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Functional magnetic resonance imaging reveals abnormal brain connectivity in EGR3 gene transfected rat model of schizophrenia

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ABSTRACT

Schizophrenia is characterized by the disorder of “social brain”. However, the alternation of connectivity density in brain areas of schizophrenia patients remains largely unknown. In this study, we successfully created a rat model of schizophrenia by the transfection of EGR3 gene into rat brain. We then investigated the connectivity density of schizophrenia susceptible regions in rat brain using functional magnetic resonance imaging (fMRI) in combination with multivariate Granger causality (GC) model. We found that the average signal strength in prefrontal lobe and hippocampus of schizophrenia model group was significantly higher than the control group. Bidirectional Granger causality connection was observed between hippocampus and thalamic in schizophrenia model group. Both connectivity density and Granger causality connection were changed in prefrontal lobe, hippocampus and thalamus after risperidone treatment. Our results indicated that fMRI in combination with GC connection analysis may be used as an important method in diagnosis of schizophrenia and evaluation the effect of antipsychotic treatment. These findings support the connectivity disorder hypothesis of schizophrenia and increase our understanding of the neural mechanisms of schizophrenia.

1. Introduction

Schizophrenia is a prevalent mental disorder affecting approximately 0.5–1% of the world's population. The pathogenesis of schizophrenia is complex including multiple genes and contributing environmental effects that adversely impact neurodevelopment. Nevertheless, a final common result, present in many subjects with schizophrenia, is the disorder of “social brain” and disturbance of functional connectivity in brain is especially emphasized [1].

Early growth response (EGR) genes play important roles in signal transduction in brain including neuronal activation, brain development, and synaptic plasticity [2]. In particular, EGR3 gene has been reported to as a potential susceptibility gene and is abnormally expressed in the brains of schizophrenic patients [3]. EGR3 might be the key regulatory factor in the calcineurin/NFAT signaling pathway. Dysfunction of this pathway led to the development of schizophrenia [4]. EGR3 protein can also regulate a variety of other signaling pathways related to neural and brain development, including nerve growth factor (NGF)-, brain-derived neurotrophic factor (BDNF)- and neuregulin 1 (NRG1)-mediated

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conduction pathways [4–7]. Furthermore, Egr-3 knockout mice are resistant to the adverse effects of antipsychotic drugs [8], which is similar to patients with schizophrenia.

Great interest has been focused on brain structure, functional connectivity and metabolism of individuals with schizophrenia. It was reported that the volume of bilateral hippocampus, especially left hippocampus was significantly reduced in schizophrenia [9–12]. Decrease in NAA (N-acetylaspartate) level was observed in multiple brain regions of patients with chronic schizophrenia including prefrontal cortex, thalamus, cingulate gyrus, hippocampus, which may be related to the presence of mitochondrial dysfunction [13–15]. Thalamus plays a key role in regulation of emotional experience and expression. Both NAA concentration and NAA/Cr ratio were significantly reduced in thalamus of patients with schizophrenia [16]. Our previous study also revealed the abnormal accumulation of choline (Cho) in hippocampus [17]. Therefore, in this study, hippocampus, prefrontal cortex and thalamus were selected as schizophrenia susceptible brain regions.

The integration of functional magnetic resonance imaging (fMRI) with cognitive and affective neuroscience paradigms enables examination of multiple brain systems and alternations associated with psychiatric diseases [18]. Multivariate neural data provide the basis for assessing interactions in brain networks. Among myriad connectivity measures, Granger causality (GC) has proven to be statistically intuitive, easy to implement, and a viable technique for analyzing fMRI Data [19]. Limited study is available on fMRI study of schizophrenia using Granger causality model.

In the present study, the rat model of schizophrenia was successfully created and characterized by the transfection of lentiviral particles carrying EGR3 gene into hippocampus and dentate gyrus. Granger causal model and time cluster analysis TCA method were used to investigate the function changes in the prefrontal lobe, hippocampus and thalamus associated with schizophrenia. The interaction patterns and functions of prefrontal lobe, hippocampus and thalamus in the schizophrenia rats before and after risperidone therapy were explored.

2. Materials and methods

2.1. Animals and husbandry

A randomized, controlled animal study was conducted in School of Medicine, Peking University, China in accordance with the Guidance Suggestions for the Care and Use of Laboratory Animals, issued by the Ministry of Science and Technology of the People's Republic of China. Twenty-four healthy, male Sprague–Dawley rats (aged 4 weeks, weighing 100 ± 10 g) were purchased from Vital River, Beijing, China (license no. SCXK (Jing) 2012-0001). Animals were housed in a temperature- (22–24 °C) and humidity (40–55%)-controlled vivarium.

2.2. Viral vector

Lentiviral particles containing EGR3 gene were purchased from the Fuyishengke Biomedical Scientific Research Service Center, Shanghai, China. The sequence is based on NCBI Reference Sequence: NM_018781.2. The lentivirus was packaged using a four-plasmid system containing psPAX2, pMD2G, pLVX-IRES-ZsGreen1 and PLVX-IRES-ZsGreen1-EGR3. PLVX-IRES-ZsGreen1 can express GFP (green fluorescent protein) as described before [17].

2.3. EGR3 transfection

To initiate the rat schizophrenia model, the lentivirus particle carrying the EGR3 gene was injected bilaterally into the hippocampus and dentate gyrus of rats as previously described [17]. Briefly, the rats were anesthetized by intraperitoneal injection with 10% chloral hydrate (10 ml/kg), and placed in a stereotaxic frame (ST-51600, Kopf Instruments, Tujunga, CA, USA). The skin of the calvarium was sterilized with 75% alcohol, and an incision of about 0.5 cm, was made. After sterilization with 0.05% H_2O_2, the Bregma was exposed. Bilateral holes (0.8 mm) were drilled in the skull above the injection site using a cranial drill (Fig. S1A). The lentivirus particle was slowly injected into each side of the hippocampus (−3.0 mm anterioposterior and ±2.0 mm mediolateral to Bregma; −2.2 mm dorsoventral to the skull surface) using a 1-μL microinjector over 20 min (Fig. S1B). The needles were maintained in position for a further 20 min, and then the incision was sutured. The animals were returned to their home cages when they could move spontaneously. Rats in the sham-surgery group (n = 6) underwent an identical procedure except the lentivirus particle carried green fluorescent protein instead of EGR3. Morris water maze working memory test and the Open Field Test were used to characterize EGR3 transfected schizophrenia model as described before [17].

After a 2-week recovery period, EGR3 transfected rats received intraperitoneal injections of risperidone (0.2 mg/kg; Sigma, St. Louis, MO, USA) for 14 consecutive days. The other three groups received intraperitoneal injections of normal saline at each administration time point corresponding to the risperidone treatment. In summary, four groups were set up in this study with 8–13 replicates in each group including control group (rats without surgery + normal saline), sham group (rats + GFP gene + normal saline), schizophrenia model group (rats + EGR3 gene + normal saline) and RT group (rats + EGR3 gene + risperidone).

2.4. Immunofluorescence staining of rat brain tissues

Immunohistochemistry staining for ERR3 was performed on frozen rat brain tissue sections as described by Schneider et al. [20]. Briefly, The 12–16 mm-thick rat brain cryosections were mounted onto gelatin-coated slides and fixed with paraformaldehyde. The sections were incubated in primary antibody (rat anti-Egr3, 1:1000) (Santa Cruz Biotechnology, USA) for 60 min. The image was acquired using Nikon Ti Eclipse Confocal Microscope (Nikon, Japan). The number of positive cells in the visual filed was counted (n = 6).

2.5. fMRI imaging acquisition

Functional and structural images were acquired on a Phillips 3T Achieva scanner (Philips, The Netherlands) using a standard whole-body rat coil (Shanghai Chenguang Medical Science and Technology, Shanghai, China). Specific methods are as follows: The rats were anesthetized by intraperitoneal injection of 10% chloral hydrate (1.5 ml/kg) before scanning. The rats was placed in prone position and the heads of rats were fixed with a vacuum pillow and band to reduce head movement. T2WI coronal, axial and sagittal images were collected using Spin Echo method (Fig. S2). T2WI image parameters: FOV = 40 × 40 mm, TR = 1696 ms, TE = 96 ms, number of slice = 7, slice thickness = 1.5 mm, layer space = 1.5 mm, NSA = 3. Imaging parameters of fMRI: slice number: 20, interlayer space = 0 mm, TR = 2000 ms, TE = 20 ms, flip angle of 90°, FOV = 40 × 40 mm, NSA = 1.

2.6. Select of schizophrenia susceptible brain area

Bilateral prefrontal lobe, bilateral hippocampus, and bilateral thalamus were selected as regions of interest (Fig. S3).

3. Post-processing methods

3.1. The time clustering (TCA) method and granger causality analysis model (GCA)

TCA method is to determine the time of the maximum and minimum responses in regions of interest (ROIs) of fMRI images [21]. This approach could reduce the noise caused by head movements or unworthy registration. The time and space of max response can be determined without experimental paradigm. If fMRI image has m × n pieces of pixels and there are p images in a sequence, the image sequence can be represented by the following matrix:

$$S = \begin{pmatrix} S_{1,1}S_{1,2}S_{1,3}\cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots S_{1,p} \\ S_{2,1}S_{2,2}S_{2,3}\cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots S_{2,p} \\ S_{3,1}S_{3,2}S_{3,3}\cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots S_{3,p} \\ \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots \cdots \\ S_{m\times n,1}S_{m\times n,2}S_{m\times n,3}\cdots \cdots \cdots \cdots \cdots S_{m\times n,p} \end{pmatrix} \quad (1)$$

TCA principle (1): If the baseline of the ith pixel value is defined as Si, 0 and matrix V has the same dimension with matrix S, Vi, j can be calculated as follows:

$$V_{i,j} = \frac{|S_{i,j} - S_{i,0}|}{S_{i,0}} \quad (1 \le i \le m \times n, 1 \le j \le p) \quad (2)$$

$$W = [W_{i,j}], \quad (1 \le i \le m \times n, 1 \le j \le p): \\ W_{i,j} = \begin{cases} 1, & V_{i,j} = \max\{V_{i,1}, V_{i,2}, V_{i,3}, \ldots \ldots V_{i,p}\} \\ 0, & \text{otherwise} \end{cases} \quad (3)$$

Equation (3) can be used to select a set of pixels which reaches the maximum at the same time. In addition, one-dimensional time change of the time cluster size can be calculated using matrix W and K:

$$K = (K_1, K_2, K_3, \ldots \ldots K_p), \quad \text{where} \quad K_j = \sum_{i=1}^{m\times n} W_{i,j} \quad (4)$$

Firstly, the maximum number of pixels of different slices was counted and the mean number in each group was calculated (Fig. S4). The vertical axis is maximum pixels' number, the horizontal axis is the number of slice. In this study, ROIs are in 9th, 10th, 13th and 14th slices. We calculated the average number of maximum pixels in ROIs, which is the average signal strength of the ROIs. S10T1-4 in Fig. S5 showed the average number of maximum pixels in 0–190 time points of the 10th slice.

3.1.1. Granger causal model approach

Two time series sequences X and Y of fMRI images in brain ROIs were extracted and the time series sentence order was assembled using BIC (Bayesian Information Criterion) [22,23]. The regression model was then established using the following equation:

$$X_t = \sum_{i=1}^{p} a_{1i}X_{t-i} + \varepsilon_{1t}$$
$$Y_t = \sum_{i=1}^{p} b_{1i}Y_{t-i} + \varepsilon_{2t}$$

$\varepsilon_{it}, i = 1,2,3,4$ represents the prediction error, p is the model order.

The joint auto-regression model was also established:

$$X_t = \sum_{i=1}^{p} a_{2i}X_{t-i} + \sum_{i=1}^{p} c_{2i}Y_{t-i} + \varepsilon_{3t}$$
$$Y_t = \sum_{i=1}^{p} b_{2i}Y_{t-i} + \sum_{i=1}^{p} d_{2i}X_{t-i} + \varepsilon_{4t}$$

$\varepsilon_{it}, i = 1,2,3,4$ represents the prediction error, p is the model order. Granger causes is calculated using the following equation [24]:

$$F_{y2x} = \ln\left(\frac{\mathrm{var}(\varepsilon_{1t})}{\mathrm{var}(\varepsilon_{3t})}\right)$$

$\mathrm{var}(\varepsilon_{it}), i = 1,3$ represents the prediction error variance.

Finally, one-sample T test was conducted for F_{v2x}. When $F_{v,2x}>0$, Y is Granger reason to X; When $F_{v,2x} = 0$, Y is not Granger cause to X. Similarly, whether Y is Granger cause to X or not is determined.

3.2. Statistical analysis

Differences among the four groups were analyzed using one-way ANOVA followed by LSD test. $p < 0.05$ was considered to be statistically different.

4. Results

4.1. Overexpression of EGR3 in dentate gyrus of rat brain

The expression of EGR3 gene in dentate gyrus region of rat brain was analyzed at one week after the lentivirus injection. Our results showed that EGR3 positive cell count was 2.67 times as the GFP transfection sham group which indicated that over-expression of ERG3 rat model had been successfully constructed (Fig. 1A and B).

4.2. EGR3 transfected rat model of schizophrenia verification result

We used Morris water maze and open field experiment to test the behavior of rats with EGR3 gene transfection. Morris water maze was used to estimate working memory ability of rats and working memory reflects individual cognitive function. The results showed the escape latency time in rats with EGR3 gene transfection was significantly longer than control group and the sham group ($p < 0.05$), suggesting that EGR3 gene transfection group showed poor working memory capacity. Treatment of risperidone can reverse the phenomenon (Fig. 2A).

Open field experiment was designed to study spontaneous activity in rats. The total central distance can reflect the capability in exploring an unfamiliar environment and adaptability. We found that the central distance in 30 min in model group mice was significantly less than the normal group ($p < 0.05$) which indicated the decrease of the capability in exploring an unfamiliar environment. The distance of risperidone treated group were increased, which indicated that risperidone has the therapeutic effect (Fig. 2B).

These results indicated that EGR3 gene transfection rat model of schizophrenia has the key clinical characteristics of schizophrenia patients.

4.3. The average signal intensity measured by TCA in prefrontal lobe, hippocampus and thalamus

TCA showed that the gray value in prefrontal lobe and hippocampus were statistically different among the four groups ($F = 398.323$, $p = 0.000$ and $F = 414.100$, $p = 0.000$, respectively)

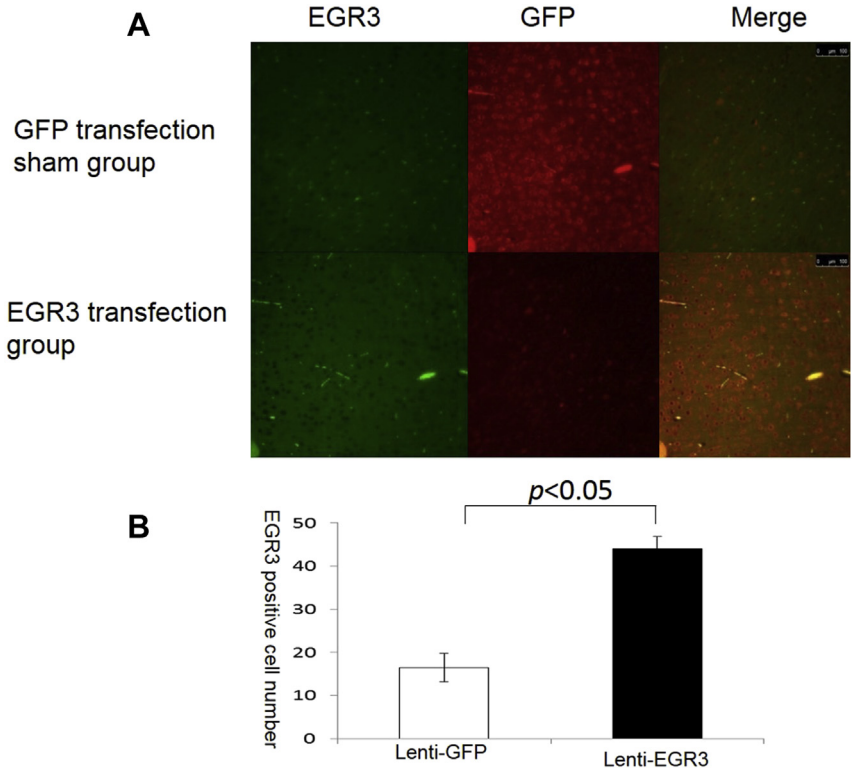

Fig. 1. The expression of EGR3 in dentate gyrus region of rat brain.

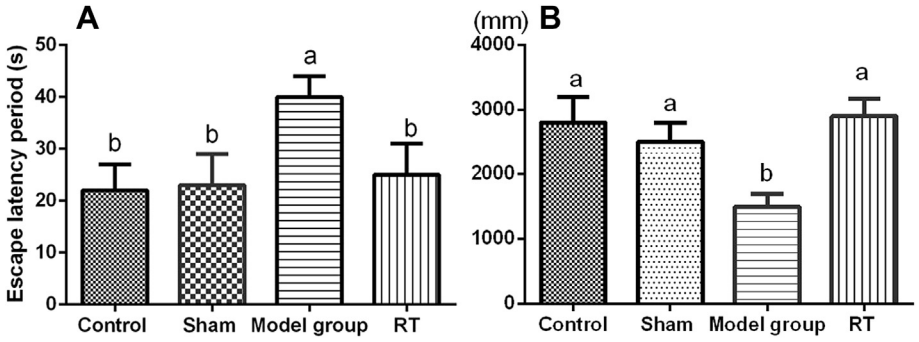

Fig. 2. Schizophrenia-like behavior abnormalities in rats transfected with EGR3 gene. A: Morris water maze test for spatial working memory capacity B: Open filed test for spontaneous activity. Data was mean ± SD (N = 8–13). Group indexed by different letters indicated significant difference ($p < 0.05$) according to one-way ANOVA followed by Tukey's test. Model group: rats with EGR3 transfection. RT group: schizophrenia-like rats treated with risperidone. Sham group: rats with GFP gene transfection and treated with normal saline. Control group: rats without gene transfection and treated with normal saline.

Table 1
Comparison of the average signal intensity value in different brain regions.

Group	Average signal intensity value		
	Prefrontal lobe	Hippocampus	Thalamus
Model group	67.03 ± 7.327[a]	76.45 ± 3.284[a]	73.67 ± 8.715[a]
RT group	39.05 ± 4.652[b]	46.66 ± 9.182[b]	81.17 ± 8.903[a]
Sham group	28.24 ± 5.973[c]	38.46 ± 4.142[c]	69.17 ± 2.222[a]
Control group	40.04 ± 8.942[b]	51.46 ± 2.051[b]	71.50 ± 9.072[a]

Data was collected by fMRI followed by TCA method. Model group: rats with EGR3 transfection. RT group: schizophrenia-like rats treated with risperidone. Sham group: rats with GFP gene transfection and treated with normal saline. Control group: rats without gene transfection and treated with normal saline. Data indexed by different letters in the same column indicated significant difference (n = 6, $p < 0.05$).

(Table 1). The order of average signal intensity in four groups was model group > control group > RT group > sham group. No statistical difference was observed in gray value of thalamus ($F = 2.198$, $p = 0.120$). These results indicated that prefrontal lobe and hippocampus might be the susceptible regions of schizophrenia (see Tables 2).

Table 2
Changes of average signal strength in brain regions after risperidone treatment.

Group	Changes of average signal strength	
	Model group	RT group
Prefrontal lobe	7.00 ± 8.585	−27.84 ± 13.377*
Hippocampus	7.02 ± 25.074	−29.66 ± 28.472*

*: $p < 0.05$ compared with model group (n = 6). Model group: rats with EGR3 transfection. RT group: schizophrenia-like rats treated with risperidone.

4.4. Granger causality connection analysis

We then analyzed the Granger causality connection in prefrontal lobe, hippocampus and thalamus in four groups (Fig. 3 and Table 3). In control group without EGR3 transfection, one-way Granger causality connection was observed from prefrontal cortex to hippocampus and from hippocampus to thalamus (Fig. 3A). Similarly, there were one-way Granger causality connections from hippocampus to prefrontal lobe and from hippocampus to thalamus in sham group (Fig. 3B). However, in EGR3 transfection schizophrenia model group, there was bidirectional Granger causality connection between prefrontal lobe and hippocampus. One-way Granger causality connections are present from thalamus to prefrontal lobe and from hippocampus to thalamus (Fig. 3C). After resperidone treatment, no bidirectional connections were found between prefrontal lobe and hippocampus. Only one-way connection from hippocampus to prefrontal lobe was present (Fig. 3D). There were bidirectional Granger causality connections from hippocampus to thalamus.

5. Discussion

5.1. Functional changes of frontal lobe in rat model of schizophrenia

Attention disorder is divided into auditory and visual attention disorder. Attention disorder can cause difficulties in processing information and is prevalent in patients with schizophrenia. It is a core cognitive dysfunction and might affect other cognitive activities [25]. Now, attention disorder is considered to be related to the functional damage of frontal lobe [26].

Previous studies showed that the role of the prefrontal cortex in working memory is quite complex, including memory attention, inhibition, management, integration and other functions [27]. It was reported that working memory training is the training of the central system related to working memory, which activates prefrontal lobe [28]. Neuropsychological evidence showed that prefrontal cortex is closely related to implementation process. Patients with the damage of prefrontal lobe display the decrease of capacity related to judging, organization, planning, decision making, behavioral inhibition and mental damage [29].

In this study, we found that the gray value in the prefrontal lobe was statistically different among the four groups. The rank order of gray value is model group > control group > risperidone group > sham group. The increase of gray value in prefrontal lobe activity in model group indicated that excessive hyperactivity might lead to attention disorders. Activity of prefrontal lobe was decreased in model group after risperidone treatment and lower than control group. Clinical symptoms of schizophrenia disappeared after risperidone treatment, suggesting that attention disorder was improved. These results also confirmed that prefrontal lobe plays an important role in schizophrenia. Enhanced activities in prefrontal lobe might be related to the pathogenesis of schizophrenia. We also found that bilateral frontal activity was suppressed in sham group, which might be related to trauma.

Table 3
Granger causality connection in prefrontal lobe (F), hippocampus (H) and thalamus (T) in four groups.

Groups	F and H		F and T		H and T	
	F->H	H->F	F->T	T->F	H->T	T->H
Sham group	–	0.17	–	–	0.2	–
RT group	–	0.17	–	0.17	0.1	0.1
Model group	0.12	0.63	0.14	–	0.15	–
Control group	0.11	–	–	–	0.13	–

5.2. Functional changes of hippocampus in schizophrenia rats

Patients with schizophrenia have extensive memory disorders and function defects of hippocampus [30]. Hippocampus is considered to be the brain area related to information storage [31]. Working memory dysfunction is an important part of memory dysfunction in schizophrenia. It takes much longer time for coding

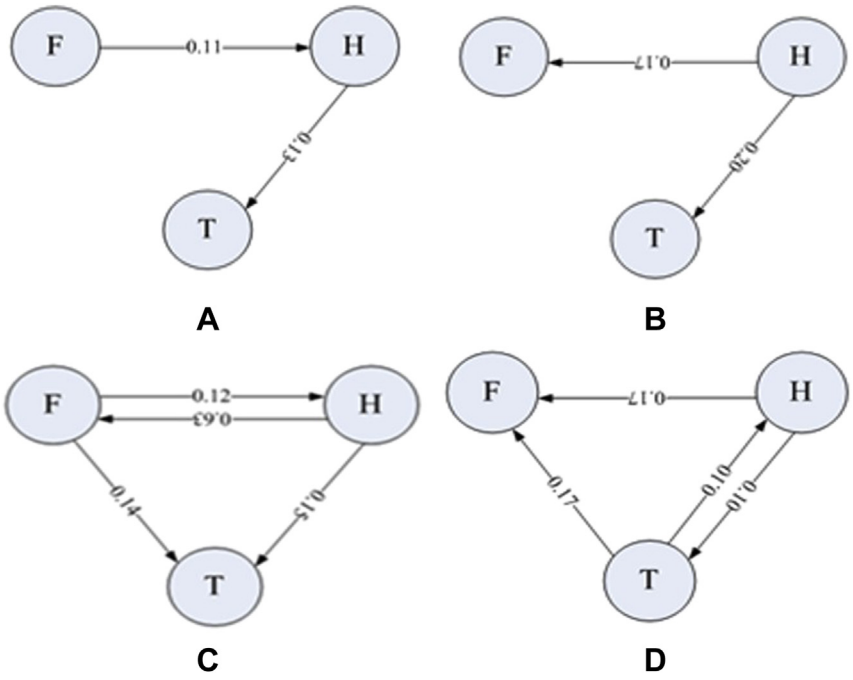

Fig. 3. The Granger causality connection in prefrontal lobe (F), hippocampus (H) and thalamus (T). Note: A: control group; B: sham group; C:model group; D: RT group. Model group: rats with EGR3 transfection. RT group: schizophrenia-like rats treated with risperidone. Sham group: rats with GFP gene transfection and treated with normal saline. Control group: rats without gene transfection and treated with normal saline.

information in schizophrenia model group and is less accurate than the control group in completing difficult tasks [32,33]. Executive function is the high-level cognitive processes and plays a role of coordination in completing a specific task [34].

We found that gray value in the hippocampus in schizophrenia model group was greater than any other groups. The rank order of gray value is schizophrenia model group > control group > risperidone group > sham group. These results indicated that excessive hyperactivity in hippocampus might lead to schizophrenia.

5.3. Thalamus functional changes in schizophrenic rats

NAA concentration decreased in multiple brain regions of patients with chronic schizophrenia such as prefrontal cortex, thalamus, cingulate gyrus and hippocampus [13]. NAA, NAA/Cr values of thalamus, as an important brain area related to emotional experience and expression, were reduced in patients with schizophrenia [16]. However, there is no statistical difference in NAA/Cr value of thalamus in model group compared with other groups. There is no significant changes in thalamic function after treated with risperidone. This indicates that the metabolic changes might occur prior to the changes of function.

The activity of prefrontal lobe and hippocampus was enhanced in schizophrenia model group. After treatment with risperidone, the activity was suppressed. The symptoms related to attention and memory disorders were also restored. Therefore, the enhanced activity in prefrontal lobe and hippocampus might be the indicator for the diagnosis of schizophrenia.

Conflict of interest

The authors declare that there are no conflicts of interest.

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Transparency document

Transparency document related to this article can be found online at http://dx.doi.org/10.1016/j.bbrc.2015.03.089.

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.bbrc.2015.03.089.

References

[1] K. Schnell, Mentalizing functions provide a conceptual link of brain function and social cognition in major mental disorders, Psychopathology 47 (2014) 408–416.
[2] K.J. O'Donovan, W.G. Tourtellotte, J. Millbrandt, J.M. Baraban, The EGR family of transcription-regulatory factors: progress at the interface of molecular and systems neuroscience, Trends Neurosci. 22 (1999) 167–173.
[3] K. Yamada, D.J. Gerber, Y. Iwayama, T. Ohnishi, H. Ohba, T. Toyota, J. Aruga, Y. Minabe, S. Tonegawa, T. Yoshikawa, Genetic analysis of the calcineurin pathway identifies members of the EGR gene family, specifically EGR3, as potential susceptibility candidates in schizophrenia, Proc. Natl. Acad. Sci. U. S. A. 104 (2007) 2815–2820.
[4] L.C. Eldredge, X.M. Gao, D.H. Quach, L. Li, X. Han, J. Lomasney, W.G. Tourtellotte, Abnormal sympathetic nervous system development and physiological dysautonomia in Egr3-deficient mice, Development 135 (2008) 2949–2957.
[5] D.S. Roberts, Y. Hu, I.V. Lund, A.R. Brooks-Kayal, S.J. Russek, Brain-derived neurotrophic factor (BDNF)-induced synthesis of early growth response factor 3 (Egr3) controls the levels of type A GABA receptor alpha 4 subunits in hippocampal neurons, J. Biol. Chem. 281 (2006) 29431–29435.
[6] A.R. Santos, C.B. Duarte, Validation of internal control genes for expression studies: effects of the neurotrophin BDNF on hippocampal neurons, J. Neurosci. Res. 86 (2008) 3684–3692.
[7] C. Jacobson, D. Duggan, G. Fischbach, Neuregulin induces the expression of transcription factors and myosin heavy chains typical of muscle spindles in cultured human muscle, Proc. Natl. Acad. Sci. U. S. A. 101 (2004) 12218–12223.
[8] A. Gallitano-Mendel, D.F. Wozniak, E.A. Pehek, J. Milbrandt, Mice lacking the immediate early gene Egr3 respond to the anti-aggressive effects of clozapine yet are relatively resistant to its sedating effects, Neuropsychopharmacology 33 (2008) 1266–1275.
[9] A. Deleon, N.C. Patel, M.L. Crismon, Aripiprazole: a comprehensive review of its pharmacology, clinical efficacy, and tolerability, Clin. Ther. 26 (2004) 649–666.
[10] H. Matsumoto, A. Simmons, S. Williams, R. Pipe, R. Murray, S. Frangou, Structural magnetic imaging of the hippocampus in early onset schizophrenia, Biol. Psychiatry 49 (2001) 824–831.
[11] L. De Peri, A. Crescini, G. Deste, P. Fusar-Poli, E. Sacchetti, A. Vita, Brain structural abnormalities at the onset of schizophrenia and bipolar disorder: a meta-analysis of controlled magnetic resonance imaging studies, Curr. Pharm. Des. 18 (2012) 486–494.
[12] M. Juuhl-Langseth, L.M. Rimol, I.A. Rasmussen Jr., R. Thormodsen, A. Holmén, K.E. Emblem, P. Due-Tønnessen, B.R. Rund, I. Agartz, Comprehensive segmentation of subcortical brain volumes in early onset schizophrenia reveals limited structural abnormalities, Psychiatry Res. 203 (2012) 14–23.
[13] M.A. Reid, L.E. Stoeckel, D.M. White, K.B. Avsar, M.S. Bolding, N.S. Akella, R.C. Knowlton, J.A. den Hollander, A.C. Lahti, Assessments of function and biochemistry of the anterior cingulate cortex in schizophrenia, Biol. Psychiatry 68 (2010) 625–633.
[14] N.V. Kraguljac, M. Reid, D. White, R. Jones, J. den Hollander, D. Lowman, A.C. Lahti, Neurometabolites in schizophrenia and bipolar disorder – a systematic review and meta-analysis, Psychiatry Res. 203 (2012) 111–125.
[15] L.S. Kegeles, X. Mao, A.D. Stanford, R. Girgis, N. Ojeil, X. Xu, R. Gil, M. Slifstein, A. Abi-Dargham, S.H. Lisanby, D.C. Shungu, Elevated prefrontal cortex gamma-aminobutyric acid and glutamate-glutamine levels in schizophrenia measured in vivo with proton magnetic resonance spectroscopy, Arch. Gen. Psychiatry 69 (2012) 449–459.
[16] J.R. Bustillo, L.M. Rowland, P. Mullins, R. Jung, H. Chen, C. Qualls, R. Hammond, W.M. Brooks, J. Lauriello, 1H-MRS at 4 tesla in minimally treated early schizophrenia, Mol. Psychiatry 15 (2010) 629–636.
[17] G. Ma, T. Song, M. Chen, Y. Fu, Y. Xu, E. Ma, W. Wang, J. Du, M. Huang, Hippocampal and thalamic neuronal metabolism in a putative rat model of schizophrenia, Neural Regen. Res. 15 (2013) 2415–2423.
[18] F. Barkhof, S. Haller, S.A. Rombouts, Resting-state functional MR imaging: a new window to the brain, Radiology 272 (2014) 29–49.
[19] X. Wen, G. Rangarajan, M. Ding, Is Granger causality a viable technique for analyzing fMRI data? PLoS One 8 (2013) e67428.
[20] E.M. Schneider Gasser, C.J. Straub, P. Panzanelli, O. Weinmann, M. Sassoè-Pognetto, J.M. Fritschy, Immunofluorescence in brain sections: simultaneous detection of presynaptic and postsynaptic proteins in identified neurons, Nat. Protoc. 1 (2006) 1887–1897.
[21] Y. Liu, J.H. Gao, H.L. Liu, F.T. Peter, The temporal response of the brain after eating revealed by functional MRI, Nature 405 (2000) 1058–1062.
[22] C.W.J. Granger, Investigating causal relations by econometric models and cross-spectral methods, Econometrica 37 (1969) 424–438.
[23] C.W.J. Granger, Testing for causality: a personal viewpoint, J. Econ. Dynam. Control 2 (1980) 329–352.
[24] L. Harrison, W.D. Penny, K. Friston, Multivariate autoregressive modeling of fMRI time series, Neuroimage 19 (2003) 1477–1491.
[25] A.S. Chan, S.L. Sze, Y.M. Han, M.C. Cheung, A chan dietary intervention enhances executive functions and anterior cingulate activity in autism spectrum disorders: a randomized controlled trial, Evid. Based Complement. Altern. Med. 2012 (2012) 262136.
[26] M. Fioravanti, O. Carlone, B. Vitale, M.E. Cinti, L. Clare, A meta-analysis of cognitive deficits in adults with a diagnosis of schizophrenia, Neuropsychol. Rev. 15 (2005) 73–95.
[27] D. Luck, J.R. Foucher, I. Offerlin-Meyer, M. Lepage, J.M. Danion, Assessment of single and bound features in a working memory task in schizophrenia, Schizophr. Res. 100 (2008) 153–160.
[28] H. Westerberg, T. Klingberg, Changes in cortical activity after training of working memory–a single-subject analysis, Physiol. Behav. 92 (2007) 186–192.
[29] L. Yang, S. Chen, C.M. Chen, F. Khan, G. Forchelli, D.C. Javitt, Schizophrenia, culture and neuropsychology: sensory deficits, language impairments and social functioning in Chinese-speaking schizophrenia patients, Psychol. Med. 42 (2012) 1485–1494.
[30] M. Krausz, S. Moritz, M. Lambert, D. Naber, Dosage of conventional neuroleptic medication and subjective cognitive functioning in schizophrenia, Int. Clin. Psychopharmacol. 15 (2000) 77–81.
[31] F. Vargha-Khadem, D.G. Gadian, K.E. Watkins, A. Connelly, W. Van Paesschen, M. Mishkin, Differential effects of early hippocampal pathology on episodic and semantic memory, Science 277 (1997) 376–380.
[32] M. Hartman, M.C. Steketee, S. Silva, K. Lanning, H. McCann, Working memory and schizophrenia: evidence for slowed encoding, Schizophr. Res. 59 (2003) 99–113.
[33] M. Stone, J.D. Gabrieli, G.T. Stebbins, E.V. Sullivan, Working and strategic memory deficits in schizophrenia, Neuropsychology 12 (1998) 278–288.
[34] S. Funahashi, Neuronal mechanisms of executive control by the prefrontal cortex, Neurosci. Res. 39 (2001) 147–165.