamine D2 receptors, and that dopaminomimetics induce psychotic symptoms in man. However, the above-mentioned dopaminergic theory of schizophrenia does not explain all aspects of the disease.

It has recently been postulated that in the course of schizophrenia there develops — apart from a dopaminergic hyperactivity – a hypofunction of glutamatergic transmission (cf. Bunney et al., 1995). In support of this view, a reduced level of glutamate was found in the cerebrospinal fluid and in different cortical regions (frontal, parietal, temporal and occipital cortices), as well as in the hippocampus of schizophrenics (Kim et al., 1980; Toru et al., 1988, 1992; Tsai et al., 1995). The significance of the decreased cortical neurotransmission for the appearence of psychoses is substantiated by the fact that phencyclidine, an uncompetitive antagonist of NMDA receptors which binds inside an ion-channel, induces both positive and negative symptoms which are very similar to those seen in schizophrenia. Other uncompetitive and competitive antagonists of NMDA receptors have also been found to induce psychotic symptoms.

Several post-mortem studies also showed an increase in NMDA receptors in the frontal, parietal and temporal cortices in schizophrenics (Toru et al., 1988, 1992; Deakin et al., 1989; Ishimaru et al., 1992; Simpson et al., 1992). On this basis it has been suggested that the presynaptic hypofunction leads to a compensatory up-regulation of glutamatergic postsynaptic receptors.

The influence of neuroleptics on NMDA receptors

It is well known that most schizophrenics are treated with neuroleptics for many years, therefore it is not possible to exclude a possibility that the observed in these patients increases in NMDA receptors result not from the illness itself but from a neuroleptic therapy. In fact, it has recently been postulated that at least some neuroleptics influence directly the NMDA receptor complex. It has been suggested that they may act as partial agonists of a glycine or another modulatory site of the NMDA receptor complex (Fletcher and MacDonald, 1993; Lidsky et al., 1993; Banerjee et al., 1995; Ilyin et al., 1996; Lynch and Gallagher, 1996; Coughenour and Cordon, 1997; Lidsky et al., 1997). In *in vitro* experiments, haloperidol and clozapine given in micromolar concentrations were found to displace [^{3}H]MK-801, [^{125}I]MK-801, [^{3}H]ifenprodil or [^{3}H]TCP from the binding to NMDA receptors. Furthermore, Gandolfi and Dall'Olio (1996) found an increase in the [^{3}H]MK-801 binding to the ion-channel of the NMDA receptor complex in the prefrontal cortex, and Ułas et al. (1993) reported a similar increase in the [^{3}H]L-glutamate binding to the recognition site of the same receptor in the parietal cortex as a result of 3-week haloperidol administration to rats. However, other studies have not supported the above-cited data. McCoy and

Richfield (1996) and Tarazi et al. (1996) showed that chronic treatment with typical or atypical neuroleptics for 3–4 weeks decreased the binding of [^{3}H]MK-801 in the medial prefrontal or the frontal cortex. Moreover, according to Tarazi et al. (1996), 8-month treatment with either haloperidol or clozapine did not change the binding of [^{3}H]MK-801 in the same cortical region.

In our recent study (Ossowska et al., 1999) we evaluated the influence of a long-term treatment of rats with the typical neuroleptic haloperidol and the atypical one clozapine on cortical NMDA receptors. Both those neuroleptics were administered to rats in drinking water (haloperidol – 1 mg/kg/day; clozapine – 30 mg/kg/day) for a period of 3 months, and were afterwards withdrawn for 4 days. Such a long period of drug administration was chosen, because neuroleptics evoke actual antipsychotic effects not earlier than after a few weeks. Therefore certain adaptive changes, responsible for the therapeutic effect of these drugs, may be expected.

We examined the frontal, parietal and insular cortices. These cortices belong to regions which have been found to be affected in schizophrenia. NMDA receptors were labelled with [^{3}H]CGP 39653 – a competitive antagonist which binds to the recognition site of the NMDA receptor complex, and with [^{3}H]MK-801 which binds inside an ion-channel of this receptor. We measured densitometrically the binding of both those ligands in cortical slices using an autoradiographic method. A saturation study for estimating the density of NMDA receptors labelled with [^{3}H]CGP 39653 (B_{max}), as well as their affinity to that ligand (K_D) was carried out in parieto-insular homogenates.

An additional autoradiographic experiment evaluated the influence of a 3-month treatment with haloperidol or clozapine on dopamine D2 receptors labelled with [^{3}H]spiperone in the striatum in rats.

We found that the long-term treatment with haloperidol increased the binding of [^{3}H]CGP 39653 in the insular, frontal and parietal cortices of rats. Similarly, the long-term treatment with clozapine increased the binding of that compound in the insular and parietal cortices.

The saturation binding of [^{3}H]CGP 39653 revealed that both haloperidol and clozapine increased the density (B_{max}) of NMDA receptors in the insular and parietal cortices by 24–28%, and had no influence on the affinity of receptors to that ligand.

In contrast to their effect on the [^{3}H]CGP 39653 binding, neither haloperidol nor clozapine influenced the binding of [^{3}H]MK-801 in any cortical regions studied.

An autoradiographic study additionally showed that a long-term treatment with haloperidol, but not with clozapine, increased the binding of [^{3}H]spiperone to striatal dopamine D2 receptors.

The above-mentioned results allow us to draw the following conclusions:

(1) a 3-month treatment with haloperidol or clozapine increases the density of cortical NMDA receptors belonging to a subpopulation labelled with [^{3}H]CGP 39653;

(2) NMDA receptors labelled with [^{3}H]MK-801 are not affected by such a treatment;
(3) the effect of both these neuroleptics on cortical NMDA receptors seems to be independent of their action on dopamine D2 receptors.

The influence of neuroleptics on the phencyclidine-induced deficit of sensorimotor gating

A question arises whether the increased density of cortical NMDA receptors is of any significance to the antipsychotic action of neuroleptics. Such a question is difficult to answer due to the lack of proper animal models of psychoses. The majority of screening tests for potential neuroleptics are based on their antidopaminergic properties.

Recently, a model of prepulse inhibition has been developed. This model uses a startle, reflex reaction induced by a brief, strong acoustic or tactile stimulus (pulse). When such a strong stimulus is preceded by a weak, subthreshold stimulus (prepulse) given less than 1,000 ms earlier, the startle response is diminished. Prepulse inhibition is a physiological reaction in normal, healthy subjects, which reflects their ability to gate sensory information. Prepulse inhibition is disrupted in some schizophrenic patients (Geyer et al., 1990) and is diminished by dopaminomimetics e.g. apomorphine, and antagonists of NMDA receptors, including phencyclidine (Masbach et al., 1988; Mansbach and Geyer, 1989). The deficit of prepulse inhibition induced by dopaminomimetics has been reported to be inhibited by acute treatment with most neuroleptics, and is proposed to be a model of a sensorimotor or an attention impairment in schizophrenia (Rigdon and Viik, 1991; Swerdlow et al., 1991). In contrast, the phencyclidine-induced prepulse inhibition deficit is not counteracted by acute administration of classic neuroleptics. Only some atypical neuroleptics (remoxipride, seroquel, olanzapine) have been reported to be effective (Johansson et al., 1994; Bakshi and Geyer, 1995; Swerdlow et al., 1996). The data concerning clozapine are conflicting: its positive effect on the phencyclidine-induced deficit in the prepulse inhibition model seems to be related to a rat strain and is not dose-dependent (Bakshi et al., 1994; Johansson et al., 1994; Swerdlow et al., 1996).

In our study (Pietraszek and Ossowska, 1998) we examined the influence of a long-term treatment with haloperidol, clozapine and raclopride on the deficit of prepulse inhibition induced by phencyclidine. Haloperidol (1 mg/kg/day) and clozapine (30 mg/kg/day) were given to rats in drinking water for 4 days, 6 weeks and 3 months. Raclopride (2 mg/kg/day) was administered also in drinking water for 4 days and 6 weeks. Phencyclidine was injected sc in a dose of 5 mg/kg.

We found that a 6-week- to a 3-month treatment with haloperidol or raclopride diminished the disruption of prepulse inhibition induced by phencyclidine. A 4-day treatment appeared to be too short and was ineffective. In contrast, neither short- nor long-term treatment with clozapine influenced the phencyclidine effect in that model.

The above-mentioned study suggested that a long-term treatment with the classic neuroleptic haloperidol or raclopride diminished the deficit of sensorimotor gating produced by the uncompetitive antagonist of NMDA receptors phencyclidine. It is supposed that such a long time of administration is necessary to produce some adaptive changes which may improve the processing of sensory information. At present it is not possible to determine whether the influence of neuroleptics on cortical NMDA receptors is responsible for such an effect, because clozapine appears to have no effect in the prepulse inhibition model. All the same, it is speculated that the increased number of cortical receptors induced by neuroleptics counteracts their blockade produced by phencyclidine. The fact that both the frontal and parietal cortices are engaged in sensorimotor functions supports this view. The lack of clozapine effect on the sensorimotor gating deficit induced by phencyclidine may result from a very complex receptor profile of this neuroleptic.

A search for the role of metabotropic glutamate receptors in the antipsychotic drug action

Moghaddam et al. (1997) and Adams and Moghaddam (1998) found that the psychotomimetic uncompetitive antagonists of NMDA receptors phencyclidine and ketamine increased the glutamate efflux in the prefrontal cortex and nucleus accumbens and, at the same time produced a working memory deficit measured by a delayed alternation procedure. In that task an animal is trained in a T-maze to find a food pellet placed alternately in either a right or a left arm. This paradigm involves continuous changing strategies, and the impairment found in this model is thought to resemble a frontal lobe dysfunction and working memory deficits associated with schizophrenia. The authors suggest that the blockade of postsynaptic NMDA receptors produces a secondary release of glutamate from presynaptic terminals whose action on non-NMDA receptors is responsible for the psychotomimetic effect of NMDA receptor antagonists.

Moghaddam and Adams (1998) also found that LY 354740 – a selective agonist of group II metabotropic glutamatergic receptors – administered in a dose of 10 mg/kg decreased the glutamate release in the prefrontal cortex, as well as the working memory deficit, locomotor activity and stereotypy induced by phencyclidine. On the basis of those findings the authors speculated that targeting group II metabotropic glutamate receptors may ameliorate symptoms of acute phencyclidine psychosis or related symptoms of schizophrenia.

The aim of our present study was to continue a research of Moghaddam and Adams (1998) and to check the effect of LY 354740 on sensorimotor and working memory deficits. The sensorimotor gating deficits were evoked by phencyclidine (5 mg/kg sc) and apomorphine (0.5 mg/kg sc) and were measured in a prepulse inhibition model. The working memory deficits were produced by the selective uncompetitive antagonist of NMDA receptors, MK-801 (0.2 mg/kg ip).

LY 354740 in a dose of 10 mg/kg ip did not influence the phencyclidine-induced decreases of prepulse inhibition. Nor did LY 354740 in doses of 2.5 and 5 mg/kg reverse the working memory deficit induced by MK-801. Moreover, in contrast to the majority of well-known neuroleptics, LY 354740 in a dose of 10 mg/kg did not influence the disturbances of prepulse inhibition produced by apomorphine.

The present results do not confirm Moghaddam and Adams' (1998) conclusion about the putative antipsychotic role of the above-mentioned agonist of group II glutamate metabotropic receptors. In our laboratory, this compound did not alleviate disturbances which seem to model deficits associated with schizophrenia in humans. Further studies are necessary to clarify this problem.

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