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Detrimental effect of postnatal blockade of N-methyl-D-aspartate receptors on sensorimotor gating is reversed by neuroleptic drugs

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Abstract:
Postnatal hypofunction of N-methyl-D-aspartate (NMDA) receptors leads to several behavioral deficits in adult rats resembling deficits typical of schizophrenia-like deficits of sensorimotor gating. Thus far, it is not known whether the above disruptions are sensitive to neuroleptic drugs. In order to verify the above model in pharmacological terms, we investigated whether deficits in the sensorimotor gating evoked by administration of NMDA receptor antagonists in the postnatal period is sensitive to neuroleptic drugs. We also investigated whether such treatment evoked alterations in the expression of dopamine D_1, D_2 and D_3 receptors in the nucleus accumbens, a key structure for dopamine-dependent alterations in sensorimotor gating. CGP 40116, a competitive antagonist of NMDA receptors was given in doses of 1.25 mg/kg on days 1, 3, 6 and 9; 2.5 mg/kg on days 12, 15 and 18; and 5 mg/kg on day 21 (all injections were *sc*). The efficacy of sensorimotor gating was tested on rats at the age of 60 days using a prepulse-induced inhibition of the startle reflex. In order to measure the expression of dopamine D_1, D_2 and D_3 receptors, we used quantitative autoradiography and tritiated ligands i.e. [^{3}H]-SCH 23390, [^{3}H]-Spiperone and [^{3}H]-7-OH-DPAT, respectively. Haloperidol (0.1 mg/kg, *sc*), risperidone (1.0 mg/kg, *sc*) and clozapine (2.5 mg/kg, *sc*) reversed deficits of sensorimotor gating observed in adult rats evoked by the postnatal administration of CGP 40116. We also observed enhanced density of dopamine D_3, but not D_1 and D_2 receptors in the nucleus accumbens of CGP 40116 treated rats. It is concluded that models of cognitive dysfunction, typical for schizophrenia based on postnatal administration of NMDA receptor antagonists, are sensitive to neuroleptic drugs and possibly not dependent on alteration in the density of dopaminergic receptors.

Key words:
developmental model of schizophrenia, NMDA receptors, sensorimotor gating, neuroleptic drugs

Introduction

A growing amount of evidence indicates that the postnatal administration of competitive and non-competitive antagonists of N-methyl-D-aspartate (NMDA) receptors may model certain symptoms of schizophrenia with substantial construct and etiological validity. In terms of construct validity, the above models fit the concepts of glutamatergic hypofunction as a common denominator of abnormalities associated with schizophrenia (for recent review see [13]). With respect to etiological validity, it has been suggested that the postnatal administration of NMDA receptor antagonists, *via* alterations of brain development and maturation (specifically interference with developmental

apoptosis), results in functional impairment [5, 35]. For example, it has been found that the administration of competitive antagonists of NMDA receptors like CGP 40116, in contrast to ketamine and phencyclidine, is devoid of psychotomimetic effects (as far as sensorimotor gating is considered [39]), can evoke deficits of working memory, sensorimotor gating, social interaction, and locomotor agitation [38]. The above functional effects have been followed by anatomical changes like alterations in the morphology of basilar dendrites [37], density of tyrosine hydroxylase axonal arbors [36], and a resemblance to pathophysiological disturbances observed postmortem in patients suffering from schizophrenia [1, 2, 8, 12]. Developmental models of schizophrenia are not only limited to administration of the above-mentioned competitive antagonists of NMDA receptors, but can also be based on non-competitive antagonists like ketamine and phencyclidine [5, 35], although it is not known to our knowledge whether antagonist of above receptors devoid of psychotomimetic activity like amantadine [21] for example evoked similar effects.

Thus, it was of interest to further verify the above model and analyze whether it has pharmacological validity, or in other words, whether deficits of sensorimotor gating are sensitive to neuroleptic drugs. In order to test the pharmacological validity in the present study, we analyzed whether defects in sensorimotor gating evoked by the postnatal administration of competitive antagonists of NMDA receptors are antagonized by typical (haloperidol) and atypical (risperidone and clozapine) neuroleptics [11]. In the present study, a prepulse inhibition (PPI) of the startle reflex was used as an operational measure of efficacy of sensorimotor gating. PPI can be studied across various species of experimental animals, and is disrupted in schizophrenic patients [7, 19, 28]. The dopaminergic components of PPI [29], which are in line with the dopaminergic theory of schizophrenia, or the mechanism of action of antipsychotic drugs on anatomical grounds, are linked with the nucleus accumbens septi [30, 34]. The peripheral or intra-accumbal administration of direct and indirect dopamine receptors agonists compromises the efficacy of PPI. The above detrimental effects of direct or indirect agonists of dopaminergic receptors can be reversed by typical and atypical antipsychotics [9, 15]. The anatomical linkage between the efficacy of PPI and the nucleus accumbens, and our previous data showing the enhancement of locomotor activity after quinpirole and amphetamine on a group of rats treated postnatally with CGP 40116 [36], inclined us to analyze the density of dopaminergic receptors with aims of quantitative autoradiography.

Materials and Methods

Animals

Pregnant dams (Wistar, IP, PAS Kraków) were housed under standard experimental conditions (constant temperature of 22 ± 2°C) with an artificial light/dark cycle (12/12; light on from 7 a.m. to 7 p.m.). All subjects used in the experiment were born in the experimental facilities of IP, PAS (Kraków). Dams and pups were housed in standard cages with wooden bedding and had free access to laboratory chow and tap water.

Administration of NMDA receptor antagonist

A day of parturition was designated as a postnatal day 0. Rat pups of both sexes were injected *sc* with increasing doses of CGP 40116 [(E)-(±)-2-amino-4-methyl--5-phosphono-3-pentenoic acid, Ciba-Geigy, Switzerland]. The drug dose was 1.25 mg/kg on days 1, 3, 6 and 9, then 2.5 mg/kg on days 12, 15, and 18 and a final dose of 5 mg/kg was administered on day 21 (post-CGP group). The volume of drug solution was 0.01 ml per 1 g of body weight. Control pups received only vehicle (0.9% NaCl, post-Veh group). On postnatal day 22, the rats were separated from the mothers and the females were sacrificed. Rats were randomly assigned to groups of 6 animals per cage. Rats were kept with water and food available *ad libitum*. All experiments were performed on adult 60-day-old male rats. Rats were used only once. The experimental protocols were approved by the Committee for Laboratory Animal Welfare and Ethics of the IP PAS Kraków and met the criteria of the International Council for Laboratory Animals and Guide for the Care and Use of Laboratory Animals (above procedure has been described previously – see: [36, 38]).

Startle apparatus

Startle reactivity was measured using twelve startle chambers (SR-LAB, San Diego Instruments, San Diego, CA). Each chamber consisted of a nonrestrictive Plexiglas cylinder (inner diameter = 38 cm) resting on

a platform inside a soundproof cabinet. A continuous background noise of 65 dB was generated by a loudspeaker inside of each cabinet. Vibrations of the Plexiglas cylinder caused by the whole-body startle response of the animal were transduced into analog signals by a piezoelectric unit attached to the platform. These signals were then digitized and stored by a computer. One hundred readings were taken at 1 ms intervals, starting at stimulus onset. Each reading was averaged automatically and given as an average amplitude of startle reflex. Additionally, the time to peak was determined by the SR-LAB software. The SR-LAB calibration unit was used routinely to ensure consistent stabilimeter sensitivity between test chambers and over time, and the sound levels in dB were measured as described previously with an external sound level meter, SM1-Sonopan (Białystok, Poland).

Prepulse inhibition (PPI) session

After habituation (5 min, background white noise, 65 dB), two types of acoustic stimuli were used in random order: acoustic stimulus alone [120 dB, 40 ms, (P)] or an acoustic stimulus preceded by an acoustic prepulse [75 dB, 20 ms, (PP)] applied 100 ms before the stimulus (P). During each experimental session, 20 trials of each type were presented with interstimulus intervals of 20 ms. The amplitudes were averaged for each individual animal, and separately for both types of trials, stimulus alone (P) or stimulus preceded by a prepulse (PP). The degree of prepulse inhibition was shown as a percentage of inhibition (%PPI) calculated as follows: %PPI = {[(startle response for PULSE-ALONE – startle response for PREPULSE + PULSE)/(startle response for PULSE-ALONE)] × 100%}, (for further details see [39]).

Drugs

Haloperidol (4-[4-(4-chlorophenyl)-4-hydroxy-1-piperidinyl]-1-(4-fluorophenyl)-1-butanone hydrochloride); Polfa, Poland; ampules of 5 mg of haloperidol in 1 ml *aqua pro injectione*) was dissolved in 0.9% NaCl and was given in a dose of 0.1 mg/kg *sc* 30 min before the test. Risperidone (3-[2-[4-(6-fluoro-1,2-benzisoxazol-3-yl)-1-piperidinyl]ethyl]-6,7,8,9-tetrahydro-2-methyl-4H-pyrido[1,2-a]pyrimidin-4-one; Janssen, Belgium) was dissolved in 1% Tween 80 and was given in a dose of 1 mg/kg *ip* 60 min before the test. Clozapine (8-chloro-11-(4-methyl-1-piperazinyl)-5H-dibenzo[b,e]-[1,4]diazepine; Sandoz, Switzerland) was dissolved in a drop of acetic acid and dissolved to final concentration in 0.9% NaCl and was given in a dose of 2.5 mg/kg *sc* 30 min before the test. All drugs were given in a volume of 2 ml/kg.

Autoradiographic studies

Tissue preparations

The rats were sacrificed by decapitation, their brains were removed, trimmed into parts containing the striatum/nucleus accumbens using rodent brain matrix (Activational Systems Inc., Mich., USA) and frozen by submersion in dry ice-cold 2-metylbutane. The frozen blocks were stored at –20°C. The coronal brain sections (10 μm) were cut using a Leica cryostat LC 3000 (temperature –17°C), thaw-mounted on gelatin-coated glass microscopic slides and stored with a desiccant at –20°C prior to the receptor assay.

Autoradiography of D_1, D_2 and D_3 receptors

For an assay of D_1 receptors, the sections were preincubated once for 5 min (room temperature) in incubation buffer (50 mM TRIS buffer, pH = 7.4, containing 120 mM NaCl, 5 mM KCl, 2 mM $CaCl_2$, and 1 mM $MgCl_2$). The sections were then incubated for 60 min (room temperature) in an incubation buffer supplemented with ascorbic acid (1 mM), bovine serum albumin (10 mg/100 ml) and 1 nM [^{3}H]SCH 23390 (Amersham, spec. activ. 70 Ci/mmol). In order to block serotonergic receptors, ketanserine (40 nM) was added to the incubation solution. The non-specific binding was determined by incubation of anatomically adjacent sections in the same amount of [^{3}H]SCH 23390 in the presence of 5 μM butaclamol. For an assay of D_2 dopamine receptors, sections were exposed to two preincubations in 50 mM TRIS buffer (pH = 7.4) for 30 min each (at 0°C and at room temperature, respectively), followed by 60 min of incubation (room temperature) in the same buffer containing 0.5 nM [^{3}H]-spiperone (Amersham, spec. activ. 18.6 Ci/mmol) and 40 nM ketanserine. The non-specific binding was determined using the same concentration of labelled spiperone in the presence of 2.5 μM butaclamol. For an assay of D_3 receptors, sections were preincubated in 50 mM TRIS buffer (pH = 7.4) for 60 min at room temperature followed by an incubation with 50 mM TRIS buffer supplemented with 1 nM [^{3}H]7-OH-DPAT

(Amersham, spec. activ. 155 Ci/mmol), 5 μM 1,3-di-(2-tolyl)guanidine (DTG) and 0.1% ascorbic acid for 60 min. The non-specific binding was determined using the same concentration of labelled 7-OH-DPAT in the presence of 10 μM quinpirol. After incubations with radioactive ligands, all the sections were rinsed and dried. Autoradiograms of D_1, D_2 and D_3 receptors were generated by apposing the labelled tissue sections to a Hyperfilm-^{3}H (Amersham) in an X-ray cassette (Du Pont). The radioactive standards (Amersham) were coexposed to the same X-ray film. After 2 or 6 weeks of exposure, the films were developed in a Kodak D-19 developer.

Quantification of autoradiograms

The optical density of each autoradiogram was quantified with an image analyzer (Java, Jandel and IBM PS/2 computers). Mean optical density readings from autoradiograms were converted by the same software (Java, Jandel) into the amount of radioligand bound per mg of tissue using the standard reference curve and a tritium-labelled autoradiographic scale (Amersham). The specific binding of ligands ([^{3}H]spiperone and [^{3}H]7-OH-DPAT) to dopamine D_2 and D_3 receptors, respectively, was calculated by subtracting non-specific binding (the radioligand in the presence of a displacer) from the total binding (the radioligand without a displacer). For D_1 dopamine receptors, the whole binding resulting from [^{3}H]SCH 23390 was considered to be specific, as virtually no binding above the background was detected in the presence of displacer (non-specific binding). In order to differentiate between histological subregions of the structures examined, tissue sections were stained with Cresyl Violet. The stained sections were then compared with the corresponding autoradiographic images.

Statistics

The data were analyzed by one-way AVOVA (quantitative autoradiography), two-way analysis of ANOVA for repeated measurements, with postnatal administration of CGP *vs.* neuroleptics as the dependent variable and repeated measurements with pulse alone *vs.* prepulse + pulse as a within factor, and finally two-way ANOVA for postnatal administration of CGP 40116 *vs.* neuroleptic drug – % of inhibition. Duncan's test was used for *post-hoc* comparisons. For calculations, Statistica package version 8.0 was used.

Results

The effects of postnatal administration of CGP 40116 on the efficacy of sensorimotor gating in adult rats – impact of neuroleptic drugs

Haloperidol

Two-way ANOVA for repeated measurements (type of acoustic stimuli as a within factor) revealed a strong effect of postnatal administration of CGP 40116 on the amplitude of startle reflex evoked by acoustic pulse [$F(1, 44) = 19.1$, $p < 0001$]; no significant effects of haloperidol (0.1 mg/kg *sc*) [$F(1, 44) = 1.11$, ns] and no significant interaction between CGP 40116 pretreatment and haloperidol administration were observed [$F(1, 44) = 0.9$, ns]. There was a significant difference between the startle amplitude evoked by pulse alone and prepulse + pulse [$F(1, 44) = 53.27$, $p < 0.001$], and on the level of amplitude of startle reflex evoked by prepulse + pulse. No significant effect of CGP 40116 [$F(1, 44) = 3.36$, ns] or haloperidol was observed [$F(1, 44) = 7.43$, $p < 0.01$], and no interaction between haloperidol and the postnatal inhibition of glutamatergic transmission was observed either [$F(1, 44) = 0.043$, ns] (Fig. 1 panel A).

In terms of % of inhibition, two-way ANOVA revealed significant effects of postnatal administration of CGP 40116 [$F(1, 44) = 11.51$, $p < 0.002$], and a significant impact of haloperidol [$F(1, 44) = 8.49$, $p < 0.002$]. Although haloperidol normalized the effects of glutamatergic receptor hypofunction evoked in the postnatal period on % of inhibition of PPI to the level of control animals (42% and 39%, respectively, for control and post-CGP), in terms of statistics, the above interaction between postnatal-CGP and haloperidol treatment did not reach the criteria for significance [$F(1, 44) = 8.49$, ns], probably due to the fact that haloperidol alone evoked a massive enhancement of % of inhibition of prepulse-evoked inhibition of startle reflex (Fig. 1 panel A).

Risperidone

Analysis of the amplitude of startle reflex evoked by an acoustic pulse in post-CGP rats treated with risperidone (1 mg/kg *sc*) and respective controls, when analyzed by two-way ANOVA for repeated measurements (type of acoustic stimuli as a within

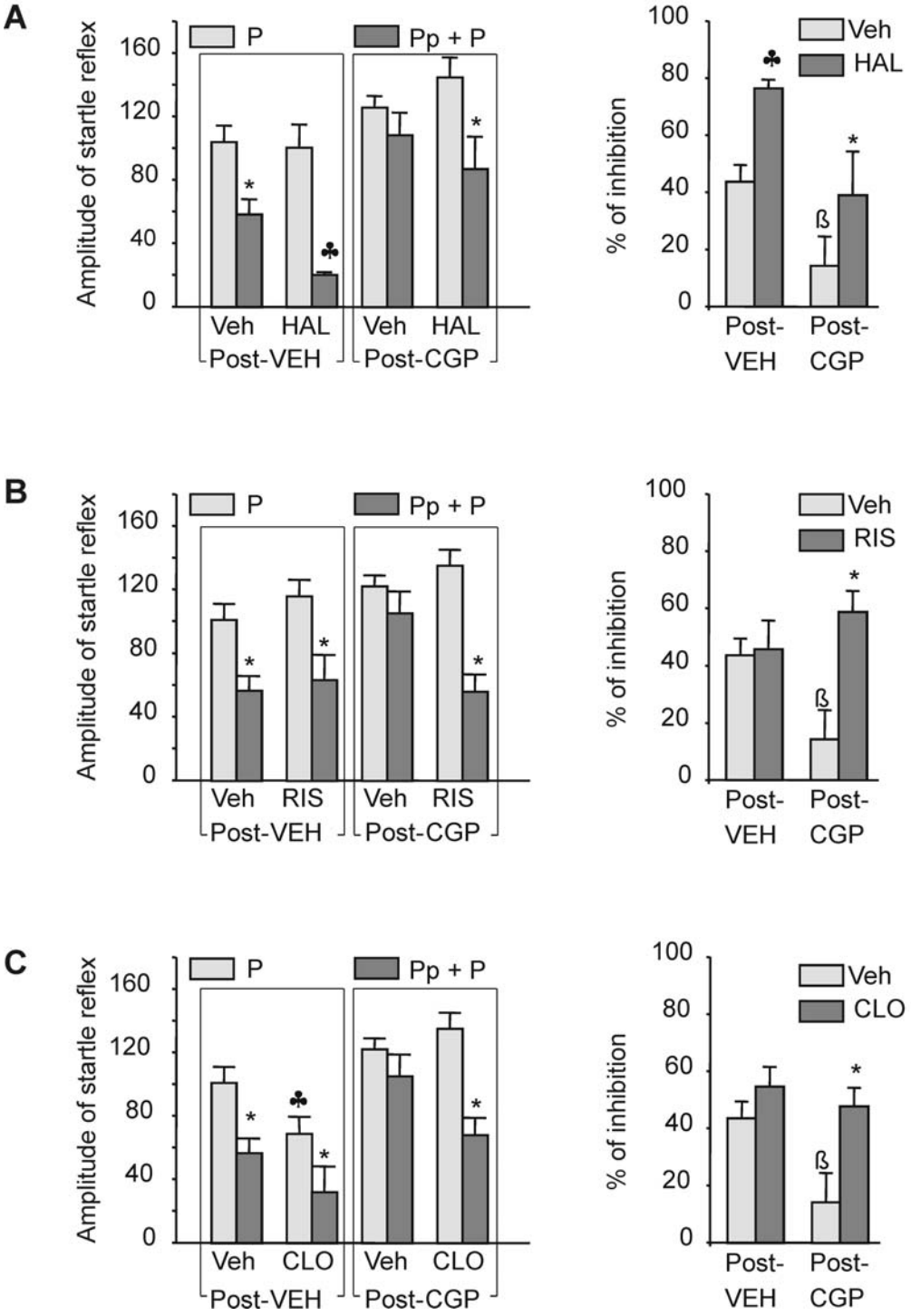

Fig. 1. The effects of **(A)** haloperidol (HAL, 0.1 mg/kg *sc*); **(B)** risperidone (RIS, 1 mg/kg *sc*) and **(C)** clozapine (CLO, 2.5 mg/kg *sc*) on deficits of prepulse-induced inhibition of acoustic startle response induced by postnatal administration of CGP 40116. Left column of graphs – amplitude of startle reflex evoked by acoustic pulse alone (P) and acoustic pulse preceded by acoustic prepulse (Pp + P) on vehicle and CGP 40116 treated animals in postnatal period (respectively Post-Veh and Post-CGP) and injected at the age of 60 days with HAL, RIS and CLO. Right panel: calculated % of inhibition (abbreviation as on left column). All data are given as arithmetical means ± SEM. Left column: two-way ANOVA for repeated measurements; right column: two-way ANOVA, Duncan test for *post-hoc* comparisons, n = 12 for each group of animals. On the left column (A–C), * – indicates significant differences between startle amplitude evoked by P and Pp. On A, ♣ – indicates difference between amplitude of startle reflex evoked by Pp in Post-Veh animals treated with vehicle and HAL. On C, ♣ – indicates differences between amplitude of startle reflex evoked by P in Post-Veh animals treated with VEH and CLO. On the right column A–C, ß – indicates detrimental effect of postnatal administration of CGP 40116, * – indicates significant effect of neuroleptic on deficits in PPI in Post-CGP animals, and ♣ – indicates enhancement of PPI evoked by HAL in Post-Veh animals

factor), revealed significant effects of postnatal administration of CGP 40116 [F(1, 44) = 4.5, p < 0.05], no significant effect of risperidone [F(1, 44) = 0.14 ns], and no significant interaction between risperidone and postnatal treatment [F(1, 44) = 2.25, ns]. There was a significant general difference between the amplitude of startle evoked by pulse alone *vs.* prepulse + pulse [F(1, 44) = 66.44, p < 0.0001]. No significant effects of postnatal pretreatment with CGP 40116 on the amplitude of prepulse + pulse [F(1, 44) = 0.07, ns] or of risperidone were observed [F(1, 44) = 8.58, p < 0.005], but significant differences between risperidone and the postnatal administration of CGP 40116 [F(1, 44) = 5.22, p < 0.03] (Fig. 1 panel B) with respect to the amplitude of startle reflex evoked by prepulse + pulse were observed.

In terms of % of inhibition, two-way ANOVA revealed significant effects of postnatal administration of CGP 40116 [F(1, 44) = 4.3, p < 0.05], a significant impact of risperidone [F(1, 44) = 8.49, p < 0.001], and a significant interaction between risperidone and postnatal treatment with CGP 40116 [F(1, 4) = 14.6, p < 0.001].

Clozapine

An analysis of data on the impact of clozapine on the effects of postnatal administration of CGP 40116 on the level of amplitude of startle reflex evoked by acoustic pulse showed (two-way ANOVA for repeated measurements) that there were significant effects of postnatal administration of CGP 40116 [F(1, 44) = 18.37, p < 0.001], a significant effect of clozapine [F(1, 44) = 6.3, p < 0.01] and a non-significant interaction between postnatal treatment with CGP 40116 and clozapine [F(1, 44) = 0.3, ns], indicating that the effects induced by the postnatal blockade of NMDA receptors was abolished by clozapine, apart from the substantial impact of clozapine on the amplitude of startle reaction evoked by an acoustic pulse alone. Again, there was a significant difference between the amplitude of startle evoked by pulse alone *vs.* prepulse + pulse [F(1, 44) = 61.38, p < 0.0001], but no significant effect of postnatal treatment with CGP 40116 [F(1, 44) = 0.21, ns]. Additionally, there was a significant effect of clozapine [F(1, 44) = 4.31, p < 0.05] and a significant interaction between postnatal pretreatment and clozapine [F(1, 44) = 4.56, p < 0.05] (Fig. 1 panel C).

In the experiments with clozapine on the level of % of inhibition, we observed with two-way ANOVA, a significant effect of clozapine [F(1, 44) = 13.3, p < 0.001], a significant effect of postnatal administration of CGP 40116 [F(1, 44) = 6.05, p < 0.05] and a significant interaction between clozapine and CGP 40116 [F(1, 44) = 4.3, p < 0.05].

The effects of postnatal administration of CGP 40116 on the expression of dopamine D_1, D_2 and D_3 receptors in adult rats – quantitative receptor autoradiography

The pattern of distribution of D_1, D_2 and D_3 receptors labelled with [^{3}H]-SCH 23390, [^{3}H]-spiperone and [^{3}H]-7-OH-DPAT, respectively, was similar to previous reports [36] and our own data published previously [36], indicating the reliable level of receptor

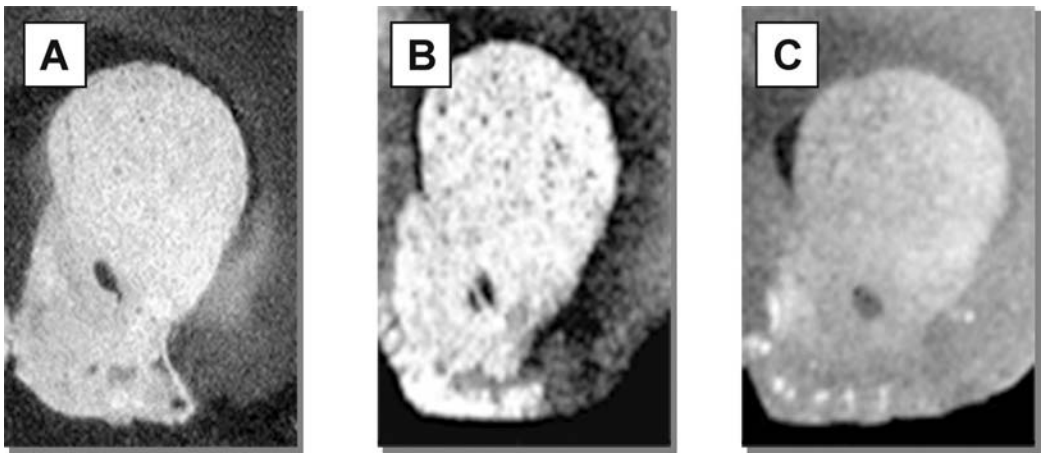

Fig. 2. The representative autoradiograms illustrating the distribution of dopamine D_1, D_2 and D_3 receptors on the level of the nucleus accumbens septi and frontal striatum

Tab. 1. The effects of postnatal administration of CGP 40116 on the density of dopaminergic receptors in the nucleus accumbens of rats

		Receptor density (fmol/mg of tissue)			
		Post-Veh		Post-CGP	
Receptor	Ligand	Shell	Core	Shell	Core
D_1	[^{3}H]-SCH 233390	189.6 ± 19.1	204.4 ± 16	245.5 ± 16.8	191.9 ± 12.7
D_2	[^{3}H]-Spiperone	51.7 ± 11.4	33.4 ± 9.6	51.2 ± 6.3	37.7 ± 5.7
D_3	[^{3}H]-7-OH-DPAT	13.7 ± 1.8	nd	2.6 ± 0.6*	nd

All data are given as arithmetical means ± SEM. One-way ANOVA Duncan's test for *post- hoc* comparisons, n = 10 for each group of animals.
* – Indicates significant difference in receptor density between Post-Veh and Post-CGP rats; nd – not detected

staining by the applied experimental protocol (Fig. 2). One-way analysis of variance revealed that the postnatal administration of CGP 40116 leads to a sustained decrease of [^{3}H]-7-OH-DPAT binding in the core of the nucleus accumbens [$F(1, 18) = 41.5$, $p < 0.001$] (Tab. 1). The binding of [^{3}H]-SCH 23390 and [^{3}H]-spiperone was not changed in subregions of the nucleus accumbens (Tab. 1) and striatum (data not shown). F values for D_1 and D_2 receptor density comparison between groups in the nucleus accumbens were (shell and core): $F(1, 18) = 4.3$, ns; $F(1, 18) = 0.3$, ns and $F(1, 18) = 0.01$, ns; $F(1, 18) = 0.14$ ns, respectively.

Discussion

The major finding of the present study indicates that the detrimental effects of postnatal administration of CGP 40116 on the efficacy of sensory motor gating are sensitive to typical (haloperidol) and atypical neuroleptic drugs (clozapine, and risperidone) [11]. Moreover, we found that postnatal blockade of NMDA receptors leads to the persistent downregulation of dopamine D_3, but not dopamine D_1 or D_2 receptors, in the nucleus accumbens of rat, a centre for the dopamine-dependent regulation of PPI.

It has been previously discussed that acceptable animal models of schizophrenia must display at least three separate schizophrenia-relevant behaviors to be accepted as a relevant model of the disease [19], since patients with schizophrenia do not manifest all symptoms of the disease. Our previous experiments suggest that hypoactivity of glutamatergic neurotransmission in the postnatal period models an "endophenotype" of schizophrenia in adulthood that is characterised by: positive symptoms like locomotor agitation (enhanced exploration), cognitive deficits (impaired sensorimotor gating and acquisition of a correct response in the working memory test) and negative symptoms like social withdrawal (social interaction test) [38], apart of fear like behavior [6]. The present data indicate that the above model is also validated in terms of susceptibility to clinically effective pharmacological intervention like the administration of typical antischizophrenic drugs [11].

Several sets of data indicate that overactivity of dopaminergic neurotransmission in the nucleus accumbens may be responsible for deficits in sensorimotor gating. It has been shown that direct or indirect agonists of dopaminergic receptors administered in the nucleus accumbens decreased prepulse-induced inhibition of the acoustic startle response [30, 33]. It is conceivable that the deficits in the PPI observed here may be associated with the dopaminergic neurotransmission in the nucleus accumbens. We found previously that post-CGP animals display enhanced locomotor stimulant effects to amphetamine and quinpirole, i.e. effects associated with functional overactivity of subcortical dopaminergic systems [36]. We mentioned the function since the density of dopamine receptors of the D_1 and D_2 subtypes had not changed, as has been shown by receptor autoradiography. Deficits of PPI are evoked not only by direct and indirect dopaminergic agonists, but also by antagonists of NMDA receptors [3, 4, 14, 16], agonists of serotonin 5-HT_{1A} and 5-HT_{2A} receptors [18, 24–26] and cannabinoids [22]. However, in our model, that haloperidol (antagonists of dopamine D_2 receptors), risperidone and clozapine attenuate deficits of PPI to the same extent suggests instead the involvement of the dopaminergic system, and not the serotonergic one, along with en-

gagement of the dopamine D_2 receptor family. Such a conclusion is in line with available data that agonists of the D_2 receptor family reliably disrupt PPI in rats [20, 31, 32], whereas the effect of dopamine agonists that nonspecifically activate D_1-family (D_1 and D_5) receptors are more variable, appearing to be strain- and even supplier-dependent [20, 31, 32]. Although involvement of dopamine D_3 receptors in the pathophysiology of schizophrenia is a matter of debate (both on the level of D_3 receptor expression and the polymorphism of the gene encoding D_3 receptors) ([27] and references there), the apparent downregulation is puzzling. Interestingly, recent data with a highly selective agonist of D_3 receptors revealed that their activation compromises the efficacy of sensorimotor gating [40]. Moreover, such a detrimental effect is antagonized by risperidone and clozapine, but not haloperidol. The above data led us to the conclusion that, for the first time, the sustained downregulation of dopamine D_3 receptors is not engaged in the detrimental effects of postnatal administration of CGP 40116, or the above effect is compensated by dopaminergic signalling based on activation of dopaminergic receptors of the D_1 and D_2 subtypes. It has to be mentioned that in our recent study, we found that postnatal administration of CGP 401116 decreased the density of tyrosine hydroxylase immunoreactive axonal arbors in the medial prefrontal cortex of rats [36], which speaks for the hypofunction of dopaminergic neurotransmission in the mesocortical dopaminergic system. Interestingly, it has been shown that decreased dopaminergic tone in the MPC evoked the enhancement of dopaminergic neurotransmission in the mesolimbic system [17]. Such a differentiating factor is based on descending glutamatergic neurotransmission from the medial prefrontal cortex to the ventral tegmental area and direct excitatory synapses on mesocortical dopaminergic cells and excitatory synapses on GABAergic neurons controlling the activity of mesolimbic dopaminergic cells [10, 23].

In conclusion, the present data indicate that the model of schizophrenia based on the administration of competitive antagonists of NMDA receptors not only models positive and negative symptoms of schizophrenia and anatomical abnormalities in the medial prefrontal cortex, but also evokes symptoms like deficits of PPI, which are sensitive to typical and atypical neuroleptics [11]. Apart from the predictive validity, it has be to mentioned that the above model also has epidemiological and pharmacological validity. The stage development of rat brain shortly after delivery is equivalent to the stage of development of the human brain around the second trimester of human pregnancy, i.e. the period of high risk to developmental malfunction resulting in the adulthood in appearance of schizophrenia.

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