Psychoneuroendocrinology (2015) 51, 472—494

Available online at www.sciencedirect.com

ScienceDirect

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

Blood-based gene-expression biomarkers of post-traumatic stress disorder among deployed marines: A pilot study

Daniel S. Tylee[a], Sharon D. Chandler[b], Caroline M. Nievergelt[c,d,e], Xiaohua Liu[b,i], Joel Pazol[b], Christopher H. Woelk[d,f], James B. Lohr[c,d,e], William S. Kremen[b,c,e], Dewleen G. Baker[c,d,e], Stephen J. Glatt[a,*], Ming T. Tsuang[b,c,d,e,g,h], Marine Resiliency Study Investigators[1]

[a] *Psychiatric Genetic Epidemiology & Neurobiology Laboratory (PsychGENe Lab), Departments of Psychiatry and Behavioral Sciences & Neuroscience and Physiology, Medical Genetics Research Center, SUNY Upstate Medical University, Syracuse, NY, USA*
[b] *Center for Behavioral Genomics, Department of Psychiatry, University of California, La Jolla, San Diego, CA, USA*
[c] *VA Center of Excellence for Stress and Mental Health, San Diego, CA, USA*
[d] *Veterans Affairs San Diego Healthcare System, San Diego, CA, USA*
[e] *Department of Psychiatry, University of California, La Jolla, San Diego, CA, USA*
[f] *Department of Medicine, University of California, La Jolla, San Diego, CA, USA*
[g] *Institute of Genomic Medicine, University of California, La Jolla, San Diego, CA, USA*
[h] *Harvard Institute of Psychiatric Epidemiology and Genetics, Department of Epidemiology, Harvard School of Public Health, Boston, MA, USA*
[i] *Shanghai Mental Health Center, Shanghai Jiao Tong University School of Medicine, Shanghai, China*

Received 14 April 2014; received in revised form 20 August 2014; accepted 22 September 2014

* Corresponding author. Tel.: +1 315 464 7742; fax: +1 315 464 7744.
E-mail address: glatts@upstate.edu (S.J. Glatt).

[1] The Marine Resiliency Study Investigators include Brett T. Litz (Veterans Affairs Boston Healthcare System, Boston, Massachusetts and Boston University); William P. Nash (University of California-San Diego); Mark A. Geyer (University of California-San Diego and VA Center of Excellence for Stress and Mental Health); Paul S. Hammer (Defense Centers of Excellence for Stress and Mental Health and Traumatic Brain Injury, Arlington, Virginia); Gerald E. Larsen (Naval Health Research Center, San Diego, California); Daniel T. O'Connor (University of California-San Diego); Victoria B. Risbrough (VA Center of Excellence for Stress and Mental Health San Diego and University of California-San Diego); Nicholas J. Schork (Scripps Translational Science Institute, La Jolla, California); Jennifer J. Vasterling (Veterans Affairs Boston Healthcare System, Boston, Massachusetts and Boston University); and Jennifer A. Webb-Murphy (Naval Center Combat and Operational Stress Control, San Diego, California).

http://dx.doi.org/10.1016/j.psyneuen.2014.09.024

KEYWORDS
Alternative splicing;
mRNA;
Peripheral blood mononuclear cells;
Microarray;
Transcriptome;
Trauma;
Diagnosis;
Biomarker;
Antioxidant;
Oxidative stress

Summary: The etiology of post-traumatic stress disorder (PTSD) likely involves the interaction of numerous genes and environmental factors. Similarly, gene-expression levels in peripheral blood are influenced by both genes and environment, and expression levels of many genes show good correspondence between peripheral blood and brain tissues. In that context, this pilot study sought to test the following hypotheses: (1) post-trauma expression levels of a gene subset in peripheral blood would differ between Marines with and without PTSD; (2) a diagnostic biomarker panel of PTSD among high-risk individuals could be developed based on gene-expression in readily assessable peripheral blood cells; and (3) a diagnostic panel based on expression of individual exons would surpass the accuracy of a model based on expression of full-length gene transcripts. Gene-expression levels in peripheral blood samples from 50 U.S. Marines (25 PTSD cases and 25 non-PTSD comparison subjects) were determined by microarray following their return from deployment to war-zones in Iraq or Afghanistan. The original sample was carved into training and test subsets for construction of support vector machine classifiers. The panel of peripheral blood biomarkers achieved 80% prediction accuracy in the test subset based on the expression of just two full-length transcripts (*GSTM1* and *GSTM2*). A biomarker panel based on 20 exons attained an improved 90% accuracy in the test subset. Though further refinement and replication of these biomarker profiles are required, these preliminary results provide proof-of-principle for the diagnostic utility of blood-based mRNA-expression in PTSD among trauma-exposed individuals.

1. Introduction

Post-traumatic stress disorder (PTSD) is a severe anxiety syndrome that is currently diagnosed based on the emergence and persistence of core clinical symptoms including hyperarousal, re-experiencing, avoidance, or emotional numbing for a period greater than one month. Early psychosocial intervention after stress exposure may help reduce some of the symptoms and prevent the development of chronic PTSD (Litz et al., 2002). However, many individuals initially presenting with PTSD-like symptoms recover spontaneously and do not develop chronic PTSD (McFarlane, 1997). Thus, identifying which individuals will benefit most from early intervention can be challenging. Despite possible benefits of early intervention and a growing knowledge of the pathophysiology accompanying PTSD, a readily assessable diagnostic biomarker for PTSD has yet to be validated.

Classical descriptions of PTSD pathophysiology have included dysregulation of the hypothalamic-pituitary-adrenal (HPA) axis, but a specific pattern of dysregulation is not consistently observed across studies. Similarly, heightened inflammatory signaling has been reported in some (but not all) contexts (Baker et al., 2012b). Some have proposed a model of insufficient regulation of inflammatory signaling (Heinzelmann and Gill, 2013). Yet, there is an apparent paradox; *i.e.*, the observation that peripheral blood mononuclear cells (PBMCs) from PTSD patients show increased sensitivity to glucocorticoid-mediated suppression of an *in-situ* inflammatory response (van Zuiden et al., 2012b).

There is considerable evidence that genetic effects, environmental influences, and their interaction play a role in the development of PTSD (Afifi et al., 2010; Koenen et al., 2009; True et al., 1993). There is a well-established body of clinical literature supporting a link between early life events, previous exposure to traumatic stress, and other psychosocial factors with the development of PTSD (Brewin et al., 2000; Ozer et al., 2003; DiGangi et al., 2013). Many biological investigations of PTSD have focused on the HPA axis and glucocorticoid receptor (GR) signaling pathways. In a cross-sectional study, Binder et al. (2008) identified an interaction between child abuse history and genetic polymorphisms in *FKBP5* (a negative regulator of GR sensitivity) in predicting adult PTSD symptomology among a sample of non-psychiatric medical clinic patients. Mehta et al. (2011) described the association between genetic polymorphisms in *FKBP5* and dysregulated neuroendocrine profiles described in PTSD. van Zuiden et al. (2012a) provided evidence that increased GR density is a pre-trauma risk factor for the development of PTSD and that dysregulation of GR density may be associated with an interaction between polymorphisms in the GR gene and concomitant early life stress. Another line of research suggests that genetic variants in corticotropin-releasing hormone type 1 receptor (*CRHR1*) are a risk factor for PTSD in children who were abused at an early age (Gillespie et al., 2009). PTSD is thus thought to be a disorder whose development is influenced by multiple genetic and environmental effects that establish a susceptible biological state; this vulnerability may be reflected in gene-expression signatures.

In light of the less-than-absolute heritability of PTSD and the prominent role of environmental factors, the pursuit of static genetic markers alone (*e.g.*, single nucleotide polymorphisms and copy-number variations) likely will not yield a suitable diagnostic biomarker. Gene-expression (*i.e.*, mRNA) levels, which potentially reflect the effects of both heredity and environment, may be better indicators of the aberrant biology underlying PTSD. PTSD clearly is a brain disorder, but assaying gene-expression levels — either acutely or longitudinally — in the brains of living human subjects at risk for PTSD is impossible. Yet, peripheral blood expression levels of many genes are moderately correlated with