Schizophrenia Research xxx (2014) xxx–xxx

Contents lists available at ScienceDirect

Schizophrenia Research

journal homepage: www.elsevier.com/locate/schres

A fundamental role for hippocampal parvalbumin in the dopamine hyperfunction associated with schizophrenia

Angela M. Boley, Stephanie M. Perez, Daniel J. Lodge *

Department of Pharmacology, Center for Biomedical Neuroscience, University of Texas Health Science Center at San Antonio, San Antonio, TX, USA

ARTICLE INFO

Article history:
Received 28 March 2014
Received in revised form 6 May 2014
Accepted 9 May 2014
Available online xxxx

Keywords:
Parvalbumin
Hippocampus
Dopamine
Schizophrenia

ABSTRACT

Postmortem studies in schizophrenia patients have demonstrated robust alterations in GABAergic markers throughout the neuraxis. It has been suggested that these alterations are restricted to subpopulations of interneurons, such as those containing the calcium binding protein parvalbumin. Indeed, a reduction in parvalbumin expression is a consistent observation in human postmortem studies, as well as, in a wide and diverse variety of animal models. However, it still remains to be determined whether this decrease in parvalbumin expression contributes to, or is a consequence of the disease. Here we utilize lentiviral delivered shRNA and demonstrate that a selective reduction in parvalbumin mRNA expression induces hyperactivity within the ventral hippocampus. In addition, we observe downstream increases in dopamine neuron population activity without changes in average firing rate or percent burst firing. These changes in dopamine neuron activity were associated with an enhanced locomotor response to amphetamine administration. These data therefore demonstrate that a reduction in ventral hippocampal parvalbumin expression is sufficient, in and of itself, to induce an augmented dopamine system function and behavioral hyper-responsivity to amphetamine, implicating a potential key role for parvalbumin in the pathophysiology of schizophrenia.

© 2014 Elsevier B.V. All rights reserved.

1. Introduction

Schizophrenia is a neuropsychiatric disease affecting up to 1% of the population (Bhugra, 2005; Saha et al., 2005). It is known to be multifactorial with both genetic and environmental factors contributing to the disease (Agid et al., 1999; Sawa and Snyder, 2002; Purcell et al., 2009). This heterogeneity has led to a number of distinct hypotheses of schizophrenia, however as yet, the specific neuropathology underlying this disease has not been conclusively determined. Nonetheless, clinical data from post mortem studies have provided some consistent observations, including an altered expression of GABAergic markers throughout both cortical and hippocampal regions (Lewis et al., 2005; Konradi et al., 2011). Specifically, decreases in glutamic acid decarboxylase (GAD)-1 mRNA and GAD-67 protein are observed throughout the cortex of schizophrenia patients (Hashimoto et al., 2003). It should be noted that these alterations in GABAergic markers are not global; rather, they are more prevalent among distinct subclasses of interneurons, including those that express the calcium binding protein parvalbumin (PV) (Lewis et al., 2005). Indeed, a decreased expression of PV is a consistent observation not only in postmortem human schizophrenia patients (Lewis et al., 2005; Konradi et al., 2011), but also in a diverse number of rodent models of the disease (Cunningham et al., 2006; Abdul-Monim et al., 2007; Behrens et al., 2007; Harte et al., 2007; Francois et al., 2009; Lodge et al., 2009). However, whether this decrease in PV expression is the cause, or a consequence, of the illness is not currently known.

Decreases in PV expression are observed in both the prefrontal cortex (Lewis et al., 2012) and the hippocampus (Konradi et al., 2011), two brain regions that are consistently implicated in schizophrenia. Deficits in prefrontal cortical function likely contribute to cognitive impairments and working memory deficits (Weinberger et al., 1986; Goldman-Rakic, 1995), whereas aberrant hippocampal function is associated with positive symptom severity (Schobel et al., 2009). We have recently demonstrated, in the methylazoxymethanol acetate (MAM) rodent model of schizophrenia (for review, see Lodge and Grace, 2009), that the dopamine hyperfunction and associated behavioral hyper-responsivity to psychomotor stimulants are driven by aberrant activity within the ventral hippocampus (vHipp) (Lodge and Grace, 2007; Perez and Lodge, 2013; Perez et al., 2013). Given that PV interneurons are perisomatic targeting (Kawaguchi and Hama, 1987; Benes and Berretta, 2001; Gonzalez-Burgos and Lewis, 2012), we posit that a loss of PV interneuron function would result in an increased pyramidal cell firing (in the vHipp) that is sufficient to induce a schizophrenia-like phenotype. Here we utilize lentiviral delivered short hairpin RNA (shRNA) to

* Corresponding author at: Department of Pharmacology, Center for Biomedical Neuroscience, University of Texas Health Science Center at San Antonio, 7703 Floyd Curl Dve, MC 7764, San Antonio, TX, USA. Tel.: +1 210 567 4188.
E-mail address: LodgeD@uthscsa.edu (D.J. Lodge).

http://dx.doi.org/10.1016/j.schres.2014.05.005
0920-9964/© 2014 Elsevier B.V. All rights reserved.

examine the consequence of vHipp PV knockdown, as it pertains to the dopamine dysfunction in schizophrenia.

2. Materials and methods

All experiments were performed in accordance with the guidelines outlined in the USPHS Guide for the Care and Use of Laboratory Animals and were approved by the Institutional Animal Care and Use Committee of the University of Texas Health Science Center.

2.1. Lentivirus administration

All survival surgical procedures were performed under general anesthesia in a semi-sterile environment. Adult male Sprague–Dawley rats obtained from Harlan Laboratories were anesthetized with sodium pentobarbital (60 mg/kg, i.p.) and placed in a stereotaxic apparatus. Anesthesia was maintained by supplemental administration of sodium pentobarbital as required to maintain suppression of limb compression withdrawal reflex. A core body temperature of 37 °C was sustained by a thermostatically controlled heating pad. The skin was reflected and bore holes drilled bilaterally overlying the vHipp (A/P +5.3, M/L +5.2, D/V −7.0 mm from bregma). Rats were administered (0.75 μl/side) commercially available (Thermo Scientific) high-titer lentivirus particles containing GIPZ vectors expressing shRNA targeting either parvalbumin (mature antisense: TAGCAGACAAGTCTCTGGC) or a non-silencing control. Rats were sutured and housed under ABSL 2 conditions for 72 h before being transferring to standard housing conditions. Rats were housed for a period of 6 weeks prior to behavioral and electrophysiological examinations to ensure stable transgene expression.

2.2. Amphetamine-induced locomotion

Rats were acclimatized to the behavioral facility for at least 1 h prior to being placed in an open field arena (Med Associates) where spontaneous locomotor activity in the X–Y plane was determined for 45 min by beam breaks and recorded with Open Field Activity Software (Med Associates). Following the baseline period, all rats were injected with D-amphetamine sulfate (0.5 mg/kg, i.p.) and locomotor activity recorded for 45 min. A subsequent dose (2.0 mg/kg, i.p.) was then administered and an additional 45 min of locomotor activity was recorded.

2.3. Extracellular recordings

Rats were anesthetized with chloral hydrate (400 mg/kg, i.p.), as this anesthetic does not significantly depress dopamine neuron activity (Hyland et al., 2002), and placed in a stereotaxic apparatus. Anesthesia was maintained by supplemental administration of chloral hydrate as required to maintain suppression of limb compression withdrawal reflex. A core body temperature of 37 °C was sustained by a thermostatically controlled heating pad. Extracellular glass microelectrodes (impedance 6–14 MΩ) were lowered into the right ventral hippocampus (A/P +5.3, M/L +5.0, D/V −5.0 to −8 mm from bregma) using a hydraulic micropositioner (KOPF — Model 640). Putative pyramidal neurons were defined as those with firing frequencies less than 2 Hz as reported previously (van der Meer and Redish, 2011; Shah and Lodge, 2013).

Electrodes were also inserted into the right ventral tegmental area (VTA; A/P −5.3, M/L +0.6 mm from bregma and −6.5 to −9.0 mm ventral of brain surface) and the activity of the population of dopamine neurons was determined by counting the number of spontaneously active dopamine neurons encountered while making multiple vertical passes (typically 6), separated by 200 μm, in a predetermined pattern to sample equivalent regions of the VTA. Spontaneously active dopamine neurons were identified with open filter settings (low pass: 30 Hz, high pass: 30 kHz) using previously established electrophysiological criteria (Grace and Bunney, 1983) and once isolated, activity was recorded for 2 − 3 min.

2.4. Quantitative PCR

Rats were decapitated following electrophysiological recordings and the brain was separated into two hemispheres. The vHipp was dissected from one hemisphere and homogenized. RNA was precipitated, separated by filtration and the concentration determined by absorbance at 260 nm. Single stranded RNA was converted to cDNA using a High Capacity cDNA Reverse Transcription Kit (Ambion). Real time PCR was performed with FAM-labeled TaqMan primers targeting either parvalbumin (Rn00574541_m1), GAD1 (Rn00690300_m1) or GAPDH (Rn01775763_g1). Detection of FAM labeled DNA was performed by a CFX384 Real-Time PCR Detection System (Bio-Rad). ΔCt was calculated as the number of PCR cycles required for mRNA detection (compared to the control mRNA, GAPDH), while fold-changes were expressed using the $2^{-\Delta\Delta Ct}$ method.

2.5. Parvalbumin immunohistochemistry

The hemisphere not used for PCR was post-fixed for at least 24 h, and cryoprotected (25% w/v sucrose in PBS) until saturated. Hemispheres were coronally sectioned (50 μm) using a cryostat (Leica). Ventral hippocampal slices were used to detect the expression of PV. Sections were washed three times (10 min) in PBS then blocked (2% normal goat serum & 0.3% Triton X-100) for 30 min at room temperature. Primary antibodies [anti-PV 1:1000 (Abcam; ab11427)] were applied (in PBS containing 1% normal goat serum and 0.3% Triton X-100) overnight at 4 °C followed by incubation with Alexa Fluor® 594 goat anti-rabbit IgG (H + L) for 1 h at room temperature. Slices were then mounted and cover slipped with ProLong gold antifade reagent. Computer assisted estimates of PV positive interneurons were performed using Neurolucida in combination with a Zeiss epifluorescent microscope.

2.6. Analysis

Locomotor data were analyzed by three separate 2-way ANOVAs (treatment and time as factors), one for each of the relevant time periods (spontaneous, 0.5 mg/kg, 2.0 mg/kg), followed by a Holm–Sidak post-hoc test, where appropriate. Electrophysiological analysis of single unit neuron activity was performed using commercial computer software (LabChart Pro — ADInstruments), and compared with student's t-test unless data failed test for normality and/or equal variance where a Mann–Whitney Rank Sum Test was utilized. PCR data was analyzed using commercial computer software (CFX Manager — BioRad). Cell counts were performed using Neurolucida and significance determined by a student's t-test. All data are represented as the mean ± standard error of the mean (SEM) unless otherwise stated. All statistical analyses were calculated using SigmaPlot (SYSTAT Software Inc.).

2.7. Materials

High titer lentiviral particles were obtained from Thermo Scientific. Pentobarbital sodium, chloral hydrate, and D-amphetamine sulfate were all purchased from Sigma. FAM-labeled TaqMan probes, the secondary Alexa Fluor® 594 goat anti-rabbit IgG (H + L) antibody as well as the ProLong gold antifade mountant were obtained from Life Technologies. The anti-parvalbumin antibody was purchased from Abcam. All other chemicals and reagents were of either analytical or laboratory grade and purchased from various suppliers.

Please cite this article as: Boley, A.M., et al., A fundamental role for hippocampal parvalbumin in the dopamine hyperfunction associated with schizophrenia, Schizophr. Res. (2014), http://dx.doi.org/10.1016/j.schres.2014.05.005

Download English Version:

https://daneshyari.com/en/article/6825184

Download Persian Version:

https://daneshyari.com/article/6825184

Daneshyari.com