ELSEVIER

Clinical Neuroscience Research 5 (2005) 15–21

Clinical Neuroscience Research

www.elsevier.com/locate/clires

Gene expression changes in schizophrenia: how do they arise and what do they mean?

David A. Lewis[a,*], Károly Mirnics[b], Pat Levitt[c]

[a]*Departments of Psychiatry and Neuroscience, University of Pittsburgh School of Medicine, W-1651 Biomedical Science Tower, 3811 O'Hara Street, Pittsburgh, PA 15213, USA*

[b]*Departments of Psychiatry and Neurobiology, University of Pittsburgh School of Medicine, Pittsburgh, PA 15213, USA*

[c]*Vanderbilt Kennedy Center for Research on Human Development and Department of Pharmacology (PL), Vanderbilt University, Nashville, TN 37203, USA*

Abstract

The advent of different methods for quantifying messenger RNAs has made it possible to assess the tissue levels of the transcription products of virtually every gene expressed in the human brain, and to determine whether the expression of each gene is altered in psychiatric disorders such as schizophrenia. However, these advances have raised a number of interpretive questions, including what causes disease-related mRNA expression changes and what such changes mean for the function of the affected brain circuits. In this paper, we consider possible answers to these questions for two genes, Regulator of G Protein Signaling 4 (RGS4) and the 67 kilodalton isoform of glutamic acid decarboxylase (GAD_{67}), both of which have been found to be under-expressed in the prefrontal cortex of subjects with schizophrenia.

Keywords: gene expression; glutamic acid decarboxylase; regulator of G-protein signaling 4; schizophrenia

1. Introduction

In recent years, the advent of a variety of new methods for quantifying messenger RNA (mRNA), the transcription product of genomic DNA, has made it possible to assess tissue levels of virtually every transcript expressed in the brain. Available techniques range from in situ hybridization, which is ideal for determining the cellular specificity and precise level of expression of individual transcripts, to DNA microarrays which can, in theory, simultaneously survey the relative expression level of all mRNAs (i.e., the transcriptome) in a tissue sample. These approaches have been applied to the study of postmortem brain specimens from subjects with schizophrenia, resulting in a number of interesting findings, with some of these independently replicated across different subject cohorts. Differences between subject groups in tissue levels of a given mRNA have been predominantly interpreted to mean that the change arises as an active regulation of the transcript. For the purposes of this paper, that same assumption will be followed, although it must be kept in mind that differences in transcript tissue levels could instead reflect changes in mRNA stability or degradation.

Although these findings are quite exciting, understanding their significance in the context of the disease process of schizophrenia requires careful consideration. Thus, the aim of this paper is to explore the two questions raised in the title. First, what factors might be responsible for disease-related transcript changes? For example, since the substantial heritability of schizophrenia appears to be polygenic in nature [1], some transcript differences might be due to DNA sequence variants that confer disease susceptibility through changes in the expression level of the encoded mRNA. Alternatively, certain transcript abnormalities might represent secondary responses of the genome that are either deleterious or compensatory. Finally, some differences in mRNA levels observed in postmortem brain samples might reflect changes in gene expression induced by the treatment of the illness, or result from other factors (e.g. co-morbid depression, substance abuse, nicotine exposure, suicide, etc.) that frequently accompany schizophrenia. Clearly, distinguishing among these alternatives is

* Corresponding author. Tel.: +1 412 624 3934; fax: +1 412 624 9910.
E-mail address: lewisda@upmc.edu (D.A. Lewis).
URL: www.tnp.pitt.edu.

1566-2772/$ - see front matter © 2005 Association for Research in Nervous and Mental Disease. Published by Elsevier B.V. All rights reserved.
doi:10.1016/j.cnr.2005.07.002

critical for assessing whether a given transcript alteration represents a cause, consequence or compensation in the disease process.

Second, what is the meaning of a given change in transcript expression for the functional circuitry of the affected brain region? Any observed difference in the tissue level of a given transcript could reflect a change in the number of the cells that normally express that mRNA, a uniform expression change in all cells that express the transcript, or an alteration in expression restricted to a subpopulation of neurons [2]. Clearly, each of these scenarios has markedly different implications for the circuitry in which the affected neurons participate.

Consequently, this article considers each of these questions in the context of two genes, Regulator of G Protein Signaling 4 (RGS4) and the 67 kilodalton isoform of glutamic acid decarboxylase (GAD_{67}), both of which have been found to be under-expressed in the prefrontal cortex of subjects with schizophrenia.

2. Factors that might contribute to altered gene expression in schizophrenia

In an initial microarray study of the prefrontal cortex in schizophrenia, we found a marked and highly consistent reduction in the RGS4 transcript [3]. This finding was particularly interesting given the critical role that RGS proteins play in controlling signaling through G-protein coupled receptors (GPCRs). The effects of a number of neurotransmitters (e.g. dopamine, serotonin and glutamate) that are thought to play a critical role in the pathophysiology or pharmacotherapy of schizophrenia are mediated by GPCRs. In the absence of their ligand, the cytoplasmic domain of these receptors is tightly bound to a heterotrimeric G protein composed of subtypes of G_α, G_β and G_γ subunits (Fig. 1). The binding of a specific ligand to a GPCR produces a conformational change in the structure of the receptor, dissociation of the G-protein and the rapid replacement of guanine diphosphate (GDP) with guanine triphosphate (GTP) at the G_α subunit. As the GTP-bound G_α has a low affinity for the $G_{\beta\gamma}$ dimer, it dissociates, allowing the independent subunits to interact with a variety of cellular effectors, thus activating or inhibiting a variety of signaling cascades. Once the G_α-bound GTP is hydrolyzed to GDP, the G_α-GDP complex regains its high affinity to $G_{\beta\gamma}$ and the heterotrimeric G-protein re-associates. This effectively ends signaling via the GPCR, and prepares the receptor for a new round of ligand binding. The spontaneous hydrolysis of G_α-bound GTP to GDP is slow, and thus limits dynamic signaling via the receptor. RGS proteins are GTPase-activating proteins that markedly facilitate the hydrolysis of G_α-bound GTP and thus play a critical role in determining the duration and timing of signaling through GPCRs [4].

To determine whether the decreased levels of RGS4 mRNA in schizophrenia reflected a specific change in this component of G-protein signaling, we analyzed the microarray data for gene expression changes in other RGS family members. Of the eleven RGS family members represented on the microarrays, only RGS4 reported

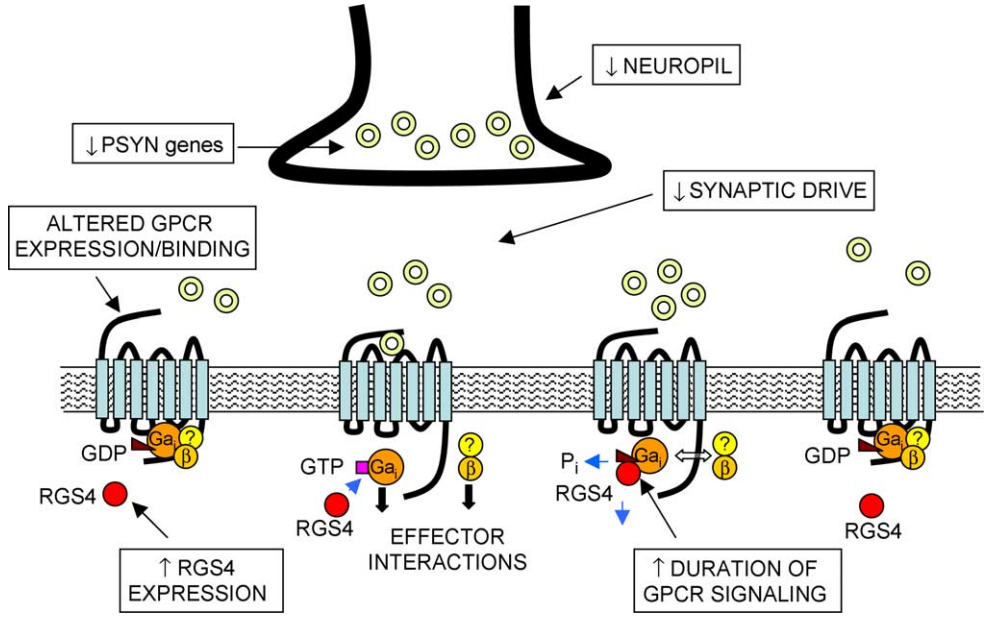

Fig. 1. RGS4 modulation of G-protein coupled receptor (GPCR) signaling in the context of schizophrenia-related findings. In schizophrenia, reduced expression of genes encoding proteins involved in the machinery of neurotransmitter release (PSYN) and/or a decreased number of synapses [54] might result in a compromised synaptic drive and altered GPCR expression or binding. In an attempt to compensate for the impaired synaptic drive, cellular levels of RGS4 levels are down-regulated. As RGS4 accelerates the hydrolysis of Gα -bound GTP to GDP + Pi, decreased RGS4 levels could increase the duration of signaling from the receptor, thus prolonging the signaling via the effector cascade. Alternatively, reduced levels of RGS4 might reflect allelic variants in the regulatory portion of this gene, and the other schizophrenia-associated changes in synaptic function secondary consequences.