Brain, Behavior, and Immunity xxx (2015) xxx–xxx

Contents lists available at ScienceDirect

Brain, Behavior, and Immunity

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

Discovery of serum biomarkers predicting development of a subsequent depressive episode in social anxiety disorder

M.G. Gottschalk [a], J.D. Cooper [a], M.K. Chan [a], M. Bot [b], B.W.J.H. Penninx [b,*,1], S. Bahn [a,c,*,1]

[a] Department of Chemical Engineering and Biotechnology, Cambridge Centre of Neuropsychiatric Research, University of Cambridge, Cambridge, UK
[b] Department of Psychiatry, EMGO Institute for Health and Care Research and Neuroscience Campus Amsterdam, VU University Medical Center, Amsterdam, The Netherlands
[c] Department of Neuroscience, Erasmus Medical Center, Rotterdam, The Netherlands

ARTICLE INFO

Article history:
Received 16 January 2015
Received in revised form 10 April 2015
Accepted 21 April 2015
Available online xxxx

Keywords:
Social phobia
SAD
Affective disorder
Mood disorder
Prediction
Major depressive disorder
Depression
Risk factors
Prognosis
Secondary depression
Dysthymia

ABSTRACT

Although social anxiety disorder (SAD) is strongly associated with the subsequent development of a depressive disorder (major depressive disorder or dysthymia), no underlying biological risk factors are known. We aimed to identify biomarkers which predict depressive episodes in SAD patients over a 2-year follow-up period. One hundred sixty-five multiplexed immunoassay analytes were investigated in blood serum of 143 SAD patients without co-morbid depressive disorders, recruited within the Netherlands Study of Depression and Anxiety (NESDA). Predictive performance of identified biomarkers, clinical variables and self-report inventories was assessed using receiver operating characteristics curves (ROC) and represented by the area under the ROC curve (AUC). Stepwise logistic regression resulted in the selection of four serum analytes (AXL receptor tyrosine kinase, vascular cell adhesion molecule 1, vitronectin, collagen IV) and four additional variables (Inventory of Depressive Symptomatology, Beck Anxiety Inventory somatic subscale, depressive disorder lifetime diagnosis, BMI) as optimal set of patient parameters. When combined, an AUC of 0.86 was achieved for the identification of SAD individuals who later developed a depressive disorder. Throughout our analyses, biomarkers yielded superior discriminative performance compared to clinical variables and self-report inventories alone. We report the discovery of a serum marker panel with good predictive performance to identify SAD individuals prone to develop subsequent depressive episodes in a naturalistic cohort design. Furthermore, we emphasise the importance to combine biological markers, clinical variables and self-report inventories for disease course predictions in psychiatry. Following replication in independent cohorts, validated biomarkers could help to identify SAD patients at risk of developing a depressive disorder, thus facilitating early intervention.

1. Introduction

Social anxiety disorder (SAD; also referred to as "social phobia") is among the most common anxiety spectrum disorders with a 12-month prevalence ranging between 2% and 7% (Kessler et al., 2012; Wittchen et al., 2011). Defined by a marked fear of social situations, the affected individual avoids situations associated with exposure to possible scrutiny by others (American-Psychiatric-Association, 2013). The age of onset is usually in childhood or adolescence (Ballenger et al., 1998). Despite the associated distress and impairment, only half of the patients fulfilling diagnostic criteria for SAD ever seek help. This results in a median delay of over two decades until correct diagnosis and initial treatment, the longest delay amongst all psychiatric disorders investigated in the US National Comorbidity Survey Replication (Wang et al., 2005). In addition to the characteristic chronic course, SAD patients frequently (20–30%) present with co-morbid major depressive disorder (MDD) Stein et al., 1990; Merikangas and Angst, 1995; Lewinsohn et al., 1997, with SAD being the most prevalent co-morbid anxiety disorder in patients suffering from depressive disorders (Pini et al., 1997; Rush et al., 2005). Co-morbidity is associated with a more severe and chronic disease course and worse clinical outcome (Stein et al., 2001; Beesdo et al., 2007). The vast majority of SAD patients present initially with social anxiety symptoms (Kessler et al., 1999) and develop a co-morbid depression on average within 5 years (Beesdo et al., 2007).

* Corresponding authors at: VU University Medical Center, AJ Ernststraat 1187, 1081 HL Amsterdam, The Netherlands. Tel.: +31 20 788 5437 (B.W.J.H. Penninx). Department of Chemical Engineering and Biotechnology, University of Cambridge, Tennis Court Road, Cambridge CB2 1QT, UK. Tel.: +44 (0) 1223334151 (S. Bahn).
E-mail addresses: b.penninx@vumc.nl (B.W.J.H. Penninx), sb209@cam.ac.uk (S. Bahn).
[1] Joined senior authorship.

http://dx.doi.org/10.1016/j.bbi.2015.04.011
0889-1591/

Consistent with these findings, SAD has been shown to be an important predictor/risk factor of a subsequent depressive disorder independent of the age of onset (Stein et al., 2001; Beesdo et al., 2007). Furthermore, apart from a SAD lifetime history, distinct psychological constructs within the SAD symptom spectrum (e.g. behavioral inhibition) have also been shown to be predictive of the future onset of depression (Beesdo et al., 2007). Other characteristics of anxiety disorders that have been linked to an increased risk of developing a depressive disorder include the level of anxiety-associated impairment (Bittner et al., 2004) and the presence of multiple anxiety disorders (Woodward and Fergusson, 2001) or panic attacks (Goodwin, 2002). However, little is known about the molecular mechanisms involved in the onset of either anxiety or depressive disorders. Changes in cortisol awaking response (Adam et al., 2010) and serum interleukin 6 (Khandaker et al., 2014) have been reported to be predictors for the future onset of depression in adolescence. So far no biomarkers have been associated with the development of depressive episodes in anxiety disorder patients. Awareness of factors that predict increased susceptibility for the onset of a depressive disorder within the SAD patient population could lead to an improved clinical outcome due to early intervention (Kessler et al., 1999).

In the present study, we investigated molecular changes in serum collected from patients diagnosed with SAD without current co-morbid depressive disorders with the objective to identify a molecular biomarker panel aiding in the prediction of the onset of a depressive disorder within a 2-year follow-up period. We analyzed the discriminative power of serum protein changes at the time of baseline clinical assessment of 165 analytes using multiplexed immunoassays. Biomarkers were initially identified in 72 patients diagnosed solely with SAD and the analysis was then expanded to include SAD patients with other co-morbid anxiety disorders (totalling 143 SAD patients) in order to account for multiple anxiety spectrum co-morbidities in SAD individuals. A selection of these candidate biomarkers was combined into an optimized panel of four serum analytes. Finally, we evaluated the predictive performance of the identified biomarker panels alone and in combination with psychiatric and somatic patient variables selected from structured patient assessments in order to determine their potential for clinical application.

2. Subjects and methods

2.1. Study sample

Data collected within NESDA, a longitudinal naturalistic cohort study were used (Penninx et al., 2008). The full baseline dataset comprises 2981 persons aged 18–65 years, including those with lifetime or current anxiety or depressive disorder ($n = 2329$; 78%) and healthy controls ($n = 652$; 22%). Individuals were recruited from the community ($n = 564$; 19%), primary care ($n = 1610$; 54%) and specialized mental health care ($n = 807$; 27%) between September 2004 and February 2007 at clinical sites in Amsterdam, Groningen and Leiden. Recruitment from community samples included sampling from two previously studied cohorts with diagnoses based on the Composite International Diagnostic Interview (CIDI) World-Health-Organization, 1997 outcomes (for more information on CIDI see Section 2.2). Of 662 individuals formerly recruited within the Netherlands Mental Health Survey and Incidence Study (NEMESIS) Bijl et al., 1998, 359 (54%) refused to participate and 303 (46%) were included. The second recruitment involved individuals previously enrolled in the Adolescent at Risk for Anxiety and Depression (ARIADNE) Landman-Peeters et al., 2005 study. 394 former participants were contacted, 11 (3%) could not be traced, 122 (31%) refused participation and 261 (66%) were included into NESDA. For the initial screening in the primary care setting 23750 questionnaires (Kessler-10 Kessler et al., 2003 with additional questions for anxiety spectrum disorders and psychotropic medication) were sent out to general practitioners (GPs) in the vicinity of the clinical sites and 10706 were returned (45%). During the subsequent screening stages 4316 (40%) people refused participation, 4532 (43%) were not contacted (due to random selections in the screened individuals) and 248 (2%) met NESDA exclusion criteria (see Section 2.2), resulting in 1610 (15%) included participants. Of 1413 people contacted for inclusion based on structured psychiatric interviews in outpatient clinics around the clinical sites, 606 (45%) refused and 807 (57%) agreed to participate in NESDA. See Penninx et al. (2008) for more details on the NESDA sample and recruitment process. Ethical approval was granted by local ethic boards of all participating centres and all participants gave written informed consent. Exclusion criteria included a clinical diagnosis of any psychotic disorder, bipolar disorder, severe addictive disorder, obsessive compulsive disorder or non-fluency in Dutch. Baseline data were collected in a 4 h interview, gathering information on clinical psychopathology, psychiatric characteristics, use of medication and sociodemographics as well as physical and psychosocial testing. Additionally the baseline evaluation included an overnight fasting blood draw. Processed serum samples were stored immediately at −80 °C until further use (Penninx et al., 2008). A 2-year follow-up assessment was conducted with a response rate of 87.1%.

In the presented study, we used blood analyte abundance information available for a subset of NESDA individuals ($n = 1840$, 62% of the total baseline assessment), who participated in the 2-year follow-up and had sufficient serum available at baseline (approximately 1 mL) for multiplexed immunoassay analysis. For our study, we included only individuals with a baseline diagnosis of SAD during the 6 months prior to sampling. We excluded SAD patients with a current depressive disorder at baseline (6 months recency) to be able to investigate subsequent depressive disorder onset or recurrence. This resulted in 143 SAD patients, of whom 72 patients had pure SAD and 71 patients had a diagnosis of SAD and another co-morbid anxiety disorder.

2.2. Diagnoses, clinical characteristics and self-report inventories

Diagnoses of anxiety disorders [SAD, panic disorder with agoraphobia (PDA), panic disorder without agoraphobia (PD), agoraphobia (AP) and generalized anxiety disorder (GAD)] and depressive disorders (major depressive disorder (MDD) and dysthymia) were established using the Composite International Diagnostic Interview (CIDI) lifetime version 2.1 (World-Health-Organization, 1997) by specially trained clinical research staff. This instrument has been used worldwide for WHO field research and has been shown to possess high validity for the detection of anxiety and depressive disorders (Wittchen et al., 1989; Wittchen, 1994) as well as high inter-rater (Wittchen et al., 1991) and test–retest reliability (Wacker et al., 2006). The CIDI assessment was used again to evaluate the course of disorders after 2 years, determining the presence of a DSM-IV classified anxiety or depressive disorder during the period between baseline and follow-up. Severity of anxiety-related symptoms at baseline was assessed with the Fear Questionnaire (FQ) Marks and Mathews, 1979 and the Beck Anxiety Inventory (BAI) Beck et al., 1988. Severity of depression-related symptoms was assessed with the Inventory of Depressive Symptomatology (IDS) Rush et al., 1996. Psychotherapy information was based on self-reported participation in any form of psychiatrist- or psychotherapist-guided psychotherapy during the

Please cite this article in press as: Gottschalk, M.G., et al. Discovery of serum biomarkers predicting development of a subsequent depressive episode in social anxiety disorder. Brain Behav. Immun. (2015), http://dx.doi.org/10.1016/j.bbi.2015.04.011