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Version: Accepted (Refereed)

Citation:

Bedford, R.; Elsabbagh, M.; Gliga, T.; Pickles, A.; Senju, A.; Charman, T.; Johnson, M.H. (2012)
Precursors to social and communication difficulties in infants at-risk for autism: gaze following and attentional engagement –
Journal of Autism and Developmental Disorders (In Press)

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Precursors to Social and Communication Difficulties in Infants At-Risk for Autism: Gaze
Following and Attentional Engagement

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(in press, *Journal of Autism and Developmental Disorders*)

Abstract

Whilst joint attention (JA) impairments in autism have been widely studied, little is known about the early development of gaze following, a precursor to establishing JA. We employed eye-tracking to record gaze following longitudinally in infants with and without a family history of autism spectrum disorder (ASD) at 7 and 13 months. No group difference was found between at-risk and control infants in gaze following behaviour at either age. However, despite following gaze successfully at 13 months, at-risk infants with later emerging socio-communication difficulties (both those with ASD and atypical development) allocated less attention to the congruent object compared to typically developing at-risk siblings and low-risk controls. This is discussed in terms of the subtle emergence of difficulties in JA.

Keywords: autism, at-risk siblings, broader autism phenotype, joint attention, gaze following.

Precursors to Social and Communication Difficulties in Infants At-Risk for Autism: Gaze Following and Attentional Engagement

In typical development sensitivity to another's gaze appears to be present from birth. Neonates show a preference for faces with eyes open (Batki, Baron-Cohen, Wheelwright, Connellan & Ahluwalia, 2000) and fixate for a greater time, with a higher number of orienting responses, to direct as compared to averted gaze faces (Farroni, Csibra, Simion, & Johnson 2002). From immediately after birth, infants can be 'cued' by the direction of an adult's gaze (Farroni, Massaccesi, Pividori, & Johnson, 2004). These authors used an attention cueing paradigm in which they presented a central face, whose eyes blinked to gain the infant's attention, followed by a gaze shift to the right or left. A target stimulus then appeared on the screen in a location congruent or incongruent with the direction of gaze. Infants were quicker to orient (based on saccadic reaction time) to the target when it was congruent with gaze direction. Frischen, Bayliss and Tipper (2007) argued that infants rely on fairly 'low-level' factors, such as the direction of movement of the pupil. However, inversion of the face or removal of preceding direct gaze removes the cueing effect (Farroni, Mansfield, Lai, & Johnson, 2003), suggesting that it is something about object-directed motion within the context of an upright face with preceding direct gaze that is important for gaze following to occur (see also Senju & Csibra, 2008).

Gaze following involves orienting attention towards a stimulus in response to another person's shift in gaze. Not only is such gaze following present early in infancy, but it is also observed in other social primates (Tomasello, Call & Hare, 1998; Deaner & Platt, 2003). There is even evidence that dogs are able to use eye-gaze and head direction cues to locate food, when these cues are not in conflict (Hare, Call & Tomasello, 1998). The preservation of this ability

across species suggests that following gaze may be a fairly low-level process. Butterworth and Jarrett (1991) suggest that in human development, early emerging biases, such as sensitivity to eye-gaze together with gaze following may form a mechanism for the later development of joint attention (JA).

Although there is consensus that JA refers to the ability to engage in *shared attention* with another individual (Baron-Cohen, 1989; Mundy, Sigman, Ungerer, & Sherman, 1986), there is a lack of clarity over precise terminology, with 'joint attention' being used to refer to several different behaviours in the literature. The classic examples of JA behaviours are spontaneous gaze following and 'protodeclarative' pointing (Scaife & Bruner, 1975). These qualify as JA because the child's response brings them into a shared focus of attention with another person. It is not restricted to these since other gestures (e.g., a nod in one direction) can produce the same end-state (the other person turning to look at the same 'topic' picked out by the first person's nod). Baron-Cohen (1994, 1995) argued that JA requires 'triadic representation' in that the infant has to represent that both they and the other person are attending to the same thing.

In this study we draw a distinction between gaze following and joint attention. By our definition JA also implies referential understanding, whereas gaze following, when taken in isolation, does not. In their developmental model, Tomasello, Carpenter, Call, Behne and Moll (2005) argue that children move from the 'understanding of pursuit of goals' stage at 9 months to 'understanding choice of plans' between 12-15 months. They suggest that the key change is a switch from 'joint perception' to 'joint attention', mediated by the development of a capacity to represent others' internal mental representations. Whilst not all researchers take this modular approach, (e.g. Mundy, Sullivan & Mastergeorge, 2009) it certainly seems that there is development, whether continuous or categorical, in infants' understanding of the meaning of eye-gaze across this time period.

The difference between the behavioural indices used to 'measure' gaze following and responding to joint attention (RJA) can be subtle. Unlike gaze following, which is usually

measured simply by correct orienting, RJA often include shifting attention back and forth between the person and the referred object, rather than simple orienting (Carpenter, Nagell, & Tomasello, 1998). Furthermore, looking time to the object is taken as a measure of infants' referential understanding. Brooks and Meltzoff (2005) found that from 10 months infants looked longer at a target object when the adult looked at the object with their eyes open (versus eyes closed). They argue that from at least this age, infants understand the importance of open eyes as a cue to the other person 'seeing' something, and that the adult's looking behaviour causes the object to acquire a new meaning for these infants. In other words it is not just the act of orienting but the subsequent looking behaviour which distinguishes infants who understand the *meaning* of gaze.

Across different studies, impairments in JA behaviours characterise young children with autism (for a review see Elsabbagh & Johnson, 2007). Charman (2003) concludes that the majority of studies based on retrospective parental report show that JA difficulties are likely to be the best 'discriminators' of emerging autism symptoms in infants between 12 and 18 months. For this reason it was a key behaviour (by its absence) in the Checklist of Autism in Toddlers (CHAT) used at 18 months of age (Baron-Cohen et al., 1992, 1996). In order to understand why difficulties in joint attention behaviours emerge early on in children who go on to develop autism, it is necessary to look at how precursors to JA might influence the trajectory of development.

Sensitivity to gaze, gaze following and attentional engagement with the gazed-at object are all theoretically important for the development of JA. Considering such behaviours in the context of development is crucial, as the ability to discriminate eye-gaze direction (Caron, Caron, Roberts & Brooks, 1997) and flexibly shift attention (Hood & Atkinson, 1993) emerge very early in typical development. Given that a diagnosis of autism rarely occurs before two years of age, looking prospectively at gaze following in infants at risk for an ASD is therefore important in understanding the developmental trajectory of this behaviour. The risk for ASD in siblings,

whilst low in absolute terms, is still much higher than in the general population (1%; Baird et al., 2006). Large scale studies have estimated that 5-10 % of later born siblings of children with ASD go on to receive a diagnosis themselves (Bolton et al., 1994; Constantino et al., 2010). Rates within infant sibling studies, however, have been much higher (Ozonoff et al., 2011; Landa, Holman and Garrett-Mayer, 2007; Rogers, 2009). Although many prospective studies have focused on determining risk factors for autism, little evidence has been found for such early behavioural markers in the first year (Rogers, 2009; Yirmiya & Charman, 2010). The prospective design also enables a broader autism phenotype (BAP) approach, looking for early group differences between those with and without a genetic risk for autism.

Elsabbagh et al. (2009) used an event-related potential (ERP) technique to look at brain responses to direct as compared to averted gaze in at-risk and low-risk 10-month-old siblings. They found that although the two groups could not be differentiated in their response to direct versus averted gaze in early latency posterior ERP components, the high-risk group showed a longer latency in response to direct gaze in a later component. Grossmann, Johnson, Farroni and Csibra (2007) suggest that such later components reflect top-down processing of the referential context of mutual gaze. Whilst understanding of communicative intent is not likely to emerge until later in development, such early differences in later components may relate to subsequent JA difficulties in these infants.

Several prospective studies have investigated JA behaviours in at-risk infants. Presmanes, Walden, Stone and Yoder (2007) tested 12-23 month olds using ten JA prompts in different combinations. Trials ranged from single cue (silent gaze shift) to highly redundant (gaze shift with point and vocalisation). At these two extremes, performance of at-risk and control children was similar. However, differences were found on the intermediate cue conditions, with reduced looking to the target by at-risk children. In an extension of the study, Yoder, Stone, Walden and Malesa (2009) used growth curve modelling to examine the relationship between early RJA abilities and later social impairment, measured by observation of RJA at outcome, and ASD

diagnosis. They found that initial level of RJA (assessed at mean age 15 months), but not its growth rate, predicted RJA impairment and ASD diagnosis at outcome (34 months). However, as the first measure is at 15 months we have no way of telling whether such early differences were present from the first few months, or alternatively developed during the first year of life.

Joint attention has also been examined prospectively using the Early Social Communication Scales (ESCS; Mundy, Hogan & Doehring, 1996), a standardised clinical observation of JA behaviours. Cassel et al. (2007) demonstrated reduced RJA on the ESCS in a group of 18-month-old at-risk siblings compared to low-risk children with a typically developing older sibling. However, unlike Yoder et al. (2009), Cassel et al. (2007) found no significant RJA impairment in the at-risk group at the younger age of 15 months, nor earlier at 8, 10 or 12 months. Other studies using the ESCS in at-risk and low-risk infants younger than 18 months have also shown no significant group differences in RJA (e.g. Goldberg et al., 2005; Yirmiya et al., 2006), although Goldberg et al. (2005) did find a non-significant reduction in frequency of RJA at 17 months in the subgroup of at-risk children who went on to develop an ASD, compared to children in the at-risk group who did not develop autism and low-risk controls. Negative findings might be attributable to differences in cue-type, with multiple cues in the ESCS (calling the child's name, pointing and looking at an object) compared to 'intermediate' levels of cues in Presmanes et al.'s (2007) experimental paradigm.

Taken together, the studies of RJA in at-risk infants suggest that impairments emerge, rather than being present from birth. It has previously been suggested that a range of subtle deficits early in development may interact with each other and the environment and become more pronounced over time (Elsabbagh & Johnson, 2010). Thus, studying precursors of joint attention, including gaze following and looking time to the referred object, may elucidate some of the inconsistent findings described earlier. Studying these behaviours in an atypical population will also inform us about their underlying mechanisms, more specifically about whether we are dealing with common or dissociable mechanisms.

The at-risk sibling paradigm lends itself to the study of gaze following behaviours, which emerge in typical development during the first year of life and thus before a diagnosis of autism can be made. In the current study eye-tracking was used to look at gaze following behaviour in an experimental task (see Senju & Csibra, 2008). In Senju and Csibra's (2008) task, 6-month-old typically developing infants viewed short videos of a model turning to look at one of two objects. Difference scores for the gazed-at ('congruent') as compared to non gazed-at ('incongruent') objects, for measures of first look, frequency of shifts between face and object and duration were then calculated. They found that the number of first looks to the congruent object was significantly greater than that to the incongruent object when the model engaged the watching infant in eye contact before shifting their gaze.

In the current study we tested a group of infants at-risk for an ASD and low-risk controls from the British Autism Study of Infant Siblings (BASIS). Infants took part in the study at around 7 months, and again at around 13 months of age. The at-risk infants were split into typically developing (TD)-sibs, atypically developing (AT)-sibs and Autism Spectrum Disorder (ASD)-sibs on the basis of clinical assessment at 36 months. To explore behavioural differences between the at-risk and low-risk groups, gaze following to the congruent object versus incongruent object and subsequent attentional engagement with the congruent object were measured. If at-risk infants, particularly ASD-sibs, have problems in orienting to gaze, they should show impairment in gaze following behaviour. If their difficulty is in the sensitivity to referential communication, they should show intact gaze following but impaired attentional engagement with the congruent object.

Method

Participants

The current study forms part of a battery of studies administered to infants as part of the *British Autism Study of Infant Siblings (BASIS: www.basisnetwork.org.uk)*. One hundred and four infants from BASIS took part in the current study (54 at-risk, and 50 low-risk). Twenty-one of the at-risk infants were male, 33 were female. Twenty-one of the low-risk infants were male, 29 were female. Along with several other measures, the infants were seen for the gaze following task at the Centre for Brain and Cognitive Development when they were 6-10 months of age (mean = 7.3 months, $sd = 1.22$) and 11-18 months of age (mean = 13.8 months, $sd = 1.46$). Subsequently, children were seen for assessment around the second birthday (24.4 months, $sd = 0.89$) and again around their third birthday (mean = 38.4, $sd = 3.01$). At the time of enrolment, none of the infants had been diagnosed with any medical or developmental condition.

At-risk infants all had an older sibling (hereafter, proband) with a community clinical diagnosis of ASD (or in 4 cases, a half-sibling). Forty-five probands were male, 9 were female. Proband diagnosis was confirmed by two expert clinicians (PB, TC) based on information using the Development and Wellbeing Assessment (DAWBA; Goodman, Ford, Richards, Gatward, & Meltzer, 2000) and the parent-report Social Communication Questionnaire (SCQ; Rutter, Bailey & Lord, 2003). Most probands met criteria for ASD on both the DAWBA and SCQ ($n = 44$). While a small number scored below threshold on the SCQ ($n = 4$) no exclusions were made, due to meeting threshold on the DAWBA and expert opinion. For 2 probands, data were only available for either the DAWBA ($n = 1$) or the SCQ ($n = 1$). For 4 probands, neither measure was available (aside from parent-confirmed local clinical ASD diagnosis at intake). Parent-reported family medical histories were examined for significant medical conditions in the proband or extended families members, with no exclusions made on this basis.

Infants in the low-risk group were recruited from a volunteer database at the Centre for Brain and Cognitive Development, Birkbeck. Inclusion criteria lack of any ASD within first-degree

family members (as confirmed through parent interview regarding family medical history). All low-risk infants had at least one older sibling (in 5 cases, only half-siblings). Twenty-eight of the older siblings were male, 22 were female. Screening for possible ASD in these older siblings was undertaken using the SCQ, with no child scoring above instrument cut-off for ASD (≥ 15).

Out of the total sample, only the 73 infants (35 at-risk and 38 low-risk) who completed the gaze following task at both visits (7 and 13 months) were included in the analysis (mean time between visits: low-risk mean = 6.45 months, $sd = 0.98$; at-risk mean = 6.37 months, $sd = 0.91$). Independent samples t-tests showed the groups did not differ significantly on age at either visit: 7 month visit (low-risk mean = 7.4, $sd = 0.11$, range 6-10; at-risk mean = 7.1, $sd = 0.09$, range 6-10), $t(71) = 0.709$, $p = 0.481$; 13 month visit (low-risk mean = 13.8, $sd = 0.10$, range 11-16; at-risk mean = 13.6, $sd = 0.13$, range 11-18), $t(71) = 0.823$, $p = 0.413$.

[insert Table 1 about here]

Behavioural assessment and outcome groups.

Infants were assessed on the *Mullen Scales of Early Learning (MSEL; Mullen, 1995)* at 7, 13, 24 and 36 months (see Table 1). The low-risk control and at-risk groups showed significantly different early learning composite scores at both visits: 7 month visit (low-risk mean = 105.7, $sd = 11.8$; at-risk mean = 91.5, $sd = 13.8$), $t(71) = 4.77$, $p < 0.001$; 13 month visit (low-risk mean = 108.0, $sd = 15.5$; at-risk mean = 97.2, $sd = 14.1$), $t(71) = 3.09$, $p = 0.003$). At the 24 and 36 month visits the *Autism Diagnostic Observation Schedule – Generic (ADOS-G; Lord et al., 2000)* assessment was administered to at-risk children. At 24 months, of the 35 children who took part in the gaze following task at both the 7 and 13 month visits, 2 toddlers completed Module 2 and 32 completed Module 1. One child did not take part in the 24m visit but was still included in group analysis of gaze following data. Control children were not administered the ADOS-G. At 36 months the 35 at-risk (33 completed Module 2 and 2 toddlers completed Module 1) and 38

low-risk control children (all 38 toddlers completed Module 2) were assessed on the ADOS-G. Assessments were administered by trained researchers who had not previously seen the children at the 7 month or 13 month visit, and were thus blind to infant performance on experimental measures. All ADOS-G assessments were double coded and a consensus code was agreed by the researchers. Intra-class correlation coefficients between coders was very high (24 months $icc = 0.73$; 36 months at-risk $icc = 0.76$, low-risk $icc = 0.87$). The ADOS-G algorithm total score combines behaviours from the social and communication domains, with higher scores indicating greater atypicality.

For the at-risk group consensus ICD-10 (World Health Organization, 1993) *ASD diagnoses* (childhood autism; atypical autism, other pervasive developmental disorder (PDD)) were achieved using all available information from all visits by experienced researchers (TC, KH, SC, GP). Given the young age of the children, and in line with the proposed changes to DSM-5, no attempt was made to assign specific sub-categories of PDD/ASD diagnosis. Toddlers from the at-risk group were considered typically developing at 36 months if they (i) did not meet ICD-10 criteria for an ASD; (ii) did not score above the cut-off on the ADOS-G or ADI; (iii) scored within 1.5 SD of the population mean on the *MSEL* Early Learning Composite (ELC) standard score (>77.5) and Receptive Language (RL) and Expressive Language (EL) subscale T scores (>35). Finally, toddlers from the at-risk group were considered to have *other developmental concerns (atypical development)* if they did not fall into either of the above groups. That is, they either scored above the ADOS-G or ADI (Risi et al., 2006) cut-off or scored $<1.5SD$ on the *MSEL* ELC or RL and EL subscales. From the 35 at-risk infants in the current study seen for diagnostic assessment at 36 months, 12 (8 boys, 4 girls) met criteria for an ASD diagnosis (34.3%), 14 (4 boys, 10 girls) were typically developing (40%) and 9 (2 boys, 7 girls) were in the atypical development group (25.7%) with 6 scoring above ADOS cut-off for ASD, 1 scoring above ADOS cut-off for ASD and below Mullen 1.5SD cut-off, 1 above ADI cut-off, and 1 scoring below Mullen 1.5SD cut-off).

Apparatus

Infants' looking behaviour was recorded using a Tobii 1750 eye-tracker. The eye-tracker has an infrared light source and a camera mounted below a 17-inch flat screen monitor to record corneal reflection data. To evaluate where on the screen the infant is looking, the Tobii system used measurements of gaze direction from each eye separately. Stimuli were presented on the screen using ClearView software. Infants sat on their parent's lap 50 centimetres away from the screen. The distance and height of the screen were adjusted for each infant in order to get good tracking of their eyes. Before starting the main experimental task, a five-point calibration sequence was run. The eye-tracking task was started when at least 4 points were marked as correctly calibrated for each eye. Gaze data were recorded at 50 Hz, and the spatial resolution was 1° after calibration.

Stimuli and Procedure

Stimuli used in this study were the same as those used in Senju and Csibra (2008). Example stills from trials presented to the infant are displayed in Figure 1. Each sequence began with two objects on a table and a female model 'looking down' (3 seconds), then looking up – 'direct gaze' (2 seconds) – and then turning her head to look at one of the objects – 'shift' (6 seconds). The 'looking down' phase was measured from the start of the trial until the model looked up and both her head and eye-gaze were directed straight ahead. The 'direct gaze' phase began as soon as the model's eyes were looking ahead, and finished when her head began to turn away. This turning marked the beginning of the 'shift' phase, which finished at the end of the trial. The object looked at by the model during 'shift' is the *congruent* object, and the other, non-gazed at object is the *incongruent* object. Each infant viewed 12 trials, and there were 6 different pairs of objects whose position with respect to the gaze was counterbalanced across trials. Thus in different trials the same object would once be the congruent object and once the incongruent

object. The direction of the female's gaze shift was fixed in the following pseudo-random order: RLLRLRRLRLR. Before the beginning of each trial, the infants' attention was directed to the screen using small animations.

[insert Figure 1 about here]

Data Analysis

For the purpose of trial exclusion, all trials were split temporally into three phases: looking down, direct gaze and shift (see Figure 1). Within each trial, three rectangular areas of interest (AOIs) were defined around the face, congruent object and incongruent object using ClearView software (face subtended 8° by 11.4° and objects by 3.7° by 4.5° for the smallest and 7.3° by 8.4° for the largest). Gaze data was extracted for each of these AOIs as well as a total for the whole slide, using a fixation filter of 60 m/s to exclude random noise unlikely to represent true fixations. Trial exclusion criteria were: (i) no looking to the face during 'direct gaze' as Senju and Csibra (2008) found this to be a prerequisite for gaze following behaviour; and (ii) looking away from the computer screen for the entire 'shift' phase. Only data from the final 'shift' phase was used to calculate the measures of interest: gaze following, defined as a higher proportion of first looks to the congruent than the incongruent object, and attentional engagement, defined as looking time to congruent object for all trials in which the first look was correct.

First look. For analysis of first look responses, infants who completed < 3 valid trials were excluded. The number of valid trials for first look did not differ across groups at either visit (see Table 2; 7 month visit: $F(2, 70) = 1.406, p = 0.252$; 13 month visit: $F(2, 70) = 0.539, p = 0.586$). First look responses were measured from the beginning of the 'shift' phase, and calculated for the congruent and incongruent objects. In this study *percentage first looks to the congruent versus incongruent object* (Moore & Corkum, 1998) was chosen as the primary measure of gaze following behaviour. This measure reflects infants' ability to follow the direction of another

person's gaze to its target. Trials in which the infant did not orient to either object, but was still looking at the screen, were included in when calculating proportions as 'other'. These trials included infants being stuck on the face or orienting to other parts of the screen. These 'other' trials were included in the model, but not explicitly analysed as a dependent variable.

Looking time. Following Brooks and Meltzoff (2005) and Senju and Csibra (2008), we also chose to analyse looking time behaviour, as this is thought to reflect referential understanding as well as being robust against noise arising from any brief loss of tracking. Looking time behaviour was analysed only for trials in which infants were correct in their first look, and a further 4 infants were excluded as they had no correct first looks. Of these excluded infants, 3 were from the at-risk group, and they did not show *MSEL* or *ADOS-G* scores systematically higher or lower than the other infants. There were no significant group differences in total looking time at either visit (see Table 2, 7 month visit: $F(2, 66) = 1.69, p = 0.192$; 13 month visit: $F(2, 66) = 1.148, p = 0.324$). Attentional engagement was defined as looking time to the congruent object (out of total looking time to the slide) during the 'shift' phase, for all first look trials that were correct. This measure reflects not only the infants' ability to follow gaze but also their subsequent engagement with the target of another person's gaze. Looking time to the incongruent object and to 'other' parts of the screen (face, torso, blank sides and table) were included in the model, but not analysed as dependent variables.

[insert Table 2 about here]

Generalised estimating equation (GEE) Analysis. These repeated measures multinomial data were analysed as a set of simple correlated proportions using a generalised estimating equation approach (Pickles, 1998). The GEE approach allows the first look data to be treated as binomial (as responses were either correct or incorrect) and the looking time data as normally distributed. In the first look analyses, for each of the two assessments the number of responses in each

category were analysed as a count, with the total number of each infant's responses at that assessment occasion as a binomial denominator and with a logit link between predictors and the expected proportion. In the looking time analyses, for each of the two assessments the proportion of time in each category was analysed as a Gaussian model with identity link between predictors and the expected proportion. Another advantage of the GEE method comes from the fact that in all cases we used Wald tests to determine the significance of effects, calculated from the sandwich estimator of the parameter covariance matrix. These tests are therefore robust to errors in the assumed correlation between the response proportions and also to the variation in the precision or overdispersion in proportions arising from the varying amounts of measurement on each infant.

Results

For each visit (7 and 13 months), we first compared the control and at-risk groups on experimental measures (first look and looking time) to examine overall group difference based on risk status. The relationship with clinical outcome was then examined, with the at-risk infants split into three groups: 'TD-sibs', 'AT-sibs', 'ASD-sibs'. For all analyses, the *MSEL* composite standard score (from either 7m or 13m visit) was included as a covariate.

7 month visit.

For the 7-month-old infants (see Table 3), a generalised estimating equation showed that both low-risk controls and at-risk infants followed gaze, looking significantly more to the congruent than incongruent object ($z = 3.7$, $p < 0.001$), with no significant difference between groups ($z < 0.001$, $p = 0.98$). Proportion of looking time was then calculated for all *correct* first look trials. No group differences between at-risk and low-risk infants were found for looking to the congruent object ($z = 0.52$, $p = 0.61$) (see Table 3).

[insert Table 3 about here]

At-risk infants were then split into TD-sibs, AT-sibs and ASD-sibs based on clinical outcomes at 36 months. No significant group difference in the proportion of first looks to the congruent versus incongruent object were found ($z = 0.28$, $p = 0.77$). Nor were there any significant group differences in terms of looking time to the congruent object ($\chi^2(3) = 1.1$, $p = 0.78$).

13 month visit.

At 13 months (see Table 4), low-risk controls and at-risk infants both had a significantly higher proportion of first looks to the congruent than incongruent object ($z=8.06$, $p<0.001$), with no group interaction ($z < 0.001$, $p = 0.98$). Nor were there any significant group differences between at-risk and low-risk infants in looking time to the congruent object ($z = 1.57$, $p = 0.12$) (see Table 4).

[insert Table 4 about here]

When split by outcome group, no significant interaction with group was found for first looks to the congruent versus incongruent object ($z = 0.1$, $p = 0.91$). For looking time, a significant overall group interaction was found for looking to the congruent object ($\chi^2(3) = 13.11$ $p = 0.004$; see Figure 2) with significantly reduced looking time in the AT-sibs compared to controls ($z = 2.88$, $p = 0.004$) and TD-sibs ($z = 2.72$, $p = 0.007$), and significantly reduced looking time in the ASD-sibs compared to controls ($z = 2.21$, $p = 0.03$) and TD-sibs ($z = 2.14$, $p = 0.03$). There were no significant differences either between TD-sibs and controls ($z = 0.1$, $p = 0.92$) or between AT-sibs and ASD-sibs ($z = 0.44$, $p = 0.66$).

[insert Figure 2 about here]

Given the finding of reduced looking time to the congruent object in both AT- and ASD-sibs, it is possible that this effect is driven by social communication difficulties, as both groups show high scores on the 24 and 36 month ADOS-G (see Table 1). Within the at-risk infants, a partial correlation accounting for *MSEL* composite score at the 13m visit showed a significant negative correlation between looking time to the congruent object at 13m and continuous 24m ADOS-G score ($r = -0.46$, $p = 0.01$). At 36 months, both controls and at-risk infants were assessed on the ADOS-G. The overall correlation with 13 month looking time to the congruent object shows the same trend ($r = -0.22$, $p = 0.07$), but when split by group, it is the at-risk infants ($r = -0.31$, $p = 0.09$) driving this effect, rather than the low-risk controls ($r = -0.1$, $p = 0.56$).

Discussion

This study aimed to determine whether early problems in spontaneous gaze following and looking behaviour during infancy is part of the broader autism phenotype, or whether such difficulties relate to autism outcome at 3 years. For the controls, if the stringent definition of gaze following is adopted, of a higher proportion of first looks to the congruent than incongruent object (Moore & Corkum, 1998), then like Senju and Csibra (2008) we found that both 7- and 13-month-old infants can follow gaze. Our finding that gaze following was neither influenced by risk status or by later emerging social and communication difficulties measured by the ADOS-G and categorical ASD outcome status within those at-risk, suggests that the early mechanisms for automatic orienting to another's gaze are intact. This is unsurprising in that such orienting is present in other primates (Tomasello, Call & Hare, 1998) whereas JA arguably is uniquely human (Povinelli, Bierschwale, & Cech, 1999; Baron-Cohen, 1995). However, by 13 months, we found reduced looking to the congruent object after gaze following in at-risk infants who go on to an ASD or atypically developing outcome at 3 years. This suggests that having followed gaze direction, these infants may not use this guidance to preferentially attend to the gazed-at object.

Our first hypothesis was that low-risk control infants would show gaze following behaviour, indexed by significantly more first looks to the congruent than the incongruent object, at both visits. The results supported this hypothesis, with controls showing a greater proportion of first looks to the congruent than the incongruent object at both 7 and 13 months. However, we also hypothesised that group differences between controls and at-risk infants would either be present from the 7 month visit and persist over time, or emerge later, at the 13 month visit. Contrary to our hypothesis, no group differences were found, with the at-risk infants also following gaze at both visits. Intact orienting to the congruent versus incongruent object was also found for infants later classified as having ASD.

This finding of gaze following behaviour in at-risk infants, even those who go on to develop ASD, might seem surprising given the evidence that real-life difficulties in responding to another's gaze are one of the earliest discriminators of children who go on to develop autism (Charman, 2003). However, according to a review by Nation and Penny (2008), unlike in 'real-life settings' the majority of published papers find no evidence for deficits in orienting to social stimuli in children already diagnosed with autism when an experimental design is utilised. For example, Swettenham, Condie, Campbell, Milne and Coleman (2003) found that, like typically developing children and adults (e.g. Hood et al., 1998; Driver et al., 1999), children with autism are also faster to orient to objects cued by moving eye gaze. Senju, Tojo, Dairoku, and Hasegawa (2004) demonstrated the existence of this cueing effect in high-functioning autistic children and controls even when it was made explicit that the cue was counterinformative (on 80% of the trials the object appeared in the uncued location). Chawarska, Klin and Volkmar (2003) used both a Posner experimental task and the ADOS-G to examine JA deficits in 2 year old children with autism. They found that whilst deficits in JA were pronounced in the ADOS-G assessment, a cueing effect of eye-gaze was nevertheless observed in the experimental measure. These findings are in line with our results that early automatic orienting to another's gaze is not impaired in the broader autism phenotype in infancy, nor is it a predictor of autism outcome.

For all trials in which first look was correct, a measure of looking time to congruent object was calculated. Attentional engagement with the target of another person's gaze is a measure more associated with referential understanding of the gaze (Brooks & Meltzoff, 2005). No statistically significant group differences between at-risk and control infants were found at either visit. When split by outcome, looking time to the congruent object at 13 months was significantly reduced in the ASD-sibs and AT-sibs compared to controls *and* TD-sibs. Reduced looking to the congruent object at 13 months suggests that whilst these infants who go on to show ASD or atypical development are able to orient correctly in response to the gaze shift, they may not be sensitive to the referential nature of the gaze. This is consistent with evidence showing joint attention difficulties to be among the earliest predictors of autism symptoms (Charman, 2003).

Given the mixed picture of impairment and non-impairment depending on the measure we choose (first look versus looking time) it is useful to understand which of these behaviours is consequential for learning or for developing typical social interactions. As discussed, Brooks and Meltzoff's (2005) study suggests that looking behaviour distinguishes infants who understand the meaning of eye-gaze. This is in line with an ERP study in 9-month-olds, in which infants saw another person looking at an object, either preceded by a period of mutual gaze (joint attention) or not (non-joint attention) (Striano, Reid & Hoehl, 2006). Infants showed an increased amplitude of a neural correlate reflecting attentional engagement when processing an object looked at by another person, as compared to an object in a non-JA situation. Taken together these studies suggest that the gaze of another person can influence subsequent object processing in infants, at both a neural and behavioural level.

Looking time at 13 months distinguishes not only infants who go on to develop ASD, but also those who show atypical development as measured by the ADOS-G, ADI or *MSEL*. Both these groups show high levels of social communication difficulties, as measured by the ADOS-G. We therefore examined the correlation between continuous ADOS-G score and looking time to the

congruent object at 13 months and found a significant relationship within the at-risk infants with the 24m ADOS-G. There was also a non-significant trend with the later 36m ADOS in the at-risk, but not control infants. This suggests that our looking time measure relates to social communication behaviour in at-risk infants, rather than autism outcome per se. The fact that this correlation is weaker with the 36 month ADOS-G is probably due to the increased time between measurement occasions.

The result of no group difference at the early 7 month visit is consistent with findings from other at-risk sibling studies looking for behavioural markers within the first year of life (e.g., Elsabbagh & Johnson, 2010; Rogers, 2009; Yirmiya & Charman, 2010). More specifically to our study, the lack of an outcome group difference in attentional engagement at the 7m visit could be due to the development, over the period 7 – 13 months, of an understanding of the meaning of gaze, and this is impaired in the infants with social communication difficulties. This fits with Tomasello et al.'s (2005) model in which they argue that infants develop from being able to follow the direction of gaze at 6 months to a full understanding of intentionality around 14 months. Alternatively, group differences in looking behaviour may have been present earlier in development but not measured by our task, either because the task was not sufficiently sensitive to detect such differences, or the differences were too subtle to measure behaviourally. It is possible that there were early group differences in neural processing (Elsabbagh et al., 2009) which compounded over time contributed to the emergence of reduced looking time by 13 months.

There is clear evidence that difficulties in responding to joint attention characterise young children with autism. Given the links between joint attention and subsequent socio-communicative development, a key area of impairment in autism, it is plausible that such behaviours play an etiological role in the condition. To understand the developmental pathways leading to diagnosis it is necessary to look at precursors to joint attention, including gaze following behaviour. Our experimental task used eye-tracking to derive measures of gaze

following. Whilst not as ecologically valid as an RJA task in a naturalistic environment, this paradigm can be used in much younger infants. It is also possible to calculate different measures related to gaze following accurately, such as subsequent attentional engagement with the target object. Further, although it was a computer-based task, the stimuli were dynamic video clips of a model turning to look at an object, and thus more ecologically valid than some attention cueing paradigms. Future research should combine this task with ERP methods over a wider developmental time-frame in order to establish whether differences in neural processing precede behavioural differences in looking time responses. While in our study there were no group differences in the number valid trials, it would be interesting in future work to look at whether later gaze following difficulties emerge because children who go on to develop autism reduce their orienting towards faces and therefore miss the referential cues (see Vivanti et al., 2011). One limitation of this study is the fact that researchers conducting the ADOS-G assessments were not blind to group status. However, care was taken to ensure that the team who saw infants for the first two visits, in which the gaze following task was conducted, were not the same researchers carrying out the ADOS-G assessments at 24 and 36 months, and assessments were double coded.

In conclusion, we found that gaze following at 7 and 13 months was not impaired in infants at risk for autism, neither in those who went on to show subsequent social communication difficulties at 24 months nor in infants who were classified as having an ASD at 3 years. However, having followed gaze correctly, infants with later social-communication problems, both those with autism and atypical development showed a reduction in looking time to the congruent object by the 13 month visit. This reduced attention may reflect difficulties in understanding the communicative relevance of eye-gaze and be part of the ongoing developmental process that leads to an ASD presentation and other developmental atypicalities.

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Table 1. Descriptive Statistics on Mullen Early Learning Composite Scores and ADOS-G Scores.

Group	7m Mullen ELC M S.E	13m Mullen ELC M S.E	24m Mullen ELC M S.E	36m Mullen ELC M S.E	24m ADOS-G score M S.E	36m ADOS-G score M S.E
Low-risk controls	106 1.91 N=38	108 2.52 N=38	117 2.53 N=33	116 2.66 N=37		5.05 0.72 N=38
At-risk 'TD-sibs'	91 2.33 N=35	97 2.39 N=35	103 3.37 N=33	105 3.84 N=32	6.88 0.66 N=34	8.80 0.96 N=35
'AT-sibs'	92 2.94 N=14	100 3.15 N=14	110 3.77 N=14	114 3.02 N=13	4.64 0.77 N=14	4.00 0.57 N=14
'ASD-sibs'	90 2.39 N=9	100 3.41 N=9	97 5.04 N=8	100 6.41 N=9	6.33 0.76 N=9	11.33 1.44 N=9
	92 5.78 N=12	92 5.29 N=12	99 7.88 N=11	98 9.73 N=10	10.18 1.15 N=11	12.5 1.62 N=12

Table 2. Number of Valid Trials and Total Duration of Looking at the 7 and 13 Month

Visits by Group and Condition.

Group	First look 7m		Attentional engagement 7m		First look 13m		Attentional engagement 13m	
	M	S.E	M	S.E	M	S.E	M	S.E
Low-risk controls	8.55		3.24		9.47		5.51	0.35
Valid trials	0.41		0.30		0.34		4231	156
Total duration			3792	202			N=37	
	N=38		N=37		N=38			
At risk								
Valid trials	8.60		2.84		9.86		5.44	0.42
Total duration	0.51		0.25		0.32		4386	145
			3243	246			N=32	
	N=35		N=32		N=35			
‘TD-sibs’								
Valid trials	8.29		2.71		9.93		5.43	0.61
Total duration	0.85		0.35		0.37		4239	234
			3216	295			N=14	
	N=14		N=14		N=14			
‘AT-sibs’								
Valid trials	8.56		3.14		9.89		5.14	1.20
Total duration	0.92		0.63		0.59		4582	351
			2967	565			N=7	
	N=9		N=7		N=9			
‘ASD-sibs’								
Valid trials	9.00		2.82		9.75		5.64	0.62
Total duration	0.93		0.42		0.72		4448	218
			3452	519			N=11	
	N=12		N=11		N=12			

Table 3. Proportion of First Looks to and Attentional Engagement with the Congruent and Incongruent Objects at the 7 month Visit.

Group	First look		Attentional engagement	
	Congruent M S.E	Incongruent M S.E	Congruent M S.E	Incongruent M S.E
Low-risk controls	0.40 0.04 N= 38	0.28 0.03 N= 38	0.28 0.03 N= 37	0.03 0.01 N= 37
At risk				
‘TD-sibs’	0.34 0.03 N=35	0.23 0.03 N=35	0.31 0.04 N=32	0.04 0.01 N=32
‘AT-sibs’	0.31 0.04 N= 14	0.24 0.05 N= 14	0.31 0.06 N= 14	0.03 0.01 N= 14
‘ASD-sibs’	0.36 0.04 N= 9	0.25 0.07 N= 9	0.27 0.05 N= 7	0.04 0.03 N= 7
	0.37 0.05 N= 12	0.20 0.05 N= 12	0.34 0.08 N= 11	0.05 0.03 N= 11

Table 4. Proportion of First Looks to and Attentional Engagement with the Congruent and Incongruent Objects at the 7 month Visit.

Group	First look		Attentional engagement	
	Congruent M S.E	Incongruent M S.E	Congruent M S.E	Incongruent M S.E
Low-risk controls	0.57 0.03 N= 38	0.28 0.02 N= 38	0.31 0.02 N= 37	0.08 0.01 N= 37
At risk				
‘TD-sibs’	0.51 0.04 N=35	0.25 0.03 N=35	0.26 0.02 N=32	0.06 0.01 N=32
‘AT-sibs’	0.54 0.05 N= 14	0.26 0.05 N= 14	0.30 0.03 N= 14	0.08 0.02 N= 14
‘ASD-sibs’	0.46 0.10 N= 9	0.21 0.05 N= 9	0.21 0.02 N= 7	0.03 0.01 N= 7
	0.51 0.07 N= 12	0.26 0.05 N= 12	0.22 0.03 N= 11	0.06 0.02 N= 11

Figure Caption

Figure 1. Screen shots of the videos presented to infants, split into the 3 phases used in analysis: looking down, direct gaze, shift; and with the congruent and incongruent object areas of interest (AOIs) highlighted. The visual angle of the overall screen took up 37.6° horizontally and 30.5° vertically. Depending on their size, the visual angle of the objects, subtended 3.7° by 4.5° for the smallest and 7.3° by 8.4° for the largest.

Figure 2. Attentional Engagement: Proportion of Looking Time to the Congruent Object at the 13 Month Visit.

Figure 1

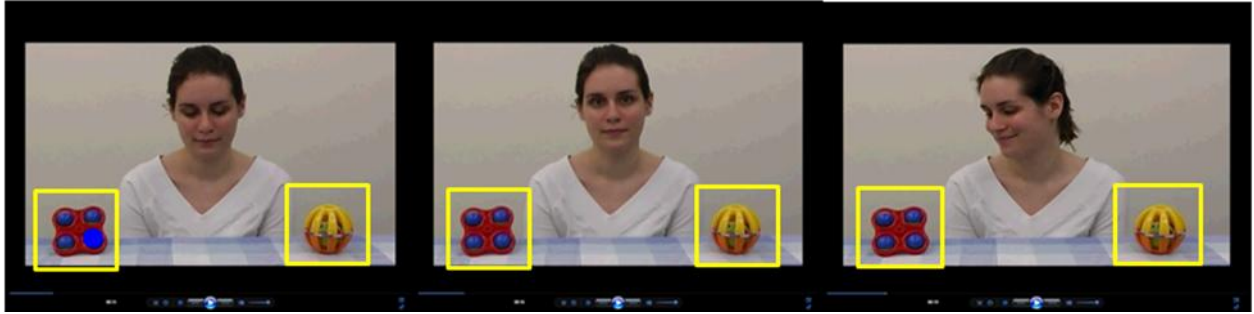


Figure 2

