Experimental Brain Research
https://doi.org/10.1007/s00221-017-5164-4

RESEARCH ARTICLE

Children with autism spectrum disorder have unstable neural responses to sound

Sebastian Otto-Meyer[1,2] · Jennifer Krizman[1,2] · Travis White-Schwoch[1,2] · Nina Kraus[1,2,3,4,5]

Received: 1 August 2017 / Accepted: 26 December 2017

Abstract

Autism spectrum disorder (ASD) is diverse, manifesting in a wide array of phenotypes. However, a consistent theme is reduced communicative and social abilities. Auditory processing deficits have been shown in individuals with ASD—these deficits may play a role in the communication difficulties ASD individuals experience. Specifically, children with ASD have delayed neural timing and poorer tracking of a changing pitch relative to their typically developing peers. Given that accurate processing of sound requires highly coordinated and consistent neural activity, we hypothesized that these auditory processing deficits stem from a failure to respond to sound in a consistent manner. Therefore, we predicted that individuals with ASD have reduced neural stability in response to sound. We recorded the frequency-following response (FFR), an evoked response that mirrors the acoustic features of its stimulus, of high-functioning children with ASD age 7–13 years. Evident across multiple speech stimuli, children with ASD have less stable FFRs to speech sounds relative to their typically developing peers. This reduced auditory stability could contribute to the language and communication profiles observed in individuals with ASD.

Keywords Autism spectrum disorder · Neural stability · Neural variability · FFR · Auditory · Sound processing

Introduction

Autism spectrum disorder (ASD) is a developmental disability that affects ~ 1% of the population (Won et al. 2013) and is characterized by impaired social-communication function and repetitive, restricted behavior (American Psychiatric Association 2013). ASD is a heterogeneous, complex disorder, with high levels of genetic (Geschwind 2011) and behavioral (Jeste and Geschwind 2014) variation among affected individuals. Despite this variation, anatomical and physiological abnormalities, such as neural excitation–inhibition imbalances (Markram et al. 2007; Peñagarikano et al. 2011), immune and mitochondrial dysfunction (Rossignol and Frye 2014), and over-pruning of neurological connections (Thomas et al. 2016), are consistently found among ASD individuals.

Noisy or variable neural processing is another physiological abnormality associated with ASD. In children and adults with ASD, unstable responses have been found with fMRI (Dinstein et al. 2010, 2012; Haigh et al. 2015), EEG (Milne 2011), and MEG (Takahashi et al. 2016) in response to somatosensory (Haigh et al. 2015), motor (Dinstein et al. 2010), and visual (Dinstein et al. 2010; Milne 2011; Takahashi et al. 2016) stimuli. Within an individual, this unreliability appears to pervade across modalities. Indeed, Dinstein et al. (2012) found increased variability in response to auditory, somatosensory, and visual stimuli in the same subjects. Collectively, these studies show an unstable, highly variable response to sensory stimuli in the ASD brain. Reduced neural stability could impede formation of a consistent representation of the sensory world, as suggested by the "neural noise" theory of ASD (Baron-Cohen

✉ Nina Kraus
nkraus@northwestern.edu
http://www.brainvolts.northwestern.edu

[1] Auditory Neuroscience Laboratory, Northwestern University, 2240 Campus Drive, Evanston, IL 60208, USA

[2] Department of Communication Sciences, Northwestern University, Evanston, IL 60208, USA

[3] Institute for Neuroscience, Northwestern University, Evanston, IL 60208, USA

[4] Department of Neurobiology and Physiology, Northwestern University, Evanston, IL 60208, USA

[5] Department of Otolaryngology, Northwestern University, Evanston, IL 60208, USA

Published online: 06 January 2018

and Belmonte 2005), which would have consequences for learning and development.

Given the prevalence of communication deficits in ASD and the vital role the auditory system plays in language learning and communication (Doupe and Kuhl 1999; Bailey and Snowling 2002), the auditory system serves as a prime location to target investigations into abnormal neural activity. Rosenhall and colleagues (2003) used click-evoked auditory brainstem responses (ABRs), a common electrophysiological method for examining basic neural function (Hall 2006), to examine auditory neural processing in children with ASD. Children with ASD had slower click-evoked ABRs relative to typically developing children (Rosenhall et al. 2003), indicating a potential problem with auditory processing. Studies using the frequency-following response, or FFR, have further shed light on auditory processing in individuals with ASD. The FFR is the brain's response to a periodic sound, including complex sounds such as speech, and gives a nuanced picture of auditory processing (Chandrasekaran and Kraus 2010; Skoe and Kraus 2010). The FFR is likely generated by the inferior colliculus (IC), an integration center in the midbrain that receives and sends inputs to and from the brainstem, but includes components from throughout the auditory system, including the cortex (Coffey et al. 2016). It has also been shown to be relatively insensitive to subject state (Skoe and Kraus 2010). Russo and colleagues found that individuals with ASD have slower neural responses to speech (Russo et al. 2009) and poorer tracking of pitch contours (Russo et al. 2008), providing objective evidence of difficulty in processing prosodic cues, which has been observed, albeit with somewhat conflicting evidence, in individuals with ASD (McCann and Peppé 2003). These results hint at deficits in the ASD brain's ability to process complex sounds, such as speech.

We hypothesized that variability within the auditory system disrupts formation of a stable representation of the auditory world, a critical component of communication and language, and manifests as deficits in auditory processing. To test the hypothesis that these neural deficits are the consequence of a common underlying bottleneck in sound processing, such as noisy or variable processing, we compared across-trial stability of click ABRs and speech-evoked FFRs of high-functioning ASD children and typically developing children. This method allowed us to specifically target the auditory system, and examine deficits in stability on a measure known to have functional consequences on language ability (Banai et al. 2009; Hornickel and Kraus 2013). Unlike the fMRI used by Dinstein et al. (2012), the FFR captures precise processing of an auditory stimulus with greater time resolution, and, therefore, provides an easily-interpretable measure with clinical ramifications that operates closer to the actual time scale of auditory signals. We predicted that children with ASD would have a less stable response than typically developing children, resulting in an FFR that is less consistent, and that this difference would not be driven by increased nonstimulus activity.

Methods

The Institutional Review Board of Northwestern University approved the methods used in this study. Informed consent and assent were obtained from the parent(s) or legal guardian(s) and the child.

The data shown here have been used in the previous analyses (Russo et al. 2008, 2009) and are analyzed retrospectively in this current report. Electrophysiological responses to a broadband click, a /d/, and a /ya/ syllable with a changing fundamental frequency were examined (stimuli described below). For all stimuli, responses were compared between children with ASD and typically developing children. Previous findings from Russo et al. 2008, 2009 showed ASD children have later latencies at peaks V, A, D, and F for the response to the /d/, and less accurate pitch tracking in their responses to the /ya/ stimuli.

Subject information

Twenty-four school-aged children were included in these analyses (mean age = 10.71 years, SD = 2.07, range of 7–13 years, 4 female). Twelve (one female) had an ASD diagnosis, while twelve (three female) were typically developing. The typically developing group was matched with the ASD group on age and verbal/nonverbal IQ using the Wechsler Abbreviated Scale of Intelligence (WASI, Pearson, San Antonio, TX). All children participated in every aspect of this study.

Inclusionary criteria were a full scale IQ ≥ 80, air conduction thresholds ≤ 20 dB nHL for octaves between 250 and 8000 Hz, and click-evoked wave V latencies consistent with laboratory norms (5.32–5.90 milliseconds). Otoscopies were performed on children prior to testing to ensure an unoccluded ear canal.

Participants with ASD were recruited from community and online groups for families of children with ASD. A diagnosis of ASD must have been made by a child neurologist or psychologist, with parents supplying the name, credentials, and office location of the medical professional, along with the date of initial evaluation and the specific diagnosis made. The participants also had to be routinely examined by their physicians and school professionals. These diagnoses were made under the DSM-IV criteria, and so include diagnoses of Asperger's and pervasive developmental disorder not otherwise specified [these diagnoses now fall under general autism spectrum disorder under the diagnostic criteria of the DSM-V (American Psychiatric Association 2013)].

In addition to formal diagnoses, most of the children with ASD were described by parents as struggling with prosody perception. Furthermore, these participants were noted by testers to have some or all of the following behaviors: speech and language characteristic of ASD (including abnormal pitch, volume, or intonation, repetitive or idiosyncratic language, echolalia, and narrow conversation interests) and social behavior characteristic of ASD (including limited eye contact, reduced social and emotional reciprocity, and distinctive body and hand movements). These behaviors were noted during social interaction with testers prior to and following the neural testing.

Typically developing participants were recruited from the community and local schools. No participants had a confounding neurological diagnosis such as active seizure disorder or cerebral palsy.

Electrophysiology

All electrophysiological recordings were completed in a sound-attenuated chamber while the child sat comfortably in a reclining chair and watched a movie at ≤ 40 dB SPL of his/her choice. The child's parent sat in the room to increase compliance and notify the tester if any problems arose during recording.

Recordings were performed using 3 Ag–AgCl scalp electrodes; active was located at Cz, reference on the right earlobe, and ground on the center of the forehead. All electrodes had an impedance of ≤ 5 kOhms throughout testing. Brain responses to the /d/ and to the click were recorded and stimuli were presented using the Navigator Pro (Bio-logic Systems Corp., a Natus Company, Mundelein, IL) with BioMARK software, while responses to the /ya/ were recorded using NeuroScan Acquire4 (Compumedics Neuroscan, Charlotte, NC) and stimuli were presented using Neuroscan Stim. These responses, retrospectively analyzed for this current report, were collected as a part of two separate studies that used these two distinct recording setups. All stimuli were played monaurally to the right ear via ear insert earphones (ER-3, Etymotic Research, Elk Grove Village, IL), while the child heard the movie soundtrack at ≤ 40 dB SPL in sound field with their left (non-test) ear.

Stimulus 1: click

A 100 µs duration click stimulus was presented 31/s at 80.3 dB SPL for ABR recordings. Responses were digitally sampled at 24 kHz in a 10.66 ms recording window and online bandpass filtered from 100 to 1500 Hz. Artifact reject limits were set at ± 23.8 µV to eliminate electromyogenic contamination resulting from jaw or neck movements. Two averages consisting of 2000 sweeps (one sweep being the neural response to a single presentation of the stimulus) each were collected at the beginning of a recording session to confirm replicability. At the end of each session, another 2000 sweeps were collected, allowing the tester to confirm using latencies that ear insert depth had not changed over the course of the recording.

Stimulus 2: /d/

The /d/ stimulus was a speech syllable 40 ms in length, representing an onset burst followed by a consonant-to-vowel transition. The /d/ was synthesized in Klatt (Klatt 1980), with voicing beginning at 5 ms. Frequency components are as follows, with the fundamental frequency and the first three formants changing linearly across the stimulus: F0: 103–125 Hz; F1: 220–720 Hz; F2: 1700–1240 Hz; F3: 2580–2500 Hz; F4: 3600 (constant); F5: 4500 (constant). The stimulus was presented at 80 dB SPL, in alternating polarities, with responses to the two polarities added to minimize stimulus artifact (Campbell et al. 2012). Sweeps were collected over a 75 ms total window, including 15 ms of prestimulus and 20 ms after the stimulus offset, at a rate of 10.9/s. Responses were sampled at 6856 Hz and filtered online from 100 to 2000 Hz with a 12 dB/octave rolloff. Responses over ± 23.8 µV were rejected online. A total of 4000 sweeps were analyzed, and collected in two blocks of 2000 sweeps, creating two subaverages of 2000 sweeps each. The two subaverages were averaged together to get a final average. The prestimulus region of this final average was used to calculate nonstimulus activity.

The /d/ can be seen in the time domain in Fig. 1c, along with its characteristic evoked neural response (1d) (Skoe et al. 2015). The neural response to the /d/ stimulus possesses six characteristic peaks (V, A, D, E, F, and O) corresponding to major acoustic features of the stimulus. The peaks occur approximately 6–8 milliseconds after their corresponding acoustic feature, with this time lag representing the neural transmission delay between the cochlea and rostral brainstem. Peaks V and A represent an onset response, and Peak O is in response to the offset, representing the end of the sound. Peaks D, E, and F are the periodic portion of the response, corresponding to the fundamental frequency of the stimulus and its harmonics. Although wave V of the FFR is similar to wave V of the click-evoked ABR (King et al. 2002), the FFR wave V has been shown to be a more sensitive measure of auditory function (Song et al. 2008; Banai et al. 2009; Krizman et al. 2010, 2012b).

Stimuli 3 and 4: /ya/ rising and /ya/ falling

The /ya/ stimuli were naturally spoken by a native English-speaking female, fully voiced with a flat pitch. The syllable was then manipulated in Praat (Boersma 2006), with the duration normalized to 230 ms and the F0 contour altered

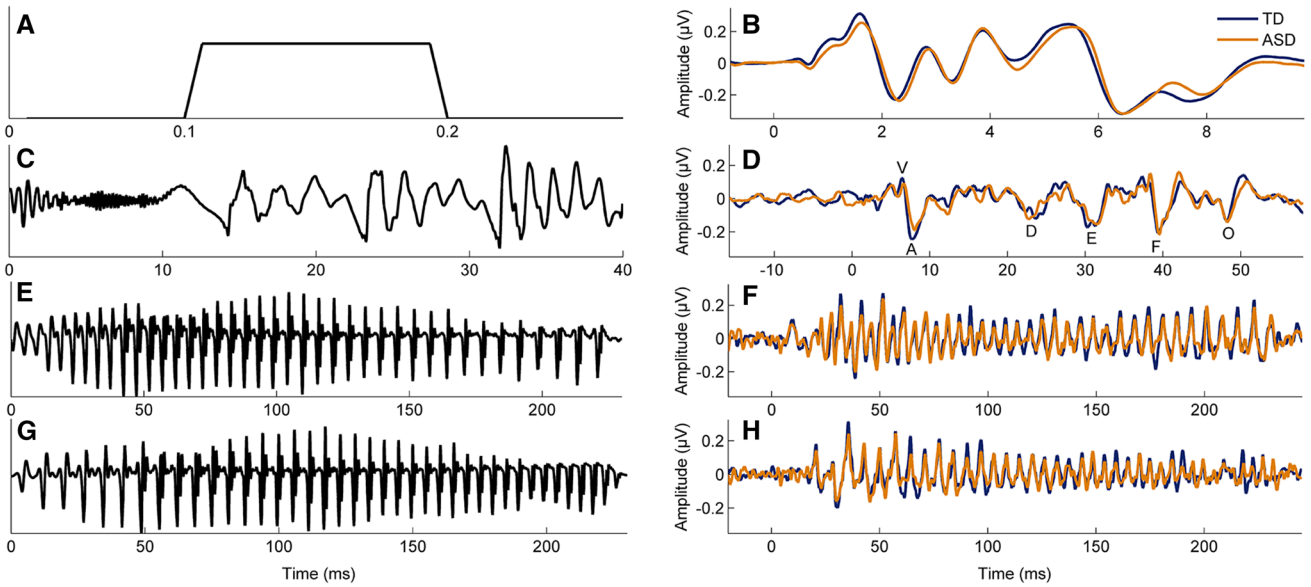

Fig. 1 FFR closely mirrors the stimulus that is played and key features are reliably expressed in a healthy system. Similarities can be seen between the FFR stimuli (left column) and grand average responses (right column). **A, B** Show, respectively, the click stimulus and the response (note that there is no similarity between the click stimulus and response—it is simply included for consistency). **C, D** Show, respectively, the /d/ stimulus and the response. This response has been well-described, and its six characteristic peaks (V, A, D, E, F, and O) are labeled. **E, F** Show the stimulus and response to the falling /ya/, while **G** and **H** show the rising /ya/. These responses also morphologically mirror the stimuli, especially in capturing the large, changing peaks of the fundamental frequency. In all response plots, the grand averages of TD versus ASD children are represented by blue and orange, respectively. No obvious gross morphological differences are present between the two groups, indicating that the FFR is a viable measure in both groups

to create rising (120–220 Hz) and falling (220–120 Hz) reciprocal F0 contours (Fig. 1). The rising and falling stimuli were otherwise identical. Stimuli were presented in alternating polarities and responses to the two polarities were added post-collection to minimize stimulus artifact (Campbell et al. 2012). During recording, the syllable was presented at 60 dB SPL, with rising and falling presented in a random order and a variable interstimulus interval (51 ± 16 ms) to prevent an anticipatory response. The response was sampled at 20 k Hz and filtered offline from 80 to 1000 Hz with a 12 dB/octave rolloff. Two blocks of 1200 sweeps per polarity were collected for each stimulus (2400 per stimulus) for a total of 4800 sweeps per block. The two blocks were then merged together, giving a final count of 4800 sweeps per stimulus (rising versus falling) at alternating polarities. The prestimulus region of this final average was used to calculate nonstimulus activity. Sweeps larger than ± 35 μV were rejected offline, consistent with previous work (Russo et al. 2008).

The characteristic neural responses to falling (Fig. 1E, F) and rising (Fig. 1G, H) /ya/ stimuli also capture acoustic aspects of the stimulus, including frequency and timing information. Because the FFR to the /ya/ occurs over the entire response, we used the entire response, with a five millisecond shift to account for neural delay, in our analyses.

Analyses

Response stability

To analyze response stability, the two subaverages of the response to the /d/ were correlated using a Pearson correlation, with an *r* value of 1 indicating a perfect correlation and an *r* value of 0 indicating no correlation. Higher *r* values, therefore, represent a more stable response across trials, while lower *r* values represent a more variable response. Click response stability was analyzed in the same manner, although all three trials were correlated separately, and an average of the three was used for analyses. Although graphs and figures report *r* values, these values were Fisher transformed prior to statistical analyses to place the values on a normal distribution. For the measures of response stability reported for the /d/, the region of the response analyzed was restricted to 19.5–44.2 ms. For the click, the 0–9.82 ms region was analyzed.

For the /ya/ responses, neural stability was determined using a similar method to previously published work (Tierney and Kraus 2013). Two thousand of the 4800 sweeps (post-artifact rejection) were randomly selected and averaged, creating a subaverage. This subaverage was then correlated with a second subaverage of 2000 different sweeps randomly selected from among the remaining 2800 trials. This was done 300 times,

and the average z correlation value was determined and used for statistical analyses. We have previously shown that different methods of calculating stability, such as comparing earlier to later sweeps versus randomly-chosen sweeps, have little to no impact on the ultimate value (Hornickel and Kraus 2013), showing that this measure is capturing true neural stability rather than a fatigue effect or change in subject state. We chose to use two separate means of assessing response stability to show that any differences we found were not due to methodology of assessing stability, but rather a consistent difference. The *z* values were transformed into *r* values for the purpose of reporting in figures.

Nonstimulus activity

For the /d/, the root-mean-square amplitude of the 15 ms prestimulus interval was measured. This value is reported as the nonstimulus activity, as it represents the level of neural activity when no stimulus is playing. For the /ya/, the root-mean-square amplitude of the 15 ms prestimulus interval was measured. The neural stability of these time periods was also assessed, using the same methods described in the "Response Stability" section above.

Statistics (all stimuli)

We predicted that neural instability would be a hallmark of ASD and would thus manifest across all stimuli. Given that the click ABR and the FFR are independent of changes to subject state, we also predicted that this instability would be independent of differences in nonstimulus activity. Therefore, we ran a repeated measures analysis of variance (RMANOVA) to compare the stability of responses of children with ASD to typically developing children across stimuli, using each stimulus as a repeated measure. Greenhouse–Geisser corrections were applied to degrees of freedom where appropriate due to a lack of sphericity. Nonstimulus activity was analyzed with a RMANOVA as well; however, the nonstimulus activity for the responses to the two /ya/ stimuli was combined, as the /ya/ responses were collected in an interleaved fashion in the same session. Planned post-hoc *t*-tests were used for follow-up comparisons of response stability for each individual stimulus. *P* values below 0.05 were considered significant and all tests were two-tailed.

Results

Responses

Grand average responses are shown in Fig. 1, displaying the brain response to the sounds played. As shown in Fig. 1, there are no gross differences in morphology in the responses to the click, /d/, or either /ya/ between the ASD and typically developing (TD) groups.

Response stability

Response stability had a main effect of disorder [$F(1,22) = 10.444$, $p = .004$] but no disorder by stimulus interaction (Fig. 2a). There was also a main effect of stimuli [$F(1.951, 42.926) = 113.794$, $p < .001$], which was expected given the differences in acoustic features across stimuli. Planned follow-up comparisons found ASD children had lower response stability than TD children to all stimuli: click [$t(22) = 2.563$, $p = 0.018$, Cohen's $d = 1.05$], /d/ [$t(22) = 2.083$, $p = 0.049$, Cohen's $d = 0.85$], /ya/ falling responses [$t(22) = 2.433$, $p = 0.024$, Cohen's $d = 0.99$], /ya/ rising responses ($t(22) = 2.275$, $p = 0.033$, Cohen's d = 0.93). Individual data are shown in Fig. 2b–e.

In Fig. 3, sample waveforms from selected participants illustrate the difference between low and high response stability. Figure 3a presents overlaid subaverages for the response to /d/ for a TD child with high response stability, while Fig. 3b shows the same for an ASD child with low response stability. Similarly, Fig. 3c, d shows subaverages for the falling /ya/ for a TD child and ASD child, respectively.

Nonstimulus activity does not differ between groups

Nonstimulus activity was compared between ASD and TD children using a RMANOVA (Fig. 4). Activity for rising and falling was averaged for the two /ya/ stimuli, as these two stimuli were presented in a randomly interleaved format in the same session. There was no main effect of disorder [$F(1,22) = 1.914$, $p = 0.180$]. There was a main effect of stimulus [$F(1,22) = 35.889$, $p < 0.001$], with the averaged /ya/ nonstimulus activity higher than the /d/; however, this was likely due to differences in filtering, recording systems, and particularly the larger artifact reject criteria for the /ya/.

Stability of the nonstimulus activity also did not differ between groups when tested using a RMANOVA, with no main effect of disorder [$F(1,22) = 0.015$, $p = 0.904$]. There was also no main effect of stimulus [$F(1.978,43.517) = 0.362$, $p = 0.698$]. The average stability (*z* value) for /d/ (ASD average = −0.035, TD average = 0.080) or the /ya/ (ASD average = 0.000, TD average = −0.042) hovered around zero.

Discussion

Children with ASD have less stable neural responses to sound than typically developing children, a difference that was observed across responses to four different stimuli:

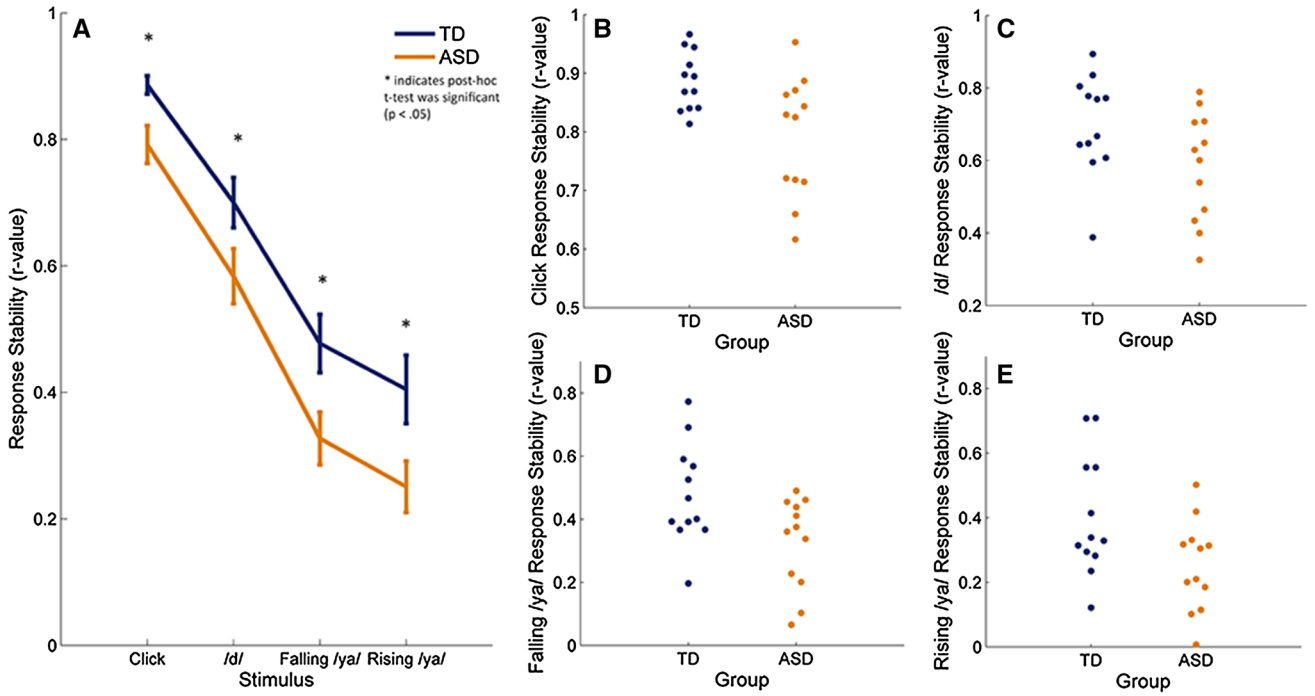

Fig. 2 **A** Shows the change in average response stability in *r* value for TD (blue) and ASD (orange) children from the click to the /d/ stimuli. ASD children had less stable responses than TD children across all stimuli [$F(1,22) = 10.444$, $p = 0.004$, results of individual *t*-tests are noted in **A** above each stimulus comparison, with $^{*}p < 0.05$, $^{**}p < 0.01$]. Different stimuli also lead to responses of different stability [$F(1.951,42.926) = 113.794$, $p < 0.001$]. Click stimuli led to relatively consistent responses, while increasingly complicated speech cues led to less consistent responses. **B**–**E** Show individual data plotted for TD and ASD children in response to the click, /d/, falling /ya/, and rising /ya/, respectively, emphasizing the shifted distribution that ASD children have relative to TD children. Mean and standard error for the groups (in *r* values) are as follows: TD click: 0.886 (0.014), ASD click: 0.792 (0.030), TD /d/: 0.700 (0.040), ASD /d/: 0.584 (0.043), TD /ya/ falling: 0.477 (0.046), ASD /ya/ falling: 0.327 (0.042), TD /ya/ rising: 0.405 (0.054), TD /ya/ falling: 0.251 (0.041)

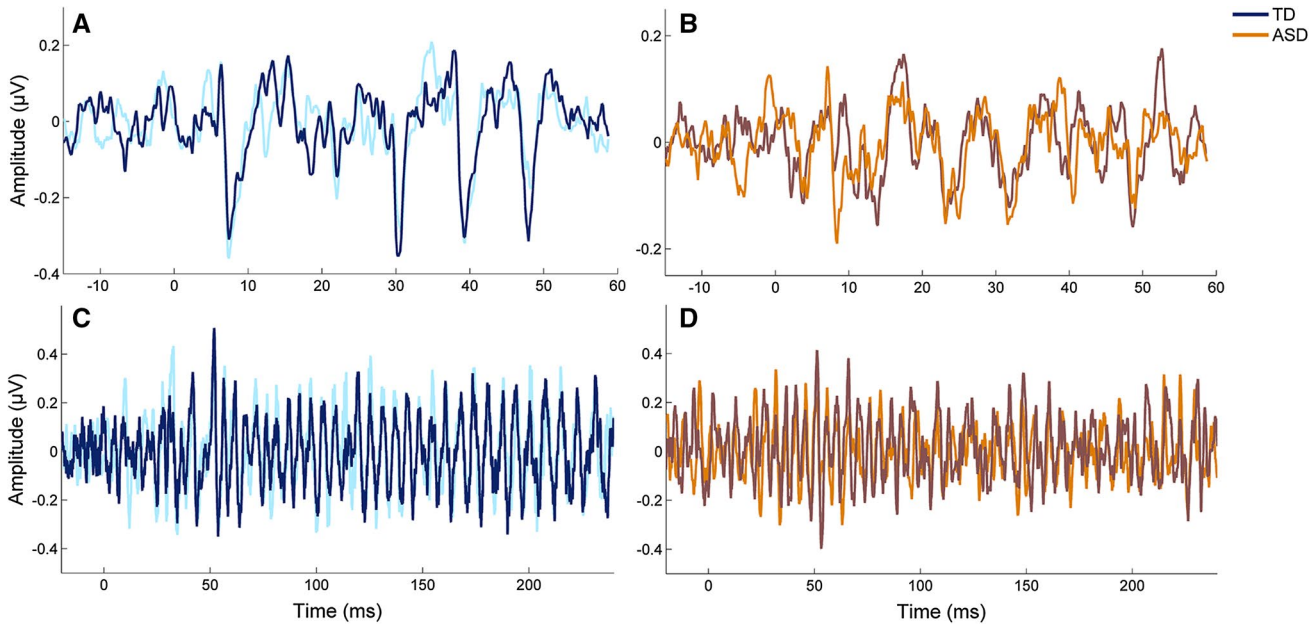

Fig. 3 Representative examples of TD and ASD responses. In each panel, an average of half the total number of sweeps collected is overlaid over an average of the remaining half. **A, B** Show a TD and ASD response to the /d/, respectively. **C, D** Show a TD and ASD response to the falling /ya/, respectively

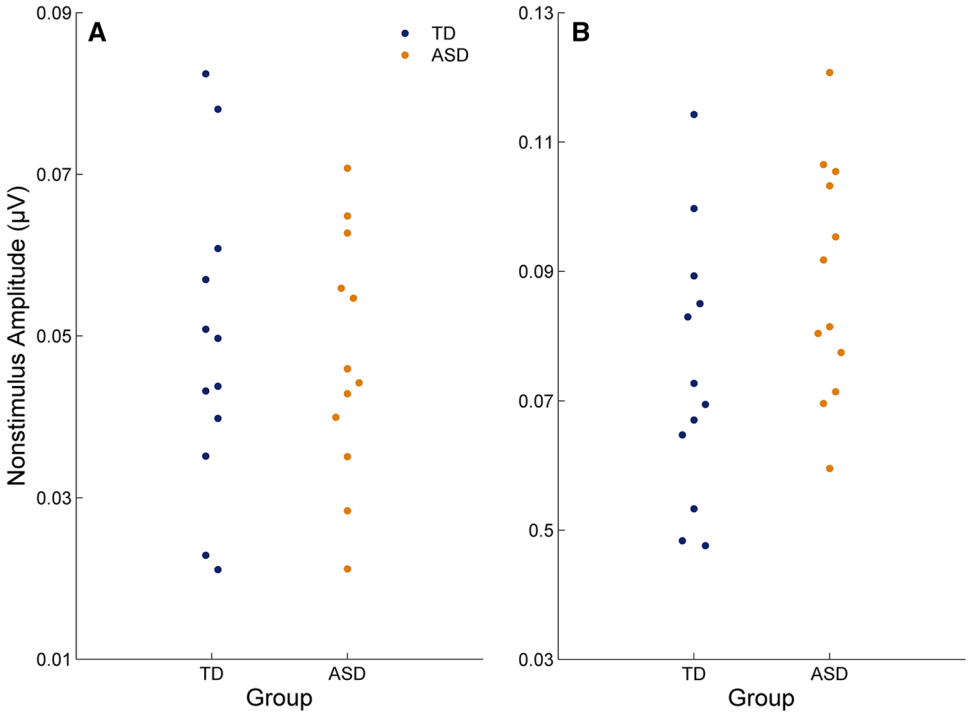

Fig. 4 Nonstimulus activity was not significantly different between the TD and ASD children ($F(1,22) = 1.914$, $p = 0.180$). 4A and B show individual data plotted for TD (blue) and ASD (orange) children for the /d/ and averaged /ya/ activities, respectively. Mean and standard error for the groups (in μV) are as follows: TD /d/: 0.049 (0.005), ASD /d/: 0.047 (0.004), TD /ya/: 0.073 (0.007), ASD /ya/: 0.091 (0.006)

click, /d/, a falling /ya/, and a rising /ya/. It is noteworthy that these stimuli varied in sound level, length, frequency, interstimulus interval, and source (synthesized versus naturally spoken). Group differences in response stability were present, even though the ASD subjects were high-functioning and the two groups were well-matched on nonverbal and verbal IQ, sex distribution, and age. Thus, lower response stability is a consistent group distinction between children with ASD and their typically developing peers. Moreover, children with ASD and typically developing children had similar levels of nonstimulus activity and no difference in neural stability of the nonstimulus region, suggesting that the observed differences in neural stability were driven by differences in their sound-evoked responses rather than by differences in nonstimulus activity.

Despite consistent group differences, the reduced neural stability we observed showed the same heterogeneity often present in individuals with ASD. Just as with typically developing children, the stability of responses for ASD children fell on a spectrum. As all of the subjects were high-functioning, it is clear that a wide variety of neural stability is present even among ASD individuals of relatively similar symptom severity. These results suggest that some subjects have difficulties with complex auditory processing, while others may not.

Differences were also found between responses to different stimuli within groups. In general, simpler stimuli evoked more stable responses, as would be expected. However, contrary to expectations, nonstimulus activity was higher for the responses to the /ya/ stimuli than the responses to the /d/ stimuli. This was likely due to the differences in recording parameters already mentioned in the methods and results. Unfortunately, the responses used for these analyses were collected as parts of different studies, as this is a retrospective reanalysis. It is possible that these differences in parameters had an impact on the stability measures of the FFRs as well. However, these differences do not diminish, but, in fact, enhance the finding of reduced stability across these stimuli. Neural stability of children with ASD was uniformly lower than TD children despite varying stimuli, collection parameters, and recording devices, showing that this finding generalizes across multiple types of FFR and ABR.

Links to behavior and neurological theories of ASD

Previous work has found unstable responses in both children and adults with ASD (Coskun et al. 2009; Dinstein et al. 2010, 2012; Milne 2011; Takahashi et al. 2016). Our findings are generally consistent with these studies. Our study, however, extended this work in several ways. First, it was able to target the midbrain, a region generally difficult to assess with measures such as fMRI. This extends the previous findings of variable neural processing in ASD to the midbrain. Second, this study targets the auditory system, a sensory system that has currently been less assessed in variability research. We were also able to examine fast brain activity on the order of milliseconds using the FFR, a contrast to the previous work which has restricted its variability analyses to slow cortical oscillations, or in the case of fMRI work, the even

slower changes in the BOLD signal. In addition, many of the studies above used and found effects with relatively simple sensory stimuli. Although our stimuli included a simple click stimulus, we also included more complex speech stimuli, including those with prosodic cues such as rising or falling voice pitch. We can, therefore, see the potential functional ramifications of this reduced stability: children with ASD are not able to process speech stimuli in a stable manner, and, therefore, may not be able to pick up on prosodic or temporal cues within speech.

As a whole, our study appears to support the increased neural noise theory of ASD, or the idea that the symptoms of ASD are based in an inability to distinguish signal from noise throughout the brain (Baron-Cohen and Belmonte 2005). This theory is supported by research showing increased connectivity between sensory regions and corresponding subcortical regions in the brains of ASD individuals (Cerliani et al. 2015) and increased local hyperconnectivity (Supekar et al. 2013; Nomi and Uddin 2015), as well as research investigating an excitation/inhibition imbalance in ASD individuals (Nelson and Valakh 2015). In the auditory midbrain, all of these factors could converge to cause processing deficits. Molecular imbalances could lead to misfiring and a lack of inhibition in a system that needs precise timing to function properly, resulting in difficulties in representing a signal accurately. Local hyperconnectivity may have a similar effect—reducing the precision of the auditory system and, therefore, reducing its stability. Increased connectivity between cortical and subcortical regions of the brain could also be implicated in reduced stability in the FFR. The subcortical auditory system relies on fine-tuning from efferent top–down connections. If these connections are not precisely defined and/or are over-proliferated, this tuning might not be functioning properly, reducing the stability and precision of subcortical auditory networks. However, it is also possible that the reduced stability we see in this study arises from an entirely different source. A study has shown lower local connectivity in sensory regions of ASD individual's brains (Dajani and Uddin 2016), rather than the increased connectivity that reduced stability (or higher variability) would imply. Other reports have disputed the increased noise theory (Davis and Plaisted-Grant 2015) and have described the inconsistencies in the connectivity research (Vasa et al. 2016). Given that we did not find a difference in resting neural noise (as measured using the nonstimulus interval between stimulus presentation), this suggests that the variability is not due to an increase in general neural noise, but rather a specific inability to extract the signal of a sound in a stable manner. Therefore, while our study identifies instability of auditory midbrain processing as a feature of ASD, future studies with animal models can pinpoint the underlying mechanisms leading to this instability.

What does reduced stability in the FFR mean?

Interestingly, an unstable FFR has been found in other populations with poor auditory function as well. These include poor readers (Hornickel and Kraus 2013), the elderly (Anderson et al. 2012), and those of low socioeconomic status (Skoe et al. 2013). All of these populations also possess hearing or language deficits compared to typical populations. While the fact that this "signature" is not unique indicates caution should be taken in analysis of an unstable FFR, its concurrence with language deficits implies that it has behavioral ramifications. However, these ramifications may differ across affected populations. For example, unstable FFRs in populations of poor readers have been postulated to have both "top–down" and "bottom–up" effects, whereby poor stability leads to difficulty with phonological and language learning (bottom–up), which in turn causes cortical refinement to be unable to properly shape the auditory system to properly process sounds (top–down) (Hornickel and Kraus 2013).

While FFRs are thought to primarily reflect pooled activity from the inferior colliculus (White-Schwoch et al. 2016), recent theories have placed the FFR as part of a more integrated system (Kraus and White-Schwoch 2015; Coffey et al. 2016) in which the FFR has some cortical contribution, along with the midbrain components of the FFR receiving consistent tuning from efferent and afferent inputs. This gives rationale for the fact that the FFR, and correspondingly its major contributors in the auditory midbrain, can be shaped by life experience (Krishnan et al. 2004, 2005; Musacchia et al. 2007; Jeng et al. 2011). If the FFR is indeed plastic, negative aspects could potentially be targeted and improved. In fact, there is some evidence that the impaired aspects of the FFR in children with ASD can be positively affected by language and phonological training (Russo et al. 2010). A rat model of ASD was also shown to improve in auditory (cortical) responses with auditory training (Engineer et al. 2014), providing additional evidence of auditory plasticity being used to assist individuals with ASD. Furthermore, the FFR has been shown to be affected by musical training and second language learning (Krizman et al. 2012a, 2015; Skoe and Kraus 2013; Kraus et al. 2014; Weiss and Bidelman 2015). Therefore, it is conceivable that the unstable responses to speech in children with ASD could be improved in a positive way given the right type of auditory and language training. Given the beneficial impacts of music and music therapy on ASD (Geretsegger et al. 2014) and brain plasticity / cognition (Wan and Schlaug 2010), music might be a good place to start. The impact of such training would conceivably be measurable with the FFR.

Limitations and future directions

With a relatively small number of high-functioning ASD children, we were able to show lower neural stability compared to their TD peers, across multiple sounds. Further research will need to extend these findings with a larger group of children. In addition, a characterization of response stability of the FFR in children with ASD at different ages is merited to chart possible developmental trends. A lack of stability relative to typically developing children may be present at birth, or may not emerge until later in childhood. Perhaps it tracks with symptom development. Consideration of different levels of severity would help expand our understanding as well, as we only worked with high-functioning children, and did not perform internal validation of their diagnoses. Furthermore, these findings could be informed by future research examining connections between neural stability and behavioral measures, particularly including the development of language and social abilities, given their strong reliance upon the auditory system.

In addition, there are inherent difficulties in working with retrospective data analyses. In particular, for this study, we were required to compare across data collected on different system, with different parameters. However, while these differences do mean that comparisons between stimuli should be handled with caution, we believe that they strengthen the main finding of differences in stability between ASD and TD groups. The fact that neural stability was lower in ASD children despite the differences in recording parameters and systems implies that it is a broad result independent of these specifics. However, a future study informed by these results could ensure that our results are consistent while using the same system and recording parameters.

Finally, given the strong theoretical relationship of these findings to the previous studies of reduced stability (higher variability) in the ASD individuals, future research could attempt to correlate these measures in one study, determining whether an individual with ASD who has an unstable FFR also has a higher EEG or fMRI variability in response to simple sensory stimuli. Doing so would help support or refute the idea that increased variability in ASD neurological measures is part of generally noisier neuronal connections.

Conclusions

We found FFRs to speech to be less stable in high-functioning children with ASD than typically developing children. This persists across multiple stimuli and aligns with previous work that has found ASD individuals to have greater variability in their sensory responses. The effects we see could have ramifications that go beyond the auditory system, given the highly-integrated nature of the generators of the FFR. Further research needs to be done to better characterize the FFR in children with ASD and other disorders and determine its possible clinical applications.

Acknowledgements We thank members of the Auditory Neuroscience Laboratory for their assistance with data collection, as well as Trent Nicol and Spencer Benjamin Smith for comments on an earlier draft of the manuscript. We would also like to acknowledge Nicole Russo's work in collecting the data used for these analyses. This work was supported by Knowles Hearing Center, Northwestern University.

Compliance with ethical standards

Conflict of Interest None of the authors have potential conflicts of interest to be disclosed.

References

American Psychiatric Association (2013) Diagnostic and statistical manual of mental disorders 5th edn. Arlington, VA

Anderson S, Parbery-Clark A, White-Schwoch T, Kraus N (2012) Aging affects neural precision of speech encoding. J Neurosci Off J Soc Neurosci 32:14156–14164. https://doi.org/10.1523/JNEUROSCI.2176-12.2012

Bailey PJ, Snowling MJ (2002) Auditory processing and the development of language and literacy. Br Med Bull 63:135–146. https://doi.org/10.1093/bmb/63.1.135

Banai K, Hornickel J, Skoe E et al (2009) Reading and subcortical auditory function. Cereb Cortex NY 19:2699–2707. https://doi.org/10.1093/cercor/bhp024

Baron-Cohen S, Belmonte MK (2005) Autism: a window onto the development of the social and the analytic brain. Annu Rev Neurosci 28:109–126. https://doi.org/10.1146/annurev.neuro.27.070203.144137

Boersma P (2006) Praat: doing phonetics by computer. http://www.praat.org/

Campbell T, Kerlin JR, Bishop CW, Miller LM (2012) Methods to eliminate stimulus transduction artifact from insert earphones during electroencephalography. Ear Hear 33:144–150. https://doi.org/10.1097/AUD.0b013e3182280353

Cerliani L, Mennes M, Thomas RM et al (2015) Increased functional connectivity between subcortical and cortical resting-state networks in autism spectrum disorder. JAMA Psychiatry 72:767–777

Chandrasekaran B, Kraus N (2010) The scalp-recorded brainstem response to speech: neural origins and plasticity. Psychophysiology 47:236–246. https://doi.org/10.1111/j.1469-8986.2009.00928.x

Coffey EBJ, Herholz SC, Chepesiuk AMP et al (2016) Cortical contributions to the auditory frequency-following response revealed by MEG. Nat Commun 7:11070. https://doi.org/10.1038/ncomms11070

Coskun MA, Varghese L, Reddoch S et al (2009) Increased response variability in autistic brains? Neuroreport 20:1543–1548. https://doi.org/10.1097/WNR.0b013e32833246b5

Dajani DR, Uddin LQ (2016) Local brain connectivity across development in autism spectrum disorder: A cross-sectional investigation. Autism Res Off J Int Soc Autism Res 9:43–54. https://doi.org/10.1002/aur.1494

Davis G, Plaisted-Grant K (2015) Low endogenous neural noise in autism. Autism Int J Res Pract 19:351–362. https://doi.org/10.1177/1362361314552198

Dinstein I, Thomas C, Humphreys K et al (2010) Normal movement selectivity in autism. Neuron 66:461–469. https://doi.org/10.1016/j.neuron.2010.03.034

Dinstein I, Heeger DJ, Lorenzi L et al (2012) Unreliable evoked responses in autism. Neuron 75:981–991. https://doi.org/10.1016/j.neuron.2012.07.026

Doupe AJ, Kuhl PK (1999) Birdsong and human speech: common themes and mechanisms. Annu Rev Neurosci 22:567–631. https://doi.org/10.1146/annurev.neuro.22.1.567

Engineer CT, Centanni TM, Im KW, Kilgard MP (2014) Speech sound discrimination training improves auditory cortex responses in a rat model of autism. Front Syst Neurosci 8:. https://doi.org/10.3389/fnsys.2014.00137

Geretsegger M, Elefant C, Mössler KA, Gold C (2014) Music therapy for people with autism spectrum disorder. Cochrane Database Syst Rev. https://doi.org/10.1002/14651858.CD004381.pub3

Geschwind DH (2011) Genetics of autism spectrum disorders. Trends Cogn Sci 15:409–416. https://doi.org/10.1016/j.tics.2011.07.003

Haigh SM, Heeger DJ, Dinstein I et al (2015) Cortical variability in the sensory-evoked response in autism. J Autism Dev Disord 45:1176–1190. https://doi.org/10.1007/s10803-014-2276-6

Hall JW (2006) New handbook for auditory evoked responses, 1 edn. Pearson, Boston

Hornickel J, Kraus N (2013) Unstable representation of sound: a biological marker of dyslexia. J Neurosci Off J Soc Neurosci 33:3500–3504. https://doi.org/10.1523/JNEUROSCI.4205-12.2013

Jeng F-C, Hu J, Dickman B et al (2011) Cross-linguistic comparison of frequency-following responses to voice pitch in American and Chinese neonates and adults. Ear Hear 32:699–707. https://doi.org/10.1097/AUD.0b013e31821cc0df

Jeste SS, Geschwind DH (2014) Disentangling the heterogeneity of autism spectrum disorder through genetic findings. Nat Rev Neurol 10:74–81. https://doi.org/10.1038/nrneurol.2013.278

King C, Warrier CM, Hayes E, Kraus N (2002) Deficits in auditory brainstem pathway encoding of speech sounds in children with learning problems. Neurosci Lett 319:111–115

Klatt DH (1980) Software for a cascade/parallel formant synthesizer. J Acoust Soc Am 67:971–995. https://doi.org/10.1121/1.383940

Kraus N, White-Schwoch T (2015) Unraveling the biology of auditory learning: a cognitive-sensorimotor-reward framework. Trends Cogn Sci 19:642–654. https://doi.org/10.1016/j.tics.2015.08.017

Kraus N, Slater J, Thompson EC et al (2014) Music enrichment programs improve the neural encoding of speech in at-risk children. J Neurosci Off J Soc Neurosci 34:11913–11918. https://doi.org/10.1523/JNEUROSCI.1881-14.2014

Krishnan A, Xu Y, Gandour JT, Cariani PA (2004) Human frequency-following response: representation of pitch contours in Chinese tones. Hear Res 189:1–12. https://doi.org/10.1016/S0378-5955(03)00402-7

Krishnan A, Xu Y, Gandour J, Cariani P (2005) Encoding of pitch in the human brainstem is sensitive to language experience. Cogn Brain Res 25:161–168. https://doi.org/10.1016/j.cogbrainres.2005.05.004

Krizman JL, Skoe E, Kraus N (2010) Stimulus rate and subcortical auditory processing of speech. Audiol Neurootol 15:332–342. https://doi.org/10.1159/000289572

Krizman J, Marian V, Shook A et al (2012a) Subcortical encoding of sound is enhanced in bilinguals and relates to executive function advantages. Proc Natl Acad Sci USA 109:7877–7881. https://doi.org/10.1073/pnas.1201575109

Krizman J, Skoe E, Kraus N (2012b) Sex differences in auditory subcortical function. Clin Neurophysiol Off J Int Fed Clin Neurophysiol 123:590–597. https://doi.org/10.1016/j.clinph.2011.07.037

Krizman J, Slater J, Skoe E et al (2015) Neural processing of speech in children is influenced by extent of bilingual experience. Neurosci Lett 585:48–53. https://doi.org/10.1016/j.neulet.2014.11.011

Markram H, Rinaldi T, Markram K (2007) The intense world syndrome—an alternative hypothesis for Autism. Front Neurosci 1:77–96. https://doi.org/10.3389/neuro.01.1.1.006.2007

McCann J, Peppé S (2003) Prosody in autism spectrum disorders: a critical review. Int J Lang Commun Disord 38:325–350. https://doi.org/10.1080/1368282031000154204

Milne E (2011) Increased intra-participant variability in children with autistic spectrum disorders: evidence from single-trial analysis of evoked EEG. Front Psychol 2:. https://doi.org/10.3389/fpsyg.2011.00051

Musacchia G, Sams M, Skoe E, Kraus N (2007) Musicians have enhanced subcortical auditory and audiovisual processing of speech and music. Proc Natl Acad Sci 104:15894–15898. https://doi.org/10.1073/pnas.0701498104

Nelson SB, Valakh V (2015) Excitatory/inhibitory balance and circuit homeostasis in autism spectrum disorders. Neuron 87:684–698. https://doi.org/10.1016/j.neuron.2015.07.033

Nomi JS, Uddin LQ (2015) Developmental changes in large-scale network connectivity in autism. NeuroImage Clin 7:732–741. https://doi.org/10.1016/j.nicl.2015.02.024

Peñagarikano O, Abrahams BS, Herman EI et al (2011) Absence of CNTNAP2 leads to epilepsy, neuronal migration abnormalities, and core autism-related deficits. Cell 147:235–246. https://doi.org/10.1016/j.cell.2011.08.040

Rosenhall U, Nordin V, Brantberg K, Gillberg C (2003) Autism and auditory brain stem responses. Ear Hear 24:206–214. https://doi.org/10.1097/01.AUD.0000069326.11466.7E

Rossignol DA, Frye RE (2014) Evidence linking oxidative stress, mitochondrial dysfunction, and inflammation in the brain of individuals with autism. Front Physiol 5:. https://doi.org/10.3389/fphys.2014.00150

Russo NM, Skoe E, Trommer B et al (2008) Deficient brainstem encoding of pitch in children with autism spectrum disorders. Clin Neurophysiol Off J Int Fed Clin Neurophysiol 119:1720–1731. https://doi.org/10.1016/j.clinph.2008.01.108

Russo N, Nicol T, Trommer B et al (2009) Brainstem transcription of speech is disrupted in children with autism spectrum disorders. Dev Sci 12:557–567. https://doi.org/10.1111/j.1467-7687.2008.00790.x

Russo NM, Hornickel J, Nicol T et al (2010) Biological changes in auditory function following training in children with autism spectrum disorders. Behav Brain Funct BBF 6:60. https://doi.org/10.1186/1744-9081-6-60

Skoe E, Kraus N (2010) Auditory brainstem response to complex sounds: a tutorial. Ear Hear 31:302–324. https://doi.org/10.1097/AUD.0b013e3181cdb272

Skoe E, Kraus N (2013) Musical training heightens auditory brainstem function during sensitive periods in development. Front Psychol https://doi.org/10.3389/fpsyg.2013.00622

Skoe E, Krizman J, Kraus N (2013) The impoverished brain: disparities in maternal education affect the neural response to sound. J Neurosci Off J Soc Neurosci 33:17221–17231. https://doi.org/10.1523/JNEUROSCI.2102-13.2013

Skoe E, Krizman J, Anderson S, Kraus N (2015) Stability and plasticity of auditory brainstem function across the lifespan. Cereb Cortex N Y N 1991 25:1415–1426. https://doi.org/10.1093/cercor/bht311

Song JH, Banai K, Kraus N (2008) Brainstem timing deficits in children with learning impairment may result from corticofugal origins. Audiol Neurootol 13:335–344. https://doi.org/10.1159/000132689

Supekar K, Uddin LQ, Khouzam A et al (2013) Brain hyper-connectivity in children with autism and its links to social deficits. Cell Rep 5:738–747. https://doi.org/10.1016/j.celrep.2013.10.001

Takahashi T, Yoshimura Y, Hiraishi H et al (2016) Enhanced brain signal variability in children with autism spectrum disorder during early childhood. Hum Brain Mapp 37:1038–1050. https://doi.org/10.1002/hbm.23089

Thomas MSC, Davis R, Karmiloff-Smith A et al (2016) The over-pruning hypothesis of autism. Dev Sci 19:284–305. https://doi.org/10.1111/desc.12303

Tierney A, Kraus N (2013) The ability to move to a beat is linked to the consistency of neural responses to sound. J Neurosci Off J Soc Neurosci 33:14981–14988. https://doi.org/10.1523/JNEUROSCI.0612-13.2013

Vasa RA, Mostofsky SH, Ewen JB (2016) The Disrupted connectivity hypothesis of autism spectrum disorders: time for the next phase in research. Biol Psychiatry Cogn Neurosci Neuroimaging 1:245–252. https://doi.org/10.1016/j.bpsc.2016.02.003

Wan CY, Schlaug G (2010) Music making as a tool for promoting brain plasticity across the life span. Neurosci Rev J Bringing Neurobiol Neurol Psychiatry 16:566–577. https://doi.org/10.1177/1073858410377805

Weiss MW, Bidelman GM (2015) Listening to the brainstem: musicianship enhances intelligibility of subcortical representations for speech. J Neurosci Off J Soc Neurosci 35:1687–1691. https://doi.org/10.1523/JNEUROSCI.3680-14.2015

White-Schwoch T, Nicol T, Warrier CM, Abrams DA, Kraus N (2016) Individual differences in human auditory processing: insights from single-trial auditory midbrain activity in an animal model. Cereb Cortex 27(11):5095–5115

Won H, Mah W, Kim E (2013) Autism spectrum disorder causes, mechanisms, and treatments: focus on neuronal synapses. Front Mol Neurosci 6:. https://doi.org/10.3389/fnmol.2013.00019