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PAPER

Animal, but not human, faces engage the distributed face network in adolescents with autism

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Abstract

Multiple hypotheses have been offered to explain the impaired face-processing behavior and the accompanying underlying disruptions in neural circuitry among individuals with autism. We explored the specificity of atypical face-processing activation and potential alterations to fusiform gyrus (FG) morphology as potential underlying mechanisms. Adolescents with high functioning autism (HFA) and age-matched typically developing (TD) adolescents were scanned with sMRI and fMRI as they observed human and animal faces. In spite of exhibiting comparable face recognition behavior, the HFA adolescents evinced hypo-activation throughout the face-processing system in response to unfamiliar human, but not animal, faces. They also exhibited greater activation in affective regions of the face-processing network in response to animal, but not human, faces. Importantly, this atypical pattern of activation in response to human faces was not related to atypical structural properties of the FG. This atypical neural response to human faces in autism may stem from abnormalities in the ability to represent the reward value of social (i.e. conspecific) stimuli.

Research highlights

- High functioning adolescents (HFA) with autism and age-matched typically developing (TD) adolescents were scanned with sMRI and fMRI as they observed human and animal faces.
- TD adolescents exhibited comparable activation to human and animal faces throughout the distributed face-processing neural network.
- HFA adolescents exhibited hypo-activation only to human, but not animal, faces, compared to the TD adolescents.
- Morphology in the fusiform gyri was comparable across the groups.

Introduction

The impaired development of face-processing behavior and the accompanying disruptions to the underlying neural circuitry are widely reported findings in the autism literature (e.g. Corbett, Carmean, Ravizza, Wendelken, Henry *et al*., 2009; Dalton, Nacewicz, Johnstone, Schaefer, Gernsbacher *et al*., 2005; Grelotti, Klin, Gauthier, Skudlarski, Cohen *et al*., 2005; Humphreys, Hasson, Avidan, Minshew & Behrmann, 2008; Pierce, Muller, Ambrose, Allen & Courchesne, 2001; Scherf, Luna, Minshew & Behrmann, 2010; Schultz, Gauthier, Klin, Fulbright, Anderson *et al*., 2000). These atypicalities are particularly apparent among children and adolescents. For example, although face recognition behavior typically improves through adolescence and early adulthood among typically developing (TD) individuals (O'Hearn, Schroer, Minshew & Luna, 2010), it fails to improve beyond childhood among individuals with high functioning autism (HFA) (Greimel, Schulte-Rüther, Kamp-Becker, Remschmidt, Herpertz-Dahlmann *et al*., 2014; O'Hearn *et al*., 2010; O'Hearn, Tanaka, Lynn, Fedor, Minshew *et al*., 2014). Similarly, the size of the fusiform face area (FFA) increases as a

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function of age from childhood to adolescence among TD individuals (Scherf, Behrmann, Humphreys & Luna, 2007; Golarai, Liberman, Yoon & Grill-Spector, 2010), yet it remains smaller and hypoactive in HFA adolescents compared to TD adolescents (Scherf *et al*., 2010). To date, there have been multiple hypotheses offered to explain these deficits. Here, we evaluated two alternative, but not mutually exclusive, theories using fMRI.

One potential explanation for this atypical face-related activation in autism is that it is derived from structural alterations in the fusiform gyrus that likely emerge early in development (Dziobek, Bahnemann, Convit & Heekeren, 2010; Raznahan, Toro, Daly, Robertson, Murphy *et al*., 2010; Trontel, Duffield, Bigler, Froehlich, Prigge *et al*., 2013; Toal, Bloemen, Deeley, Tunstall, Daly *et al*., 2009; Van Kooten, Palmen, Von Cappeln, Steinbusch, Korr *et al*., 2008). For example, in autism, alterations in left fusiform gyrus (FG) volume in children and adolescents (Trontel *et al*., 2013) and cortical thickness in adults (Dziobek *et al*., 2010) are negatively correlated with face memory abilities. This theory predicts that the fundamental structural alterations likely impact the full range of stimuli that are processed by the FG, rather than being limited to human faces in particular.

An alternative hypothesis is that face-processing difficulties stem from abnormalities in the ability to represent the reward value of social (i.e. conspecific) stimuli (Dawson, Carver, Meltzoff, Panagiotides, McPartland *et al*., 2002; Dawson, Webb & McPartland, 2005; Chevallier, Kohls, Troiani, Brodkin & Schultz, 2012). In turn, this leads to atypical tuning of neural regions that process social information about *conspecifics*, including the fusiform face area (FFA) and other face- and body-processing regions over the course of development (Schultz, 2005). In support of this hypothesis, there are hints in the literature that highly familiar stimuli, which may be particularly rewarding, do elicit more normal levels of activation, specifically in the FFA, in autism. For example, Grelotti *et al*. (2005) found that a 12-year-old boy with autism showed strong FFA activation while observing Digimon cartoon characters, which were objects of expertise for him. Another study reported comparable responses in the FFA among children and young adolescents with autism and TD individuals when they viewed personally familiar faces (e.g. own mother; Pierce & Redcay, 2008). However, there are no conditions that reportedly lead to more normal levels of activation in any of the other regions (beyond the FFA) of the distributed face-processing network in autism (Bookheimer, Wang, Scott, Sigman, Dapretto *et al*., 2008; Dapretto, Davies, Pfeifer, Scott, Sigman *et al*., 2006; Hadjikhani, Joseph, Snyder & Tager-Flusberg, 2007; Perlman, Hudac, Pegors, Minshew & Pelphrey, 2011; Pierce & Redcay, 2008). This complex set of findings leads to open questions about the full range of face properties that may or may not elicit more typical levels of neural activation in autism and the extent to which modulation of neural activity is evident in multiple neural regions or is restricted to the FG/FFA.

In the current study, we explored the extent to which atypical activation of the face-processing network is explained by either of these two mechanisms, structural alterations in FG morphology or human face-specific hypo-activation. To do so, we measured the morphologic characteristics (volume and cortical thickness) of the fusiform gyrus to determine whether face recognition behavior and/or neural responses to faces for HFA adolescents are related to aberrant morphology within these regions. We also measured neural responses to both human and animal faces. Faces of cats and dogs are ideal stimuli to evaluate the specificity of the atypical responses in the face-processing system in autism for two reasons. First, animal faces share similar perceptual features with human faces, although they are not conspecifics; thus they likely engage similar visuoperceptual processing strategies as do human faces (see Diamond & Carey, 1986). Second, a growing body of evidence suggests that animal faces (particularly dogs) evoke similar activation to human faces in the FFA (Blonder, Smith, Davis, Kesler/West, Garrity *et al*., 2004; Tong, Nakayama, Moscovitch, Weinrib & Kanwisher, 2000) and other regions involved in face processing (Anzellotti & Caramazza, 2014; Blonder *et al*., 2004; Stoeckel, Pailley, Gollub, Niemi & Evins, 2014; Yang, Bellgowan & Martin, 2012) in TD adults. Therefore, evaluating neural responses to animal and human faces in autism can help determine whether the neural circuitry involved in face processing is generally disrupted (regardless of the type of face) or is specifically affected in the processing of human faces.

We scanned HFA adolescents and age-matched TD adolescents as they observed unfamiliar human faces with a range of emotional expressions and eye gaze direction, animal faces, and common objects. We hypothesized that if there is a generalized deficit in the underlying neural system in autism, we would observe hypo-activation to both human and animal faces in the FFA (and potentially in other face-processing regions) and that FFA hypo-activation might be related to aberrant structural properties of the FG. In contrast, if the deficit in autism is highly specific to human faces, we would observe selective hypo-activation only in response to human, but not animal, faces in the FFA and other face-processing regions, and no obvious relationship with altered morphological brain characteristics.

Methods

Participants

Participants included 14 HFA adolescents (13 male, 1 female), ages 13–18 years (M = 15, SD = 2) and 14 TD adolescents (13 male, 1 female), ages 13–18 years (M = 15, SD = 2). One additional HFA adolescent was excluded due to excessive head motion. The groups were matched on sex, handedness, age [$t(26) = .23$, $p = .82$], Full Scale IQ [$t(26) = .53$, $p = .60$], Verbal IQ [$t(26) = .11$, $p = .5$], and Performance IQ [$t(26) = 1.37$, $p = .18$] (see Table 1). IQ was assessed in the HFA adolescents using the Weschler Abbreviated Scales of Intelligence (WASI; Wechsler, 1999) and in the TD adolescents using the Kauffman Brief Intelligence Test-2 (KBIT-2; Kaufman & Kaufman, 2004).

The diagnosis of autism was established using the Autism Diagnostic Interview-Revised (ADI-R; Lord, Rutter & Le Couteur, 1994), the Autism Diagnostic Observation Schedule-G (ADOS; Lord, Rutter, DiLavore & Risi, 2001), and expert clinical diagnosis (Minshew, 1996). The HFA adolescents were medically healthy; had no identifiable genetic, metabolic, or infectious etiology for their disorder; and were free of traumatic brain injury, seizures, attention deficit disorder, and depression. HFA participants were not asked to withhold medication prior to testing. TD participants were medically healthy, free of regular medication usage, had no history of autism, neurological or psychiatric illness, acquired brain injury, learning disabilities, developmental disabilities, school problems, or substance abuse in themselves or their first-degree relatives.

Adolescents were recruited from several participant databases to be part of a longitudinal study investigating the effects of visuoperceptual training. The data reported here are from the pre-training assessment. Written informed consent was obtained from the parents of the adolescents, along with written assent or consent from the adolescents, using procedures approved by the Internal Review Boards of Penn State University and Carnegie Mellon University.

Table 1 *Demographic characteristics for the adolescents with high functioning autism (HFA) and typically developing (TD) adolescents*

	HFA	TD
# of participants	14	14
Age (years)	15 (2)	15 (2)
Full Scale IQ[a]	112 (11)	110 (12)
Verbal IQ	109 (12)	109 (13)
Performance IQ	113 (13)	107 (10)
Handedness	12R / 2L	12R / 2L
Gender	13M / 1F	13M / 1F
ADOS Total	13 (3)	
ADOS Comm	5 (1)	
ADOS Social	9 (2)	

Note: [a]The HFA adolescents completed the Weschler Abbreviated Scale of Intelligence and the TD adolescents completed the Kauffman Brief Intelligence Test-2. SRS = Social Responsiveness Scale

Measures

Face recognition behavior

The upright and inverted versions of the Cambridge Face Memory Test (CFMT) were used to measure unfamiliar face recognition outside the scanner (Duchaine & Nakayama, 2006). This task has been used previously to measure face recognition abilities in TD and HFA adolescents (O'Hearn *et al.*, 2010). One HFA and one TD adolescent did not complete the inverted version of the task. Accuracy was collected as a measure of performance.

MRI acquisition

Prior to scanning, all participants were placed in a mock MR scanner for approximately 20 minutes and completed practice versions of the tasks that were administered in the full scan. This procedure acclimates participants to the scanner environment and minimizes motion artifact and anxiety.

Participants were scanned using a Siemens 3T Magnetom Trio whole body MRI scanner with a 12-channel head coil. High-resolution T1-weighted anatomical images (3D-MPRAGE) were acquired with 176 sagittal slices (TR/TE/TI = 1700, 1.78, 850 ms; voxel size = 1 mm^3, FOV = 256 × 256). Functional EPI images were acquired in 34 slices (3 mm isotropic voxels; TR/TE = 2000, 25 ms; FOV = 210; matrix 70 × 70; Flip angle = 80°). The functional images were aligned approximately 30° in the rostral direction from the AC-PC line, which minimizes noise from the eye orbits and nasal sinuses and maximizes signal in the medial temporal lobes (Whalen, Johnstone, Somerville, Nitschke, Polis *et al.*, 2008). This protocol allowed for full coverage of the ventral visual pathway as well as of the frontal and occipital lobes. For participants with larger head size, the superior parietal lobe was not completely covered. High-resolution structural images and functional images were acquired in a single session.

fMRI human–animal face task

Participants observed color pictures from four categories of human faces (fearful, happy, and neutral expressions with direct eye gaze, and neutral expressions with averted

Figure 1 *Examples of stimuli from each visual category represented in the fMRI human–animal face task: fearful faces, happy faces, neutral faces with direct gaze, neutral faces with averted gaze, common objects, and animal faces. Images are presented in greyscale here for the purposes of presentation but were presented in color to the participants.*

eye gaze), common objects (inanimate objects), and animal faces (cats/dogs) (see Figure 1). Images of human faces were selected from several existing databases (Langer, Dotsch, Gijsbert, Wigboldus, Hawk *et al.*, 2010; Thomaz & Giraldi, 2010; Tottenham, Tanaka, Leon, McCarry, Nurse *et al.*, 2009; pics.stir.ac.uk). Some of the human face identities repeated across categories (e.g. fearful and happy faces), but no exact image of a face ever repeated. Images of objects and animals were downloaded from the Internet. Images were cropped and resized to 280 × 320 pixels.

The task was a block design with six 12-second blocks of each visual category, the order of which was randomized for each participant, interleaved with 6-second fixation blocks. Within each task block, 12 images were each presented for 800 ms followed by a 200 ms fixation. Participants completed a 1-back memory task while viewing the pictures and responded by button press when they saw a picture repeat. Two images repeated in each stimulus block, the order of which was counterbalanced across blocks. Accuracy was collected as a measure of task performance. The duration of the task was 9 min 42 seconds.

Data analysis

Behavioral data

For the 1-back scanner task, both misses and false alarms were counted as errors in calculating accuracy. Accuracy scores for each participant on both the CFMT and 1-back memory tasks were submitted to separate repeated-measures ANOVAs, with stimulus category as the within-subject factor and group as the between-subject factor.

fMRI data

The neuroimaging data were analyzed using Brain Voyager QX v2.3 (Brain Innovation, Maastricht, The Netherlands). Preprocessing included 3D-motion correction and filtering out low frequencies up to three cycles. Participants' data were rejected if motion in any of the six directions exceeded 3 mm for any TR in the functional run. The average motion in each group was less than 1 mm in all six directions and did not differ between groups ($p > .4$); thus any group effects were not the result of motion differences.

For each participant, the time series images for each brain volume were analyzed for category differences in a fixed-factor GLM. Each category was defined as a separate predictor and modeled with a box-car function adjusted for the delay in hemodynamic response. The functional data were not spatially smoothed, following previous recommendations (Weiner & Grill-Spector, 2012). The timeseries images were spatially normalized into Talairach space, which is common practice in autism neuroimaging research (Redcay & Courschesne, 2005; Scherf *et al.*, 2010).

Whole-brain group differences

To examine potential group differences in human face and animal face activation, the fMRI data from the TD and HFA groups were directly compared in a whole-brain voxelwise mixed-model ANOVA including group and visual category as fixed factors and subject as a random factor. Group differences for human face-related activation were examined using the following balanced interaction: [TD (all human faces – 4*objects) > HFA (all human faces – 4*objects)]. Group differences for animal face activation were examined with the following interaction: [TD (animal faces – objects) > HFA (animal faces – objects)]. A Monte Carlo simulation was used to correct the resulting interaction maps for false positive activation (corrected $p < .05$ required a minimum of 31 contiguous voxels at a t-value ≥ 2.0). To quantify and graph group differences in category selectivity, beta weights for each condition for each individual participant were extracted from each

significant ROI and difference scores were plotted for human (average of human face categories – objects) and animal (animal – objects) faces.

Individually defined fusiform face area

Given our *a priori* predictions about group differences in the profile of activation for the FFA, individually defined FFA regions were demarcated separately for human and animal faces in each hemisphere in each participant (Scherf *et al.*, 2010; Weiner & Grill-Spector, 2012). Selectivity for human faces was defined in a balanced contrast: [(happy + fearful + neutral-direct + neutral-averted human faces) − (4*objects)]. Animal face selectivity was defined as: [(animal faces) − (objects)]. Each resulting individual contrast map was corrected for false positive activation using the False Discovery Rate procedure (Genovese, Lazar & Nichols, 2002) with a $q < .05$. The right and left FFA ROIs were defined as the most anterior cluster of contiguous significant voxels in the fusiform gyrus (FG) separately for each participant's human-selective and animal-selective activation, and were quantified in terms of the number of voxels (e.g, ROI size). These ROIs were defined to ensure that the BOLD signal comparison between groups was derived from the optimal (i.e. most selective) region on a case-by-case basis rather than superimposing a group-defined ROI on each brain. This approach allowed us to differentially optimize the BOLD signal for human and animal faces separately. Therefore, any emerging group differences in BOLD response resulting from these individual ROI analyses cannot be attributed to partial volume effects or differences in ROI definition across the two groups.

Within each individually defined ROI for each participant, a separate ROI-based GLM was conducted that modeled stimulus condition as a fixed factor. The resulting beta weights were extracted for each of the six visual categories. Beta weight difference scores were computed to examine the magnitude of face-related activation in the FFA for human faces (average of human face categories – objects) and animal faces (animal faces – objects). The size and the beta weight difference scores were then submitted to separate repeated-measures ANOVAs with the factors of face category (human, animal), hemisphere (right, left), and group (HFA, TD).

Independently defined fusiform face area

To confirm the results from the individually defined ROI analysis, beta weights were also extracted from an independently defined right FFA ROI. This ROI was identified in previous work evaluating human face-related activation in TD adolescents and adults. We centered a 4 mm sphere on the functionally defined Talairach coordinates for the right FFA (40, −41, −21) reported in a previous study (Scherf *et al.*, 2007).

sMRI analyses

To evaluate whether structural differences might account for observed functional differences, we extracted FG volume and cortical thickness for each participant in each hemisphere, as well as whole-brain volume. The structural data were analyzed using the standard cortical reconstruction pipeline in FreeSurfer v5.3.0 (surfer.nmr.mgh.harvard.edu), including standardization to a common atlas (Fischl, Salat, Busa, Albert, Dieterich *et al.*, 2002), which is a validated approach for use with children (Ghosh, Kakunoori, Augustinack, Nieto-Castanon, Kovelman *et al.*, 2010). Once the cortical model was completed for each participant, the cerebral cortex was parcellated into units based on gyral and sulcal structure, creating surface-based maps of gyral curvature and sulcal depth (Fischl, Sereno & Dale, 1999). Calculations of cortical and subcortical volumes using this method are comparable to manual segmentation (e.g. Lehmann, Douiri, Kim, Modat, Chan *et al.*, 2010). We normalized the FG volumes for each participant using their whole-brain volume (e.g. [region/whole brain volume]*1000). Whole brain volume was calculated as the SupraTentorial volume, excluding ventricles, CSF, and choroid plexus. The structural measures were submitted to separate ANOVAs, with the factors of group and hemisphere.

Structure–function correlations

To test the hypothesis that structural properties of the left and right FG may predict atypical behavior or functional activation for HFA adolescents or TD adolescents, Pearson product correlations were conducted between the structural and functional measures obtained for these regions separately for each group. We examined the relations between structural properties of the FG (volume and cortical thickness) with FFA size (separately for human and animal faces) and activation (beta weight difference scores). These correlations were conducted separately within each hemisphere. For each significant correlation, we evaluated robustness in separate bootstrap analyses using 1000 iterations to obtain a 95% confidence interval (CI).

Results

Behavioral tasks

On the CFMT, both groups exhibited a face inversion effect; there was a main effect of orientation, with higher performance for upright than inverted faces, $F(1, 24) = 83.78$, $p < .001$, $\eta^2 = .78$. There was no main effect of group nor an interaction between group and orientation (Supplemental Figure 1a). During the scanning task, recognition performance was comparable across the two groups for all conditions and there were no main effects or interactions (all $p > .05$) (Supplemental Figure 1b). Therefore, group differences in the BOLD response cannot be attributed to differences in behavioral performance.

Whole-Brain Group Differences

Human face activation comparison

In spite of the comparable face recognition behavior across the groups, the TD adolescents exhibited greater human face activation than HFA adolescents in the right FFA, right occipital face area (OFA), left amygdala, left putamen, and the posterior cingulate cortex (PCC) (Figure 2). There were no regions where HFA adolescents showed greater activation than TD adolescents for human faces (Table 2). Figure 2 shows the activation for human and animal faces extracted from the right FFA, right OFA, left amygdala, and PCC. In each of these regions there was a group difference for human faces (by definition). The key question concerns whether there

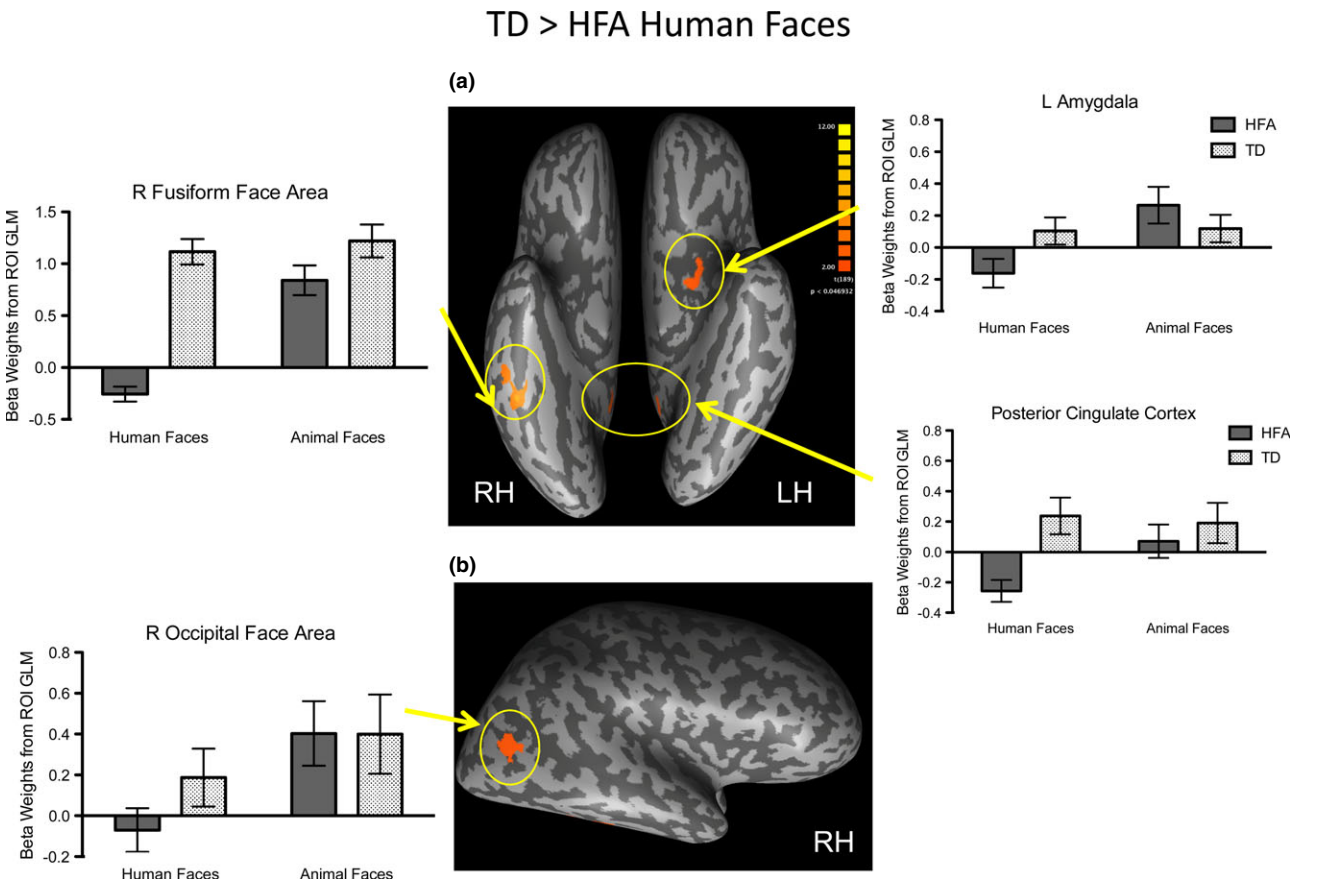

Figure 2 *Group differences for HFA adolescents and TD adolescents in response to human faces compared to objects. The maps were generated by computing a whole-brain analysis, corrected at p < .05. Adolescents with HFA exhibited less activation than TD adolescents for human faces compared to objects in the left putamen (not pictured), (a) right fusiform face area, left amygdala, posterior cingulate cortex, and (b) the right occipital face area. The mean (SEM) beta weight difference scores (human faces minus objects) and (animal faces minus objects) were extracted from these regions. Adolescents with HFA show hypo-activation for human faces, but not animal faces, in these regions.*

Table 2 *Results from whole-brain comparisons between TD and HFA adolescents for human and animal face activation*

Group	Contrast	Region	Hemisphere	Size (#voxels)	Talairach coordinates X	Y	Z
TD > HFA	Human > object	FFA	R	940	41	−41	−22
		OFA	R	1642	45	−65	8
		Amygdala	L	417	−20	3	−13
		Putamen	L	870	−22	4	0
		PCC		2327	15	−51	−0
TD > HFA	Animal > Object	*ns*					
HFA > TD	Human > Object	*ns*					
HFA > TD	Animal > Object	*ns*					

were group differences for animal faces. Interestingly, there were no significant differences between HFA adolescents and TD adolescents for the beta-weights extracted for animal faces in any of these ROIs (p = *ns*). In addition, while TD adolescents did not show differences between human and animal activation in these ROIs, the HFA adolescents showed significantly reduced activation for human compared to animal faces (all $p < .05$), indicating that the hypo-activation was *specific to human faces.*

Animal face activation comparison

The whole-brain analysis contrasting group responses to animal faces revealed no regions of significant difference between HFA and TD adolescents for animal face-selective activation.

Individually defined fusiform face area

FFA size

Figure 3a–b shows the size of the human- and animal-defined right and left FFA ROIs for the two groups. There were no main effects of group or of hemisphere. However, there were interactions between face category × group, $F(1, 26) = 4.75$, $p < .05$, $\eta^2 = .15$, and between hemisphere × group, $F(1, 26) = 89.99$, $p < .01$, $\eta^2 = .28$. To investigate the face category × group interaction, we evaluated category effects within each group collapsed across hemisphere. Across both hemispheres, the HFA adolescents had smaller human- than animal-defined FFA regions, $t(13) = -2.17$, $p < .05$. Among TD adolescents, the size of the human-defined and animal-defined FFA regions did not differ (p = *ns*). To investigate the hemisphere by group interaction, we evaluated hemisphere effects within each group collapsed across face category. HFA adolescents had larger left than right FFA regions, $t(13) = -2.30$, $p < .05$. In contrast, TD adolescents, had larger right than left FFA regions, $t(13) = 2.55$, $p < .05$.

FFA magnitude

Figure 3c–d shows the beta weight difference scores in the human-defined bilateral FFA regions, for both human faces and animal faces. There was a main effect of face category, $F(1, 25) = 6.21$, $p < .05$, $\eta^2 = .19$. Surprisingly, *animal faces* elicited higher magnitude responses than did human faces in the human-defined FFA. However, this was qualified by a face category × group interaction, $F(1, 25) = 11.06$, $p < .01$, $\eta^2 = .31$, and no main effect or interactions with hemisphere. To investigate this interaction, we evaluated the effect of face category within each group. HFA adolescents had lower activation for human than animal faces in their human-defined FFA regions, $t(12) = 3.37$, $p < .05$. In contrast, TD adolescents showed comparable activation for human and animal faces in the FFA (p = *ns*).

Independently defined right fusiform face area

Within the independently defined right FFA, HFA adolescents showed hypo-activation for human faces compared to TD adolescents, $t(26) = -3.00$, $p < .01$, but not for animal faces, replicating the pattern found for the whole-brain analysis (Figure 3e).

Structural MRI

For all ROIs, there was no main effect of group, no main effect of hemisphere, and no interaction between group and hemisphere (p = *ns*) (Supplemental Figure 2a–c).

Structure–function correlations

For HFA adolescents, in the right hemisphere, there were no correlations between FG structure and either behavior or functional activation. In the left hemisphere, FG cortical thickness correlated with the size of the left FFA ROI for animal faces, $r(14) = .65$, $p = .012$, (bootstrap analysis: $r = .63$, 95% CIs of .34/.92, which is different from 0 at $p < .05$), but not for human faces (Figure 4).

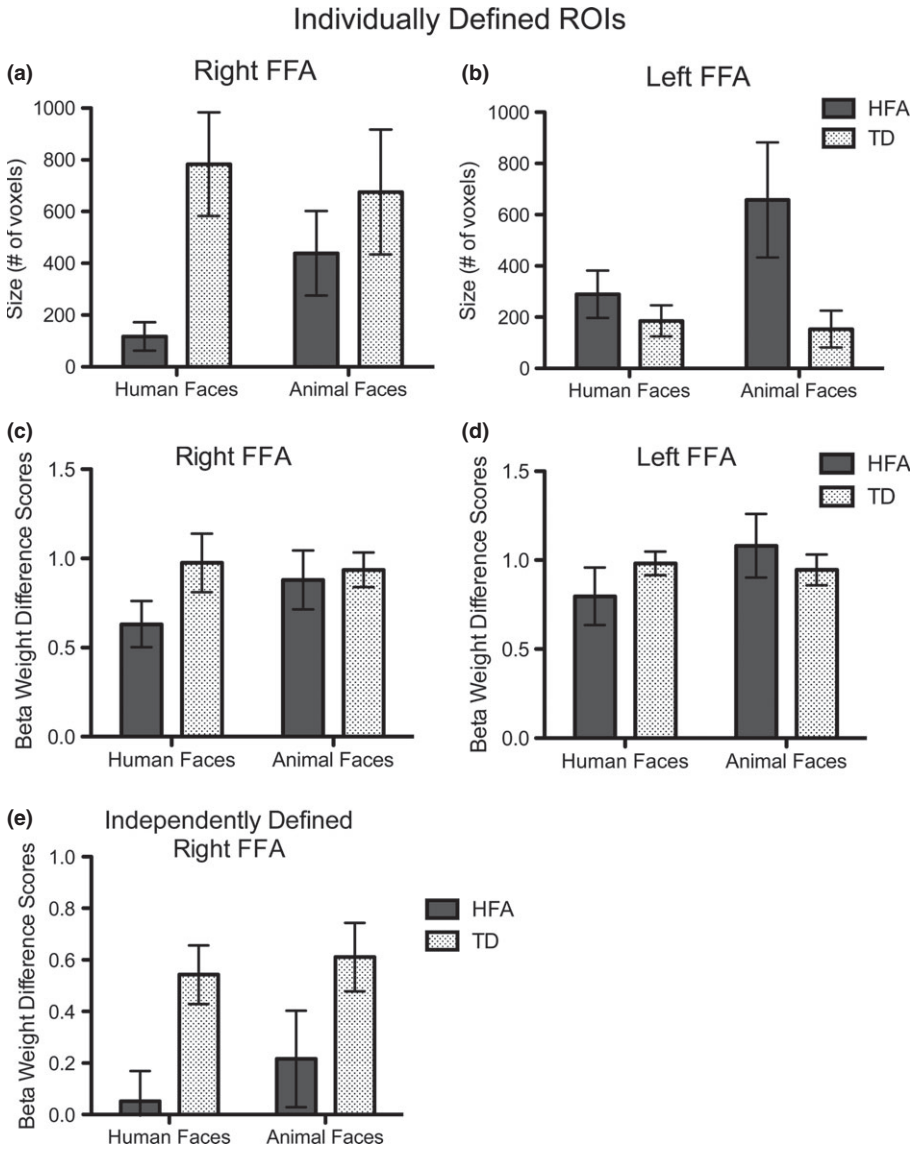

Figure 3 *Mean (SEM) number of significantly active voxels for the individually defined (a) right FFA and (b) left FFA regions, for human-defined (all human faces versus objects), and animal-defined (animal faces versus objects) regions for HFA adolescents and TD adolescents. Mean (SEM) beta weight difference scores (human faces minus objects, and animal faces minus objects) from ROI GLMs for the individually human-defined (c) right FFA and (d) left FFA ROIs for HFA adolescents and TD adolescents. (e) Mean (SEM) beta weight difference scores (human faces minus objects, and animal faces minus objects) from ROI GLMs for the independently defined right FFA (from Scherf et al., 2007).*

No other measures correlated with left FG volume or cortical thickness (Table 3). There were no significant correlations in either hemisphere in the TD adolescents.

Discussion

The current study examined the specificity of atypical face-processing activation and FG morphology among HFA and TD adolescents. We evaluated neural responses to animal and human faces in autism to help determine whether the neural circuitry involved in face processing is generally disrupted (regardless of the type of face) or is specifically affected in the processing of human faces. Cat and dog faces were ideal comparison stimuli to human faces because they likely engage similar visuo-perceptual processing strategies as do human faces and they activate the distributed face-processing network in

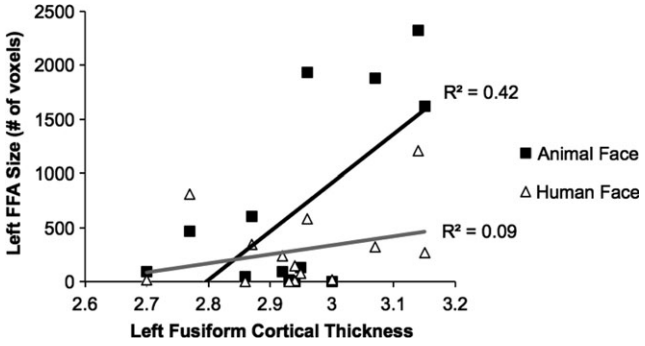

Figure 4 *Correlations for adolescents with autism between the left fusiform gyrus (FG) cortical thickness and the size (# of voxels) in the independently-defined left FFA for human faces (n.s.) and animal faces (r = .65, p < .05).*

Table 3 *Correlations between fusiform gyrus structure (volume and cortical thickness) and CFMT accuracy, FFA size and FFA activation (beta weight difference scores). All correlations were conducted within hemisphere*

	R. fusiform volume	R. fusiform cortical thickness	L. fusiform volume	L. fusiform cortical thickness
CFMT accuracy	0.05	0.01	0.17	0.12
FFA human size	0.39	0.12	0.06	0.30
FFA animal size	0.33	0.44	0.13	**0.65***
FFA human activation	−0.40	−0.12	−0.25	0.00
FFA animal activation	−0.42	−0.06	−0.45	0.25

Note: *p = .012.

TD adults (Anzellotti & Caramazza, 2014; Blonder *et al.*, 2004; Tong *et al.*, 2000). As in TD adults, we observed that TD adolescents engage both core and extended face-processing regions when recognizing human and cat/dog faces; there were no differences for TD adolescents when viewing human versus animal faces in any analyses.

Most important, we observed converging evidence (across whole-brain, individually defined, and independently-defined ROI analyses) of the specificity of right FFA hypo-activation in response to human, but not animal, faces in HFA adolescents. Our findings of reduced activation for the HFA group in both core (right FFA and right OFA) and extended (left amgydala) regions when viewing human faces are consistent with and extend previous findings (e.g. Grelotti *et al.*, 2005; Pierce & Redcay, 2008; Ashwin, Baron-Cohen, Wheelwright, O'Riordan & Bullmore, 2007; Kohls, Schulte-Ruther, Nehrkorn, Muller, Fink *et al.*, 2013).

The equivalent activation for both groups in response to animal faces stands out from previous findings in that we show conditions under which normal activation can be elicited when the stimuli are not personally familiar to the participant. In other words, our findings reveal that personal familiarity is not a prerequisite stimulus property for eliciting normal responses in the face-processing neural network in autism. In addition, we report a novel finding of hypo-activation in the PCC as HFA adolescents observed *unfamiliar* faces. This hypo-activation could reflect relative difficulty among the HFA adolescents in recognizing that the identities of the faces repeated across conditions in the experiment (e.g. same identity faces in the fearful and angry conditions and in the averted and direct eye gaze conditions), given the reported role of the PCC in acquiring facial familiarity (Kosaka, Omori, Iidaka, Murata, Shimoyama *et al.*, 2003).

Critically, this study is the first to report normal levels of face-related activation in adolescents with HFA in response to faces that are not those of conspecifics. HFA adolescents only exhibited TD-like levels of activation in response to animal, but not human, faces. Thus, these findings show that the hypo-activation in the face-processing system is *specific to human faces* in autism. Furthermore, weakened human face-related activation in adolescents with autism was not related to atypical structural properties of the FG. This is evident from the results of the morphometric analyses, which show comparable volume and cortical thickness in the right FG in both groups of adolescents, as well as the robust activation that was observed in this region in response to animal faces. Together, these results suggest that there are no gross structural anomalies (that can be measured with sMRI at the millimeter level) that fundamentally limit the development of face-selective activation in this region of cortex in autism. While it is possible that a small sample size may have limited the ability to detect structural differences, we were able to find differences in functional activation when viewing human faces, suggesting that functional differences are present in the absence of structural differences in this sample. In addition, recent findings in a much larger sample (n = 295) of 6–65-year-olds also failed to find differences in FG structural morphology in individuals with autism (Haar, Berman, Behrmann & Dinstein, 2014).

With respect to our original hypotheses, our findings are consistent with the notion that in autism, human faces are not imbued with the same reward value or processed with the same attentional and motivational

resources as they are in TD individuals (Chevallier *et al.*, 2012; Schultz, 2005). Our findings of stronger responses to animal faces in this same network may indicate that individuals with autism find animal faces more socially rewarding than human faces. There is behavioral evidence that is consistent with this notion. Individuals with autism often show strong motivational preferences and social behaviors directed towards common animals such as cats and dogs (Carlisle, 2014; Celani, 2002; O'Haire, McKenzie, Beck & Slaughter, 2013; Prothmann, Ettrich & Prothmann, 2009). In support of this interpretation, we found greater activation in areas related to reward and emotional arousal (amygdala, putamen) in response to animal faces but not human faces among the HFA adolescents. Future work investigating the association between perceived reward value of both human and animal faces and category-selective neural activation is needed to directly evaluate this interpretation. In addition, future research using eye-tracking technology may provide specific information about whether and how visually attention to human and animal faces is deployed differently and is or is not related to the differential patterns of activation in either the visuoperceptual and/or affective regions of the face-processing network.

There were no differences in human face-processing behavior between the HFA and TD adolescents. In spite of the comparable behavior, the adolescents with autism still showed reduced activation for human faces, particularly in the right hemisphere. Also, the two groups exhibited different patterns of laterality in face activation. The TD adolescents exhibited right hemisphere laterality for both human and animal faces, while the HFA adolescents exhibited left hemisphere laterality. Together, this pattern of results suggests that the HFA adolescents may be using different strategies from the TD adolescents to recognize both human and animal faces. We suggest that these processing strategies may rely disproportionately on the left hemisphere. This hypothesis is informed by the results from the structure–function analyses. Among the HFA adolescents, there was a relation between the cortical thickness of the left FG and the size of the animal face defined functional ROI. All together, these results may reflect individual differences in the extent of cortical thinning (or rather, lack thereof) during development that have consequences for and/or are related to the emerging specialization for faces in the autism brain. Consistent with this notion are previous findings that children with autism do not begin to show a left visual hemifield advantage for processing both human and dog faces that young TD children do (consistent with emerging right-lateralization for face processing) (Guillon, Hadjikhani, Baduel, Kruck, Arnaud *et al.*, 2014). Longitudinal data will determine whether this left lateralization and heightened selectivity for animal faces is contributing to the development of hypo-activation in the face-processing system during human face processing, is a compensatory response, or is even unrelated to this hypo-activation.

In conclusion, the current study found that adolescents with autism evince consistent hypo-activation throughout the face-processing network specifically when viewing human, but not animal, faces. Group differences in social motivation specific to human faces may be central to this atypical neural response in autism. Future research should examine how motivational salience impacts activation of face-processing regions for both individuals with autism and typical development.

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References

Anzellotti, S., & Caramazza, A. (2014). Individuating the neural bases for the recognition of conspecifics with MVPA. *NeuroImage*, **89**, 165–170.

Ashwin, C., Baron-Cohen, S., Wheelwright, S., O'Riordan, M., & Bullmore, E.T. (2007). Differential activation of the amygdala and the 'social brain' during fearful face processing in asperger syndrome. *Neuropsychologia*, **45** (1), 2–14.

Blonder, L.X., Smith, C.D., Davis, C.E., Kesler/West, M.L., & Garrity, T.F. *et al.* (2004). Regional brain response to faces of humans and dogs. *Cognitive Brain Research*, **20**, 384–394.

Bookheimer, S.Y., Wang, A.T., Scott, A., Sigman, M., Dapretto, M. *et al.* (2008). Frontal contributions to face processing differences in autism: evidence from fMRI of inverted face processing. *Journal of the International Neuropsychological Society*, **14** (06), 922–932.

Carlisle, G.K. (2014). Pet ownership decisions for parents of children with autism spectrum disorder. *Journal of Pediatric Nursing*, **29** (2), 114–123.

Celani, G. (2002). Human beings, animals and inanimate objects: what do people with autism like? *Autism*, **6** (1), 93–102.

Chevallier, C., Kohls, G., Troriani, V., Brodkin, E.S., & Schultz, R.T. (2012). The social motivation theory of autism. *Trends in Cognitive Sciences*, **16** (4), 231–239.

Corbett, B., Carmean, V., Ravizza, S., Wendelken, C., Henry, M. *et al.* (2009). A functional and structural study of emotion and face processing in children with autism. *Psychiatry Research: Neuroimaging*, **173** (3), 196–205.

Dalton, K.M., Nacewicz, B.M., Johnstone, T., Schaefer, H.S., Gernsbacher, M.A. *et al.* (2005). Gaze fixation and the neural circuitry of face processing in autism. *Nature Neuroscience*, **8** (4), 519–526.

Dapretto, M., Davies, M.S., Pfeifer, J.H., Scott, A.A., Sigman, M. *et al.* (2006). Understanding emotions in others: mirror neuron dysfunction in children with autism spectrum disorders. *Nature Neuroscience*, **9** (1), 28–30.

Dawson, G., Carver, L., Meltzoff, A.N., Panagiotides, H., McPartland, J. *et al.* (2002). Neural correlates of face and object recognition in young children with autism spectrum disorder, developmental delay, and typical development. *Child Development*, **73** (3), 700–717.

Dawson, G., Webb, S.J., & McPartland, J. (2005). Understanding the nature of face processing impairment in autism: insights from behavioral and electrophysiological studies. *Developmental Neuropsychology*, **27** (3), 403–424.

Diamond, R., & Carey, S. (1986). Why faces are and are not special: an effect of expertise. *Journal of Experimental Psychology*, **115** (2), 107–117.

Duchaine, B., & Nakayama, K. (2006). The Cambridge face memory test: results for neurologically intact individuals and an investigation of its validity using inverted face stimuli and prosopagnosic participants. *Neuropsychologia*, **44** (4), 576–585.

Dziobek, I., Bahnemann, M., Convit, A., & Heekeren, H.R. (2010). The role of the fusiform-amygdala system in the pathophysiology of autism. *Archives of General Psychiatry*, **67** (4), 397–405. doi:10.1001/archgenpsychiatry.2010.31

Fischl, B., Salat, D.H., Busa, E., Albert, M., Dieterich, M. *et al.* (2002). Whole brain segmentation: automated labeling of neuroanatomical structures in the human brain. *Neuron*, **33** (3), 341–355.

Fischl, B., Sereno, M.I., & Dale, A.M. (1999). Cortical surface-based analysis. II: Inflation, flattening, and a surface-based coordinate system. *NeuroImage*, **9** (2), 195–207.

Genovese, C.R., Lazar, N.A., & Nichols, T. (2002). Thresholding of statistical maps in functional neuroimaging using the false discovery rate. *NeuroImage*, **15** (4), 870–878.

Ghosh, S.S., Kakunoori, S., Augustinack, J., Nieto-Castanon, A., Kovelman, I. *et al.* (2010). Evaluating the validity of volume-based and surface-based brain image registration for developmental cognitive neuroscience studies in children 4 to 11 years of age. *NeuroImage*, **53** (1), 85–93.

Golarai, G., Liberman, A., Yoon, J.M., & Grill-Spector, K. (2010). Differential development of the ventral visual cortex extends through adolescence. *Frontiers in Human Neuroscience*, **3**, 80.

Greimel, E., Schulte-Rüther, M., Kamp-Becker, I., Remschmidt, H., Herpertz-Dahlmann, B. *et al.* (2014). Impairment in face processing in autism spectrum disorder: a developmental perspective. *Journal of Neural Transmission*, **121** (9), 1171–1181.

Grelotti, D.J., Klin, A.J., Gautheir, I., Skudlarski, P., Cohen, D.J. *et al.* (2005). fMRI activation of the fusiform gyrus and amygdala to cartoon characters but not to faces in a boy with autism. *Neuropsychologia*, **43** (4), 373–385.

Guillon, Q., Hadjikhani, N., Baduel, S., Kruck, J., Arnaud, M. *et al.* (2014). Both dog and human faces are explored abnormally by young children with autism spectrum disorders. *NeuroReport*, **25** (15), 1237–1241.

Haar, S., Berman, S., Behrmann, M., & Dinstein, I. (2014). Anatomical abnormalities in autism? *Cerebral Cortex*. Advance online publication. doi:10.1093/cercor/bhu242

Hadjikhani, N., Joseph, R.M., Snyder, J., & Tager-Flusberg, H. (2007). Abnormal activation of the social brain during face perception in autism. *Human Brain Mapping*, **28** (5), 441–449.

Humphreys, K., Hasson, U., Avidan, G., Minshew, N., & Behrmann, M. (2008). Cortical patterns of category-selective activation for faces, places and objects in adults with autism. *Autism Research*, **1** (1), 52–63.

Kaufman, A.S., & Kaufman, N.L. (2004). *Kaufman Brief Intelligence Test* (2nd edn.). Circle Pines, MN: AGS Publishing.

Kohls, G., Schulte-Ruther, M., Nehrkorn, B., Muller, K., Fink, G.R. *et al.* (2013). Reward system dysfunction in autism spectrum disorders. *Social Cognitive and Affective Neuroscience*, **8** (5), 565–572.

Kosaka, H., Omori, M., Iidaka, T., Murata, T., Shimoyama, T. *et al.* (2003). Neural substrates participating in acquisition of facial familiarity: an fMRI study. *NeuroImage*, **20**, 1734–1742.

Langer, O., Dotsch, R., Gijsbert, B., Wigboldus, D.H.J., Hawk, S.T. *et al.* (2010). Presentation and validation of the Radboud Faces Database. *Cognition and Emotion*, **24** (8), 1377–1388.

Lehmann, M., Douiri, A., Kim, L.G., Modat, M., Chan, D. *et al.* (2010). Atrophy patterns in Alzheimer's disease and semantic dementia: a comparison of FreeSurfer and manual volumetric measurements. *NeuroImage*, **49** (3), 2264–2274.

Lord, C., Rutter, M., DiLavore, P.C., & Risi, S. (2001). *Autism Diagnostic Observation Schedule (ADOS)*. Lost Angeles, CA: Western Psychological Services.

Lord, C., Rutter, M., & Le Couteur, A. (1994). Autism diagnostic interview-revised: a revised version of a diagnostic interview for caregivers of individuals with possible pervasive developmental disorders. *Journal of Autism and Developmental Disorders*, **24** (5), 659–685.

Minshew, N. (1996). Autism. In B.O. Berg (Ed.), *Principles of child neurology* (pp. 1713–1729). New York: McGraw-Hill.

O'Haire, M.E., McKenzie, S.J., Beck, A.M., & Slaughter, V. (2013). Social behaviors increase in children with autism in the presence of animals compared to toys. *Plos ONE*, **8** (2), e57010.

O'Hearn, K., Schroer, E., Minshew, N., & Luna, B. (2010). Lack of developmental improvement on a face memory task

during adolescence in autism. *Neuropsychologia*, **48** (13), 3955–3960.

O'Hearn, K., Tanaka, J., Lynn, A., Fedor, J., Minshew, N. *et al.* (2014). Developmental plateau in visual object processing from adolescence to adulthood in autism. *Brain and Cognition*, **90**, 124–134. doi:10.1016/j.bandc.2014.06.004

Perlman, S.B., Hudac, C.M., Pegors, T., Minshew, N.J., & Pelphrey, K.A. (2011). Experimental manipulation of face-evoked activity in the fusiform gyrus of individuals with autism. *Social Neuroscience*, **6** (1), 22–30.

Pierce, K., Muller, R.A., Ambrose, J., Allen, G., & Courchesne, E. (2001). Face processing occurs outside the fusiform 'face area' in autism: evidence from functional MRI. *Brain*, **124**, 2059–2073.

Pierce, K., & Redcay, E. (2008). Fusiform function in children with autism spectrum disorder is a matter of 'who'. *Biological Psychiatry*, **64** (7), 552–560.

Prothmann, A., Ettrich, E., & Prothmann, S. (2009). Preference for, and responsiveness to, people, dogs, and objects in children with autism. *Anthrozoos: A Multidisciplinary Journal of the Interactions of People and Animals*, **22** (2), 161–171.

Raznahan, A., Toro, R., Daly, E., Robertson, D., Murphy, C. *et al.* (2010). Cortical anatomy in autism spectrum disorder: an in vivo MRI study on the effect of age. *Cerebral Cortex*, **20** (6), 1332–1340.

Redcay, E., & Courchesne, E. (2005). When is the brain enlarged in autism? A meta-analysis of all brain size reports. *Biological Psychiatry*, **58** (1), 1–9.

Scherf, K., Behrmann, M., Humphreys, K., & Luna, B. (2007). Visual category-selectivity for faces, places and objects emerges along different developmental trajectories. *Developmental Science*, **10** (4), F15–F31.

Scherf, K., Luna, B., Minshew, N., & Behrmann, M. (2010). Location, location, location: alterations in the functional topography of face- but not object- or place-related cortex in adolescents with autism. *Frontiers in Human Neuroscience*, **4**, 26.

Schultz, R.T. (2005). Developmental deficits in social perception in autism: the role of the amygdala and fusiform face area. *International Journal of Developmental Neuroscience*, **23**, 125–141.

Schultz, R.T., Gauthier, I., Klin, A., Fulbright, R.K., Anderson, A.W. *et al.* (2000). Abnormal ventral temporal cortical activity during face discrimination among individuals with autism and Asperger syndrome. *Archives in General Psychiatry*, **57** (4), 331–340.

Stoeckel, L.E., Palley, L.S., Gollub, R.L., Niemi, S.M., & Evins, A.E. (2014). Patterns of brain activation when mothers view their own child and dog: an fMRI study. *PloS ONE*, **9** (10), e107205.

Thomaz, C.E., & Giraldi, G.A. (2010). A new ranking method for principal components analysis and its application to face image analysis. *Image and Vision Computing*, **28** (6), 902–913.

Toal, F., Bloemen, O.J., Deeley, Q., Tunstall, N., Daly, E.M. *et al.* (2009). Psychosis and autism: magnetic resonance imaging study of brain anatomy. *British Journal of Psychiatry: The Journal of Mental Science*, **194** (5), 418–425.

Tong, F., Nakayama, K., Moscovitch, M., Weinrib, O., & Kanwisher, N. (2000). Response properties of the human fusiform face area. *Cognitive Neuropsychology*, **17** (1), 257–280.

Tottenham, N., Tanaka, J.W., Leon, A.C., McCarry, T., Nurse, M. *et al.* (2009). The Nimstim set of facial expressions: judgments from untrained research participants. *Psychiatry Research*, **168** (3), 242–249.

Trontel, H.G., Duffield, T.C., Bigler, E.D., Froehlich, A., Prigge, M.B. *et al.* (2013). Fusiform correlates of facial memory in autism. *Behavioral Sciences*, **3** (3), 348–371.

Van Kooten, I., Palmen, S., Von Cappeln, P., Steinbusch, H., & Korr, H. *et al.* (2008). Neurons in the fusiform gyrus are fewer and smaller in autism. *Brain: A Journal of Neurology*, **131** (4), 987–999.

Wechsler, D. (1999). *Wechsler Abbreviated Scale of Intelligence (WASI)*. San Antonio, TX: The Psychological Corporation.

Weiner, K.S., & Grill-Spector, K. (2012). The improbable simplicity of the fusiform face area. *Trends in Cognitive Sciences*, **16** (5), 251–254.

Whalen, P.J., Johnstone, T., Somerville, L.H., Nitschke, J.B., Polis, S. *et al.* (2008). A functional magnetic resonance imaging predictor of treatment response to venlafaxine in generalized anxiety disorder. *Biological Psychiatry*, **63** (9), 858–863.

Yang, J., Bellgowan, P.S., & Martin, A. (2012). Threat, domain-specificity and the human amygdala. *Neuropsychologia*, **50** (11), 2566–2572.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Supplemental Figure 1. Mean (SEM) behavioral accuracy on the A) Cambridge Face Memory Test (CFMT) and B) 1-back memory task performed during scanning by HFA adolescents and TD adolescents.

Supplemental Figure 2. The mean (SEM) structural data for A) fusiform volume corrected by whole brain volume, B) fusiform cortical thickness, and C) whole brain volume for HFA adolescents and TD adolescents.