Neurobiology of Disease 46 (2012) 88–92

Contents lists available at SciVerse ScienceDirect

Neurobiology of Disease

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

MeCP2+/− mouse model of RTT reproduces auditory phenotypes associated with Rett syndrome and replicate select EEG endophenotypes of autism spectrum disorder

Wenlin Liao [1], Michael J. Gandal, Richard S. Ehrlichman, Steven J. Siegel, Greg C. Carlson *

Department of Psychiatry, University of Pennsylvania, Philadelphia, PA 19104, USA

A R T I C L E I N F O

Article history:
Received 24 August 2011
Revised 16 December 2011
Accepted 31 December 2011
Available online 9 January 2012

Keywords:
Sensory evoked potential
SEP
Auditory evoked potential
AEP
ERP
MeCP2
Preclinical model
Mouse
Gamma oscillation

A B S T R A C T

Impairments in cortical sensory processing have been demonstrated in Rett syndrome (RTT) and Autism Spectrum Disorders (ASD) and are thought to contribute to high-order phenotypic deficits. However, underlying pathophysiological mechanisms for these abnormalities are unknown. This study investigated auditory sensory processing in a mouse model of RTT with a heterozygous loss of MeCP2 function. Cortical abnormalities in a number of neuropsychiatric disorders, including ASD are reflected in auditory evoked potentials and fields measured by EEG and MEG. One of these abnormalities, increased latency of cortically sourced components, is associated with language and developmental delay in autism. Additionally, gamma-band abnormalities have recently been identified as an endophenotype of idiopathic autism. Both of these cortical abnormalities are potential clinical endpoints for assessing treatment. While ascribing similar mechanisms of idiopathic ASD to Rett syndrome (RTT) has been controversial, we sought to determine if mouse models of RTT replicate these intermediate phenotypes. Mice heterozygous for the null mutations of the gene MeCP2, were implanted for EEG. In response to auditory stimulation, these mice recapitulated specific latency differences as well as select gamma and beta band abnormalities associated with ASD. MeCP2 disruption is the predominant cause of RTT, and reductions in MeCP2 expression predominate in ASD. This work further suggests a common cortical pathophysiology for RTT and ASD, and indicates that the MeCP2+/− model may be useful for preclinical development targeting specific cortical processing abnormalities in RTT with potential relevance to ASD.

© 2012 Elsevier Inc. All rights reserved.

Introduction

Event-related magnetoencephalography (MEG) and electroencephalography (EEG) studies of auditory and language processing have identified intermediate phenotypes associated with autism spectrum disorders (ASD) and abnormal responses in Rett syndrome (RTT) (Bader et al., 1989; Badr et al., 1987; Kalmanchey, 1990; Oram Cardy et al., 2008; Roberts et al., 2008; Stach et al., 1994). In autism, delayed middle latency components of the auditory-evoked response have been observed in the cortex (superior temporal gyrus) and have been linked to higher-order language impairments (Roberts et al., 2011). Likewise, abnormalities in cortical gamma-band (30–80 Hz) synchrony which have been observed in ASD, are thought to reflect deficits in excitatory–inhibitory balance (Gandal et al., 2010; Rojas et al., 2008; Wilson et al., 2007). Fewer and a less conclusive set or studies have been performed in girls with RTT (Kalmanchey, 1990) (Stach et al., 1994; Yamanouchi et al., 1993). The relative lack of preclinical studies investigating these auditory response deficits limits our ability to test for relationships between intermediate clinical phenotypes and neuronal circuit abnormalities in RTT.

Rett is a unique disorder, but shares proposed mechanistic and core symptoms of autism. In contrast to the complex genetic etiology of idiopathic ASD, RTT has a clear monogenetic basis with mutations in the X-linked gene MeCP2 occurring in approximately 90% of patients with RTT. Nevertheless, RTT is characterized by developmental regression, similar to that observed in a subset of severely affected autistic children, combined with the loss of age-appropriate social interaction and speech. Clinically, RTT patients often present with core autism-like behavioral deficits, along with RTT specific components that include severe motor abnormalities. In some cases, patients with RTT associated mutations in MeCP2 nevertheless present preserved but affected speech and limited motor abnormalities leading to a diagnosis of ASD that is clinically undifferentiated from idiopathic ASD (Young et al., 2007). Reduced MeCP2 expression is also found in forebrain post-mortem tissue from the majority of idiopathic ASD subjects, suggesting similar epigenetic dysregulation in many cases of idiopathic ASD and RTT (Samaco et al., 2004, 2005). Such links between MeCP2 and ASD suggest that mice with reduced MeCP2 expression may have construct validity for ASD as well as RTT, and

* Corresponding author at: Center for Neurobiology and Behavior, Upenn, Room 2226, TRL, 125 S. 31st Street, Philadelphia, PA 19104-3403, USA.
E-mail address: gregc@mail.med.upenn.edu (G.C. Carlson).
[1] Present Address: Institute of Neuroscience, National Cheng-Chi University, 64, Sec. 2, ZhiNan Rd., Wenshan District, Taipei City 11605, Taiwan.
Available online on ScienceDirect (www.sciencedirect.com).

0969-9961/$ – see front matter © 2012 Elsevier Inc. All rights reserved.
doi:10.1016/j.nbd.2011.12.048

thus models of MeCP2 dysfunction may help understand the distinction between RTT and ASD mechanisms and symptoms. Owing to the lack of well-characterized models of idiopathic ASD yet strong clinical data, changes in auditory and visual evoked phenotypes in a mouse model of RTT may provide additional insight into auditory and visual sensory processing abnormalities found in ASD.

To investigate the role of MeCP2 function on the integrity of sensory processing, this study measured auditory and visual event-related potentials (ERPs) in female mice carrying a single null allele of MeCP2, which replicates the genetic condition leading to RTT (Guy et al., 2001). A number of ERP features common to idiopathic ASD and RTT were observed, including delayed auditory-evoked responses, increased component amplitudes, and gamma-band abnormalities. Each of these differences has been identified in the ASD or RTT clinical population (Castren et al., 2003). These findings suggest that ASD and RTT may share a subset of underlying local circuit abnormalities that contribute to endophenotypic and behavioral abnormalities. Finally, by demonstrating intermediate phenotypic deficits in MeCP2+/− mice, this work helps bridge the divide between clinical and preclinical studies, providing a basis for future pathophysiological investigation and indicates targets for therapeutic development.

Materials and methods

Event-related potentials

Animals and implantation surgery

Female heterozygous Mecp2 null mice (*Mecp2*$^{tm1.1Bird/J}$) and littermate controls (n = 9/group) were obtained from The Jackson Laboratory (Bar Harbor ME) and bred in house using males from the background strain (C57/B6J). At 4 months of age, mice underwent stereotaxic implantation of bipolar, twisted, stainless steel electrodes into region CA1 of the hippocampus (AP −2.2 mm, ML 2.0 mm, DV −1.9 mm; 100 μm diameter, Plastics One, Roanoke VA). A reference skull screw was implanted over the primary visual cortex (AP −3.4 mm, ML −2.7 mm) and a ground screw was placed above the frontal cortex (AP 3.1 mm, ML −1.0 mm).

Recording of event-related potentials (ERPs)

Recording of auditory and visual ERPs was performed between 10 AM and 4 PM, after a minimum of two weeks recovery from surgery. All studies were performed in mice between the ages of 4 and 5 months, a period where these mice are largely presymptomatic (Stearns et al., 2007). The mice were tested in their home cages, which are fitted with special tops to accommodate speakers and electrode cables, and placed inside a Faraday cage. The mice were then acclimatized to the testing apparatus for 30 min before first the stimulus onset. The head stage is connected to a 30 cm six channel electrode cables, which is in turn connected to a high-impedance differential AC amplifier (A-M Systems, Carlsborg WA). Auditory stimuli were generated by Micro1401 hardware and Spike 6 software (Cambridge Electronic Design) and delivered through speakers attached to the cage top. In the presence of background white noise of 55 dB, 150 single white-noise clicks (10 msec in duration) were issued at 82 dB 8 s apart. For gating experiments, 150 white-noise clicks pairs (S1, S2) were presented with a 500 msec inter-stimulus interval and a 9 s inter-trial interval. Auditory brain stem responses were derived from stimuli consisting of a 4000 white noise clicks (0.1 ms duration, 125 ms ISI) sampled at 10 kHz, repeated at 85 dB similar to as previously published (Connolly et al., 2003). Recordings were filtered 100–500 Hz offline (Digital IIR filter, Butterworth Bandpass, order 4) and grand averages were compared. Recording of visual evoked potentials was performed as previously published (Halene et al., 2009). Visual stimuli were delivered through a flash box (PS40/R Photic Stimulator, Grass Technologies, West Warwick, RI) 30 cm above home cages with transparent cage tops. During the 15 min acclimation period and subsequent stimulus presentation, mice were entirely in the dark. Average waves were created for the response to the visual stimulus for each mouse separately. Recording sessions consisted of an acclimation phase (15 min) and subsequent data collection. Analysis was performed as described above for auditory ERPs.

EEG signal was bandpass filtered online between 1 and 500 Hz, and grand average waveforms were created from −500 ms to 1000 ms relative to the auditory stimulus. To remove movement artifacts, trials containing activity over 2 SD of the mean were rejected. Initial peak analysis was performed in Microsoft Excel (Redmond, WA) or Igor (Wavemetrics, OR) on the remaining averaged trials. The baseline was corrected at stimulus onset of S1 and S2 independently. Peak components were extracted from grand-average waveforms as follows: P1/P20 (most positive deflection between 10 and 30 msec), N1/N40 (most negative deflection between 25 and 60 msec) and P2/P80 (most positive deflection between 60 and 250 msec) in the averaged waves were analyzed by two-way analysis of variance (ANOVA). To also test for differences *independent of* peak identification each time point was compared to using a *T*-test and corrected for multiple comparisons using a non-parametric bootstrap method fully described bellow for time frequency analysis and additionally described by Carlson et al. (2011).

Time-frequency analysis

Spectral decomposition of auditory-evoked response waveforms was performed using the EEGLab toolbox in Matlab (Delorme and Makeig, 2004), as published (Gandal et al., 2010). Single-trial epochs between −0.3 and 0.8 s relative to the first stimulus (S1) were extracted from the continuous EEG data sampled at 1667 Hz. For each epoch, total power (i.e., event-related spectral perturbation, ERSP) and phase-locking values (i.e., intertrial coherence, ITC) were calculated using Morlet wavelets in 100 linearly spaced frequency bins between 5.0 and 100 Hz, with wavelet cycles increasing from 3 (at low frequencies) to 6 (at high frequencies). Total power was calculated in decibels (dB) relative to baseline power (−200 to 0 ms) in each frequency band. Phase locking factor (PLF) is expressed as a unitless ratio between 0 and 1, where 1 represents complete phase synchrony at a given frequency and time across trials. Significance for total power and PLF differences between the group-derived time-frequency graphs was assessed by unpaired T-tests at each time-frequency bin (Carlson et al., 2011). Correction for multiple comparisons was achieved using a non-parametric permutation bootstrap method as implemented in EEGlab, using 500 permuted samples. The permutation bootstrap method involves pooling all of the single-subject 2D time-frequency images from both groups, randomly shuffling and partitioning the pooled samples into two “random” groups of the same size as the initial comparison. Unpaired t-tests are then conducted between these two randomly chosen groups at each time-frequency bin and the maximum T-statistic is retained. Repeating this procedure 500 hundred times generates a distribution of T-statistics. One can then compare T-statistics from the original group comparisons to this permuted bootstrap distribution, retaining any points that have *P* values less than 0.05. Such correction for multiple comparisons has been commonly used in the neuroimaging community (Singh et al., 2003).

Results

Auditory and visual evoked responses produce a typical set of middle latency positive going (P) and negative going (N) components that are labeled P1–N1–P2 in humans as well as in mice. In response to single stimuli spaced 8 s apart, the ERP responses in both MeCP2+/− and wildtype littermate mice showed this same typical pattern (Fig. 1). Following the P2 there can be another wide positivity (P3) associated with novelty and often an even broader negativity sometimes described as the slow wave. These middle and late-latency events in mice are

Download English Version:

https://daneshyari.com/en/article/3069488

Download Persian Version:

https://daneshyari.com/article/3069488

Daneshyari.com