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Running head: SPONTANEOUS BELIEF ATTRIBUTION IN SIBLINGS AT-RISK
FOR AUTISM

Spontaneous Belief Attribution in Younger Siblings of Children on the Autism
Spectrum

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Abstract

The recent development in the measurements of spontaneous mental state understanding, employing eye-movements instead of verbal responses, has opened new opportunities for understanding the developmental origin of “mind-reading” impairments frequently described in ASD. Our main aim was to characterize the relationship between mental state understanding and the broader autism phenotype, early in childhood. An eye-tracker was used to capture anticipatory looking as a measure of false beliefs attribution in 3 year-old-children with a family history of autism (At-risk participants, $n = 47$) and controls (Control participants, $n = 39$). Unlike controls, the at-risk group, independent of their clinical outcome (ASD, broader autism phenotype or typically developing), performed at chance. Performance was not related to children’s verbal or general IQ and nor was it explained by children “missing out” on crucial information, as shown by an analysis of visual scanning during the task. We conclude that difficulties with using mental state understanding for action prediction may be an endophenotype of autism spectrum disorders.

Keywords: autism, family risk, mental state understanding, false belief, eye-tracking, social communication abilities

Spontaneous Belief Attribution in Younger Siblings of Children on the Autism Spectrum

It has been proposed that the social and communication impairments that characterise autism spectrum disorders (ASD) stem from delays and/or difficulties with understanding that human actions are a result of mental states (e.g. desires, knowledge and beliefs) which may not always conform to reality (Baron-Cohen, 2005, but see Gernsbacher & Frymiare, 2005). Although “Theory of Mind” studies often show participants with ASD at a disadvantage, compared to control participants, exceptions do exist, which make it difficult to describe mindreading difficulties as a *core* deficit in ASD. For example, tasks that assess false belief understanding (e.g. the Sally-Anne task, the Smarties task) have repeatedly found that children with ASD fail at an age where typically developing children had long overcome any difficulties (Baron-Cohen, Leslie, & Frith, 1985; Leslie & Thaiss, 1992) but also that performance is dependent on linguistic abilities (Milligan, Astington, & Dack, 2007). Older children with ASD, and/or those with more advanced language, easily pass these tests (Bowler, 1992; Happe, 1995; Steele, Joseph, & Tager-Flusberg, 2003). This, and the fact that difficulties with false belief understanding can also result from language difficulties such as specific language impairment (SLI) (Farrar et al., 2009; van Buijsen, Hendriks, Ketelaars, & Verhoeven, 2011) suggested that reasoning about mental states may be difficult only for less linguistically able individuals, therefore not specific nor universal for ASD.

However, a non-linguistic false belief task, designed to test this ability in pre-verbal children (Southgate, Senju, & Csibra, 2007) has recently revealed difficulties with using mental states for action prediction, even in verbally able adults with autism (Senju, Southgate, White, & Frith, 2009). In this task an eye-tracker is used to monitor

gaze behaviour while participants watch a videoclip depicting a false belief attribution scenario similar in structure to the Sally-Anne task. Instead of asking participants to verbally report where someone would look for an object displaced without that person knowing, eye-tracking is used to measure whether the participant looks in anticipation towards the location where the person is expected to search. Reality biases in responses (i.e. not being able to inhibit looking to the true location of the object instead of the location where the person would search) were prevented by having the object removed from the scene instead of just changing its location. These modifications allowed children as young as 2 years of age to succeed in this task and thus show evidence for mental state understanding (Southgate et al., 2007). In contrast to the good performance of typically developing toddlers, adults with Asperger syndrome and older children with autism perform at chance (Senju et al., 2009). Measures of anticipatory looking reflect spontaneous, on-line computation of others' mental states, which may be distinct from and developing in parallel with the ability to reason about behaviours in terms of mental states when explicitly asked to do so. Predicting other people's actions based on their mental states as important for functioning in a social world as being able to reason and communicate about these mental states. Taking part in joint activities is just one example in which action prediction is believed to be instrumental (Sebanz & Knoblich, 2009). Unlike performance in classical Sally-Ann tasks, anticipatory looking is unrelated to language skills (Ruffman, Garnham, & Rideout, 2001; Senju et al., 2010) and is possibly present in typically developing infants as young as 7 months (Kovacs, Teglas, & Endress, 2011).

The reliance on language skills to succeed in the standard theory of mind task has clouded our understanding of the relationship between mindreading difficulties and symptomatology of autism in terms of social and communication abilities. One strategy

employed to investigate this relationship is to take advantage of the wider range of social abilities manifested by relatives of individuals with ASD. For example, Losh and Piven (2007) showed poor sociability in relatives was reflected in poor performance on the ‘Reading the Mind in the Eyes Test’, a test which requires inferring emotional mental states from only the eyes region of faces (Baron-Cohen and Hammer, 1997; Dorris et al, 2004). In contrast, non-affected siblings of children with autism performed as well as controls, in standard false belief tasks (Shaked, Gamliel, & Yirmiya, 2006). In this study though, good performance could be explained by the good verbal and non-verbal skills siblings had, which may have allowed them to infer mental states off-line, when asked explicitly. However, we know that even those individuals with ASD capable of reasoning about a false belief situation like Sally-Anne may still have difficulties with using this information in real time (Senju et al., 2009).

It is the above considerations that motivated the current study in which action anticipation based on mental states understanding was measured in 3-year-old siblings of children with ASD, using the same paradigm as in Senju et al (2009). Younger siblings manifest a wide variety of clinical and sub-clinical ASD-like traits. Recurrence rates in these populations vary, but in the largest study to date (N = 664 at-risk siblings) around 20% of at-risk participants developed ASD when assessed around three years of age (Ozonoff et al., 2011). Moreover, other siblings at high familial risk for autism, despite not reaching the clinical threshold for ASD, can subsequently manifest a wide range of social and communicative difficulties – considered to be manifestations of the broader autism phenotype (Rogers, 2009; Yirmiya & Charman, 2010). Showing difficulties in this task not only in those children with ASD but also in those on the broader autism phenotype will reinforce the hypothesis of a common origin for the social skills and the “mindreading” difficulties characteristic of ASD (Baron-Cohen,

Leslie, & Frith, 1985).

Failure in mental state understanding tasks must be interpreted cautiously as difficulties with aspects of the tasks other than mental state understanding can result in poor performance. A lack of a bias or motivation to attend to socially relevant information as well as attention disengagement difficulties have both been proposed as alternative explanations for the apparent “mind-blindness” of people with autism. It has been suggested that people with ASD are not motivated to infer other people’s mental states and intentions (Andari et al., 2011; Dawson et al., 2004; Liebal, Colombi, Rogers, Warneken, & Tomasello, 2008), which could make them “miss out on” information needed to succeed in theory of mind tasks. Equally, failure could result from difficulties with disengaging from irrelevant aspects of the scene (Elsabbagh et al., 2009; Landry & Bryson, 2004) in order to notice when the actor attends or does not attend to the displacement of the object. Toddlers who eventually received a diagnosis of autism were also shown to explore objects in atypical ways, by placing them in their peripheral vision (Ozonoff et al, 2008). Because we used an eye-tracker to monitor gaze we were able to quantify differences in looking behavior during the task and thus address the above concerns. Senju, Southgate et al (2009) did not find a relationship between performance and gaze distribution in older children with ASD. Other studies on this population have found a dissociation between looking at and processing the looked-at information. For example although children at-risk for ASD had no difficulties following someone’s gaze, they did not succeed in learning the name of the gazed-at object (Gliga, Elsabbagh, Hudry, Charman, & Johnson, 2012). We therefore expect no relationship between visual attention distribution and performance in the false belief task. As in the above-mentioned word learning study we expect all children, including poor performers, to attend to key events (e.g. to look at the actress when she turns away

and cannot see an object being moved).

This study aims to characterize the relationship between mental state understanding and clinical and subclinical ASD profiles, in 3 year olds with a family history of this disorder. For the first time with this population, we use eye-gaze as a measure of using mental state understanding for action prediction, a measure not confounded by children's poor verbal skills. The nature of the clinical outcomes within the at-risk group offers the unique opportunity to test whether difficulties with mental state understanding are characteristic of children with a diagnosis of ASD only or of children with poor social and communicative abilities in general (the BAP). Having access to detailed gaze behaviour during the task, a further aim of this study is to show that poor performance is not due to poor or atypical visual attention.

Method

Participants

Participants took part in a longitudinal study of children at risk for autism. Recruitment, ethical approval (London Research Ethics Committee, ref: 09/H0718/14) and informed consent, as well as background data on participating families, were made available for the current study through BASIS, a UK collaborative network facilitating research with infants at-risk for autism. Families enroll when their babies are younger than 5 months of age and they are invited to attend multiple research visits until their children reach three years of age or beyond. Measures collected are anonymised and shared among scientists to maximize collaborative value and to minimize burden on the families. A clinical advisory team of senior consultants works closely together with the research team/s and, if necessary, with the families' local health services, to ensure that any concerns about the child, arising during the study are adequately addressed. At the

time of enrollment, none of the infants had been diagnosed with any medical or developmental condition. Of the initial 50 Control and 54 At-risk participants, 39 *Control* and 47 *At-risk* contributed data to this study. Two Control and one At-risk did not take part in the 36 months visit. Nine other Control were excluded because data was not collected due to technical problems (4) or because of having accumulated less than 20% looking data (5). Six At-risk participants were not included because of home visits (2), having accumulated less than 20% looking data (3) or being more than one year older than the group average at this visit (1). Participants' characteristics (age, gender, IQ) are present in Table 1.

At-risk infants had an older sibling (hereafter, proband) with a community clinical diagnosis of ASD (in 3 cases, a half-sibling), and in one case 2 probands with an ASD. Thirty-eight probands were male, 9 were female. Proband diagnosis was confirmed by two expert clinicians (PB, TC) based on information using the Development and Well Being Assessment (DAWBA, Goodman, Ford, & Richards, 2000) and the parent-report Social Communication Questionnaire (SCQ, Rutter, Bailey, & Lord, 2003). The DAWBA is a parent-completed web-based assessment that asks parents to rate symptoms of autism, relevant to making DSM-IV-TR and ICD-10 diagnosis of autism spectrum disorders. Descriptive information about the child is also included. The experts review the forms using both the scores and the narrative text to assign a diagnosis. The SCQ is a widely-used 40-item questionnaire that ask about current and past autism symptoms. Most probands met criteria for ASD on both the DAWBA and SCQ ($n = 42$). 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 one proband, data were only available for the DAWBA. Parent-reported family medical histories were examined for significant medical conditions in the

proband or extended family members, with no exclusions made on this basis. Infants in the Control group were recruited from a volunteer database. Inclusion criteria included full-term birth, normal birth weight, and lack of any ASD within first-degree family members (as confirmed through parent interview regarding family medical history). All Control infants had at least one older-sibling (in 3 cases, only half-sibling/s). Screening for possible ASD in these older siblings was undertaken using the SCQ, with no child scoring above instrument cut-off for ASD (≥ 15). 62% of controls and 85% of at-risk participants were only exposed to one language.

----- insert Table 1 here -----

Stimuli

The stimulus was a video recording, which depicted five main events: two familiarization trials, two true belief trials (TB) and one final false belief (FB) trial (see Figure 1). We familiarized children with two events in which an actress reached through two doors for a toy strawberry placed on the left (first trial) or the right of two boxes (second trial). An audio-visual cue (the windows were illuminated and a chime sounded) was given and 2.5 seconds later the actor reached through the window and grasped the strawberry. The actor wore a visor so that her gaze direction could not betray the direction of her reach through the windows. The purpose of the familiarization trials was (a) to show the children that the actor's goal was to reach for the object and (b) to teach the children that when the audio-visual cue was presented one of the windows was about to open. At the beginning of the two TB trials, a puppet monkey appeared and placed a banana in the left box (first TB trial) or the right box (second TB trial). After leaving the scene and 2.5 seconds after the cue appeared the

actress reached through the door behind the box that contained the banana. The FB trial is depicted in Figure 1. Crucially, in this trial, the actor turned away from the scene and the puppet monkey returned to remove the banana from the right side box, which induced a false belief in the actor. After the cue was given in this trial the scene froze for another 5 seconds. Because we could not counterbalance the locations of the banana in the FB trial within each outcome group (the outcome was not known at the time when the study was carried out), the same video-clip was used for all participants.

----- insert Figure 1 here -----

Procedure

An integrated Tobii (Stockholm, Sweden) T120 17'' Eye Tracker was used to collect data on direction of gaze. Data were collected at 60 Hz. Tobii Studio was used to present the stimuli and for data analysis. Children sat on their own on a chair, at approximately 60 cm from the Tobii monitor. At this distance the diagonal of the screen subtended app. 40°. A five-point calibration was run before stimulus presentations. Children were told that they would see a movie about a cheeky monkey. An experimenter stood behind the child and encouraged her to look if she got distracted.

Data reduction and analysis

The 2 minutes 45 seconds long video was segmented into scenes of various lengths corresponding to the various important events. To measure anticipatory eye-movements in the second true belief trial we defined a 2.5 second interval after the visual cue appeared, which corresponded roughly to the time taken for the person to reach through the doors in both the familiarization trials and the TB trials. Because the

actor never reached through the door in the false belief trial, a 5 second interval (until the end of the movie) was used for analysis. For clarity, details about the length of other intervals analysed are given together with the results of those particular analyses, in the Results section. Three areas of interest (AOI) were defined manually for all scenes analysed (see Figure 1), two covering the left and right doors and boxes and another one corresponding to the face. Cumulative looking time within areas of interest was calculated automatically using Tobii Studio software. Only fixations longer than 100 ms. were included in the analyses. Data loss could occur during the video presentation at different time points (either due to looking away or to the eye-tracker not detecting the eyes despite the fact that the child was looking). We decided to only exclude children if they accumulated less than 20% data overall and not if only certain intervals had valid data, the consequence of which was that slightly different numbers of participants were entered in the analysis of different events (e.g. in the TB and the FB trial analysis).

Outcome characterization of the At-risk and Control groups

Standard measures of cognitive development – Mullen Scales for Early Learning (MSEL; Mullen, 1995) and adaptive development - Vineland Adaptive Behavior Scale (VABS; Sparrow, et al., 2005) were collected. The MSEL is a standardized direct developmental assessment that yields a standardized score (mean = 100, SD = 15) of overall intellectual ability (Early Learning Composite, and subscale T-scores (mean = 50, SD = 10) for receptive language (RL) and expressive language (EL), as well as non-verbal fine motor (FM) and visual reasoning (VR) abilities. The VABS is a standardized parent-reported interview of everyday adaptive functioning that measures social, communication, daily living and motor skills. In addition (and for both groups) a semi-

structured play-based assessment, the Autism Diagnostic Observation Schedule - Generic (ADOS-G; Lord et al., 2000) was used to assess autism-related social and communication behavioural characteristics (44 children were administered Module 2 and the other 3 children Module 1 of the ADOS-G). This was augmented (At-risk group only) with the parent-report Autism Diagnostic Interview – Revised (ADI-R; Lord et al., 1994). In common with other research groups studying familial at-risk siblings (Zwaigenbaum et al, 2007) a ‘best estimate clinical consensus’ approach to diagnosis was taken following review by experienced clinical researchers (TC, KH, SC, GP) taking account of all information about the child (i.e. MSEL, VABS, informal observation), in addition to information from the ADI-R and ADOS-G. Children were included in the At-risk ASD group if they met ICD-10 (World Health Organisation, 1993) criteria for ASD. 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. Children from the At-risk group were considered typically developing (At-risk Typical) if they (i) did not meet ICD-10 criteria for an ASD; (ii) did not score above the ASD cut-off on the ADOS or ADI; (iii) scored within 1.5 SD of the population mean on the Mullen Early Learning Composite (ELC) score (>77.5) and Receptive Language (RL) and Expressive Language (EL) subscale T scores (>35). Children from the At-risk group were considered to have atypical development if they did not fall into either of the above groups. That is, they either scored above the ADOS or ADI cut-off for ASD or scored $<1.5SD$ on the Mullen ELC or RL and EL but did not meet ICD-10 criteria for an ASD. From the 47 At-risk participants taking part in this task, 17 met criteria for an ASD diagnosis, 18 were At-risk Typical and 12 were in the At-risk Atypical group (9 scoring above ADOS ASD cut-off, 1 scoring above ADOS

ASD cut-off and $<1.5SD$ Mullen ELC cut-off, 1 scoring above ADI ASD cut-off and 1 scoring $<1.5SD$ Mullen ELC cut-off).

Results

We analysed separately the true belief (TB) and the false belief (FB) trials. As in previous studies (Southgate et al., 2007) only the second TB trial was analysed. By not including the first TB trial we thus gave children more opportunities to understand the actor's goal - to reach for the objects - as well as the role of the audio-visual cue. For each trial we assess performance by analysing the difference between the looking time to the correct and incorrect doors' AOIs, scaled by the amount of looking to those AOIs: $(LT_{Correct} - LT_{Incorrect}) / (LT_{Correct} + LT_{Incorrect})$. Values go from -1 (exclusive looking towards the Incorrect location) to 1 (exclusive looking to the Correct location), with chance level at zero. In the TB trial, the correct location was that which contained the banana. In the FB trial, the correct location was that in which the actor thought the banana was. We start the analysis by comparing the Control and At-risk groups to chance levels and to each other and then compare the three outcome groups within the At-risk participants (At-risk ASD, At-risk Atypical and At-risk TD) to chance levels and to each other (using Bonferroni correction for multiple comparisons). Where a significant difference between Controls and At-risk is found, we also test whether all at-risk groups are significantly different than Controls (using Dunnett correction for multiple comparisons). We also test whether any individual variables that showed groups differences, like Total IQ, Verbal IQ or Age (see Table 1) explain group differences in TB or FB performance. Finally, we examine whether visual attention distribution during the false belief trial may account for children's performance. Three

AOIs were entered in this analysis, the door AOIs and another AOI corresponding to the actor (Figure 1).

True belief

Looking time differential scores were significantly above chance (zero) for both Control and At-risk participants (Control: $t(37) = 3.01$, $p = .005$; At-risk: $t(40) = 2.99$, $p = .005$). A univariate ANOVA with Group (At-risk vs. Control) as between-subjects factor yielded a non-significant effect of Group ($F(1,78) = 0.13$, $p = .716$, $\eta^2 = .002$). Verbal and General IQ significantly predicted performance (Verbal IQ: $F(1,76) = 1.78$, $p = .03$, $\eta^2 = .06$; General IQ: $F(1,76) = 1.78$, $p = .02$, $\eta^2 = .06$), however entering these factors in the above ANOVA did not change the significance level for the main Group factor. When analysing the behaviour of the 3 at-risk groups separately, they only performed marginally better than chance (At-risk Typical: $t(15) = 1.24$, $p = .23$; At-risk Atypical: $t(10) = 2.01$, $p = .07$; At-risk ASD: $t(13) = 2.03$, $p = .06$), possibly also because of the reduced power of this analysis. A univariate ANOVA comparing the three at-risk subgroups (At-risk ASD, At-risk Atypical and At-risk Typical) yielded no significant effect of Group ($F(2,40) = .19$, $p = .82$, $\eta^2 = .01$).

False belief

At the point at which anticipatory looking is measured in the False Belief trial the two boxes were empty, thus preventing a reality bias. Correct anticipation is reflected in longer looking towards the box that contained the banana just before the person looked away. As seen in Figure 2, looking time differential scores were higher for the Control group than for the High-risk groups. Preliminary analyses confirmed that Total IQ, Verbal IQ or Age did not have a main effect on looking time distribution

nor did they interact with the factor Group. We therefore removed these factors from further analyses. There was also no group difference in the overall amount of time spent looking at the three target AOIs (correct, incorrect and face) after the light prompt, in the FB trial ($M_{Control} = 3.35$, $SD_{Control} = 1.4$ sec; $M_{At-risk} = 3.56$, $SD_{At-risk} = 1.15$ sec; $t(82) = -.71$, $p = .47$). Mean looking time difference scores were significantly above chance only for the Control participants (Control: $t(35) = 5.13$, $p < 0.001$; At-risk: $t(42) = .86$, $p = .39$). A univariate ANOVA with Group (Control and At-risk) as between-subject variable resulted in a significant main effect of Group ($F(1,78) = 9.35$, $p = .003$, $\eta^2 = .10$). The significance level of the Group factor did not change when the TB looking time performance was entered as a covariate and TB performance did not have a significant impact on FB performance. When the 3 At-risk sub-groups (At-risk ASD, At-risk Atypical and At-risk Typical) performance was analysed separately, none of the groups performed different than chance (At-risk Typical: $t(16) = 1.21$, $p = .23$; At-risk Atypical: $t(9) = .36$, $p = .72$; At-risk ASD: $t(15) = -.36$, $p = .71$). A univariate ANOVA comparing the looking time difference scores for the 3 At-risk sub-groups yielded a non-significant effect of Group ($F(2,42) = 0.79$, $p = 0.46$, $\eta^2 = 0.03$). Post-hoc t-tests, were used to compare each at-risk group to the Control participants. Only At-risk ASD significantly differed from Control participants ($p = .009$), At-risk Atypical and At-risk Typical were not significantly different from Control ($p = 0.11$ and $p = 0.33$).

----- insert Figure 2 here-----

Relationship with social and communication abilities (ADOS)

The lack of a difference in performance between the three at-risk groups suggests that difficulties with mental state understanding may be unrelated to ASD symptom severity.

To confirm that performance in this task is only related to the risk status and not to children's social and communication abilities as measured by the ADOS, we split the Control and At-risk groups depending on their ADOS scores into a *Low ADOS* (ADOS < 8; 25 out of 35 Controls and 21 out of 42 At-risk participants) and *High ADOS* group (ADOS ≥ 8). Looking time performance was entered in a univariate ANOVA with Group (Control, At-risk) and ADOS (Low, High ADOS). This analysis yielded a main effect of risk Group ($F(1,76) = 9.41, p = .003, \eta^2 = 0.11$). The ADOS scores did not significantly predict performance ($F(1,76) = .19, p = .66, \eta^2 = 0.003$) and there was no significant interaction between risk Group and ADOS levels ($F(1,76) = 1.52, p = .22, \eta^2 = 0.02$).

Differences in attention to the placement/displacement events

What can explain the poorer performance of the At-risk participants in the FB trial? We were interested in determining whether children's looking behaviour during the task differed in any way that would explain their performance. One possible source of error could arise from not paying attention to the hiding and displacement events during the FB interval, especially the last hiding event before the actress looks away. Visual inspection of looking time distribution along the FB trial suggests that all groups followed closely this event (Figure 3a: "Banana placed in the right box"; "Monkey steals banana from right box"), and that major differences between groups only emerge at the very end, when FB is tested (Figure 3a: "Person turns back"). We looked more specifically at attention distribution during key events. Groups spent equal amounts of time looking at the box during the 8 sec that the monkey took to place the banana ($M_{Control} = 4.9, SD_{Control} = 2.0; M_{At-risk\ Typical} = 5.3, SD_{At-risk\ Typical} = 1.6; M_{At-risk\ Atypical} = 4.9, SD_{At-risk\ Atypical} = 1.7; M_{At-risk\ ASD} = 5.3, SD_{At-risk\ ASD} = 1.6; F(3,84) = .51, p = .67, \eta^2 = .01$).

Groups also spent equal amounts of time looking at the box from which the monkey surreptitiously removed the banana ($M_{Control} = 7.4$, $SD_{Control} = 2.9$; $M_{At-risk Typical} = 6.5$, $SD_{At-risk Typical} = 3.0$; $M_{At-risk Atypical} = 7.9$, $SD_{At-risk Atypical} = 2.3$; $M_{At-risk ASD} = 6.7$, $SD_{At-risk ASD} = 2.7$; $F(3,84) = .91$, $p = .44$, $\eta^2 = .03$). It is also important to have noticed that, when the banana was removed from the box, the person was looking away. Visual inspection of looking time spent on the face during the FB trial does not highlight consistent group differences (Figure 3b) and indeed, when we compared the amount of time spent looking at the person's face while the monkey removed the banana from the box no group difference was found ($M_{Control} = 3.8$, $SD_{Control} = 1.9$; $M_{At-risk Typical} = 3.1$, $SD_{At-risk Typical} = 2.4$; $M_{At-risk Atypical} = 4.0$, $SD_{At-risk Atypical} = 2.7$; $M_{At-risk ASD} = 4.5$, $SD_{At-risk ASD} = 2.6$; $F(3,84) = 1$, $p = .41$, $\eta^2 = .03$). None of these measures correlate with the FB looking time difference score, for either the whole group or the low and at-risk groups separately.

Closer exploration of the data revealed that at the point in the video where the monkey had placed the banana in the right box and left the screen and before the person turned away, children looked up at the person ($M_{Control} = 2.0$, $SD_{Control} = 1.1$; $M_{At-risk Typical} = 2.2$, $SD_{At-risk Typical} = 1.0$; $M_{At-risk Atypical} = 2.7$, $SD_{At-risk Atypical} = 1.3$; $M_{At-risk ASD} = 2.1$, $SD_{At-risk ASD} = 0.9$; $F(3,80) = 1.42$, $p = .24$, $\eta^2 = .05$) and then looked towards the right door and box. Encoding where the person last saw the object or her goal at this point where a TB is still held may be crucial for predicting their behaviour later. We analysed looking time distribution to correct (here where the banana had been placed) and incorrect locations at this time point. Both Low-risk and High-risk participants looked longer at the Correct side (average and SD for Correct vs. Incorrect for Low-risk: 620 ms (105) vs. 370 ms (81) and High-risk: 552 ms (90) vs. 369 ms (69)). A 2x2 ANOVA with Side and Group confirmed that there was a main effect of Side ($F(1,79) =$

5.01, $p = .02$, $\eta^2 = .06$), but no main effect of Group ($F(1,79) = .21$, $p = .64$) and no interaction between Side and Group ($F(1,79) = .11$, $p = .74$), which means that both groups looked longer at the box containing the banana. To investigate whether looking time distribution at this point was related to performance later in the FB trial we calculated difference scores in both cases (Looking time Correct – Looking time Incorrect). These measures were correlated in the whole sample ($r(70) = .27$, $p = .01$), as well as in the Low-risk group ($r(31) = .40$, $p = .02$) but not in the High-risk group ($r(39) = .13$, $p = .41$). A Chow test demonstrated that the slope and intercept of the regression analysis predicting test performance from looking time distribution when the person last saw the object was not significantly different for the high-risk and low-risk participants ($F(1,69) = 2.02$, $p = .15$).

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Discussion

Previous studies of mental state understanding have documented difficulties with on-line computation of mental states in older children with ASD (Senju et al., 2010) as well as in adults with ASD (Senju et al., 2009). Here we provide evidence that this impairment is measurable as early as 3 years of age in children at familial risk for this disorder and that it is not restricted to those children having received a diagnosis of ASD. Control participants, as a group, performed above chance, confirming previous findings at 24 months of age (Southgate et al., 2007) and suggesting this ability is continuously present during development from 2 years of age through adulthood (Senju et al., 2009, 2010). The At-risk participants, included in the study on the basis of having an older sibling with ASD, developed a wide range of social and communication

abilities by three years of age, with some children receiving a diagnosis of ASD and others manifesting other developmental problems, including sub-clinical scores on the ADOS-G, which measures ASD-like social and communication atypicalities.

Based on previous findings of subtle difficulties with inferring mental states in relatives with poor sociability (Losh & Piven, 2007), we hypothesised that all children with poor social and communication abilities, i.e. both At-risk ASD and At-risk Atypical groups, would show difficulties with mental state understanding. Interestingly, all groups of at-risk children found the task difficult, *including those at-risk that developed typically. Moreover, performance was not related to social and communication abilities, as measured by the ADOS.* The performance of the At-risk Typical group was not significantly different from that of the other at-risk groups but was also not different from that of Controls, suggesting that they may have intermediary abilities, with more participants succeeding at the task than in the other at risk groups. Notwithstanding these findings, the At-risk Typical group's performance was not significantly different than chance. Although similar in terms of IQ to Controls, the At-risk Typical group is more similar to the other at-risk participants in terms of both genetic and family background.

The picture of autism emerging from recent genetic studies is of a multi-factorial disorder, in which outcomes are a result not of a small number of deterministic factors but of the combination of a great number of risk and protective factors (Geschwind, 2011). This model is supported by recent findings from prospective studies of infants at-risk (Elsabbagh & Johnson, 2010). Difficulties with mental state understanding could be one of these many risk factors, which impacts on symptom severity only in combination with other concurrent factors. Family environment is expected to mediate shared genetic influences on the outcome phenotype both in terms of mental state understanding and

social and communication abilities. Previous studies have shown that having an older sibling positively impacts on the development of mental state understanding in typically developing children (Ruffman, Perner, Naito, Parkin, & Clements, 1998). Many of our at-risk participants did not have a typically developing older sibling (they only had an older sibling with ASD). This may place them at a disadvantage in mindreading abilities with respect to Control participants, all of whom had a typically developing older sibling. At this point our sample is too small to properly investigate the interaction between genetic and environmental factors, something future studies will have to clarify.

Although we take failure in our task to mean difficulties with computing and using mental states, alternative explanations are possible. The design of the task, in particular the existence of a True Belief condition, allows us to rule out some of them. Above chance performance in the True Belief trial is evidence that both controls and at-risk participants understood the task and were motivated and able to anticipate someone's actions. Although they did not look to the correct location, a majority of at-risk participants (43/47) did look towards one of the two possible locations in response to the audio-visual cue, in the False Belief trial, which again is not compatible with a lack of motivation. Using eye-tracking we could also ask whether performance can be explained by any differences in the looking distribution during the task. It has previously been proposed that attention disengagement difficulties or a lack of a bias to attend to social information can result in missing crucial information necessary to succeed in theory of mind tasks (Dawson et al., 2004). This would be even more problematic in on-line assessments of mental state understanding, than in classical, slower paced tasks. We therefore analysed looking behaviour at various points during the false belief trial and showed that all children looked at the object placement and

displacement actions and also looked at the person when she turned away from the scene. There was thus no difference in the way children with ASD attended to the sequence of actions. More importantly, the amount of looking did not correlate with looking time performance at test, confirming our initial hypothesis that poor performance was not due to poor attention. Similarities in looking behavior in response to various key events (e.g. looking at the person when she turned away or at the monkey when she was engaged with the banana) also speak against any oculo-motor differences between groups (Ozonoff et al, 2008). Of course, looking is necessary but not sufficient for attending to and processing the information fixated. As previously shown, in a word learning task, children at risk-for ASD could follow someone's gaze to an object, but did not learn the word-object association as well as controls (Gliga, Elsabbagh, Hudry, Charman, & Johnson, 2012). Brain imaging studies of face processing have also shown that even when asked to fixate faces, ASD participants activate a less extensive network of brain areas than neurotypical participants (Hadjikhani, Joseph, Snyder, & Tager-Flusberg, 2007). Thus, similarities in scanning of visual scenes in autism may mask learning and processing differences associated with this condition.

Some differences in looking behavior did appear during the false belief trial. When visually inspecting the data we noticed that children made saccades towards the location containing the banana earlier during the FB trial, before the cue was given, at the point where the person still held a true belief and could have reached for the box. At this point groups were again indistinguishable, suggesting that they had again correctly encoded the person's reaching goal. The positive correlation between looking towards the box containing the banana at this point and looking towards this same box later in the trial, when the banana had been removed, suggests that, at least in controls, success in the FB trial depends on how well one encodes the goal of the actress at the moment at which it

still corresponds to a true belief. This is compatible with one current model that explains spontaneous mental state attribution on the basis of co-representations of people and the objects they encounter and act upon (Apperly & Butterfill, 2009; Samson, Apperly, Braithwaite, Andrews, & Bodley Scott, 2010). Thus, when someone repeatedly reaches for a particular object, their goals or beliefs about that object's location are stored in memory together with that person's identity for later re-enactment. However, this correlation only holds in the control group. The lack of a correlation in the at-risk participants could reflect encoding of different information when children look at the box just before the person turns away (e.g. the location of the banana instead of the person's goal) or difficulties with maintaining that information in memory. Future behavioural studies could test the impact of memory by varying the time delay between hiding, displacement and test. Brain imaging studies could measure the nature of the information encoded initially (e.g. object location or action goal). Although at this point we can not tell whether these differences in performance are due to at-risk participants not being able to compute mental states nor whether they are due to not being able to keep in memory someone's representation of the world while monitoring changes in the world itself, we subscribe to a recently made case for the importance of theory of mind tasks to reveal not just conceptual understanding but also functional usage of these abilities (Apperly, 2012). Whether or not they can compute mental states, we show that children with a family background of autism did not use these abilities online to anticipate another's actions, which is of importance knowing that action prediction is believed to be crucial for joint activities (e.g. for example Sebanz & Knoblich, 2009).

The current study advances our understanding of mental state attribution in ASD by providing the earliest evidence that these difficulties are not restricted to those children that fulfill diagnostic criteria for ASD but characterize the whole at-risk group. It is

unclear at this point whether these difficulties are specific to ASD-risk, whether they reflect a genetic susceptibility, the influence of the social environment or maybe the interaction between these two factors. Difficulties with classical theory of mind tasks have been documented in other developmental disorders like Down syndrome or mental retardation (Yirmiya, Solomonica-Levi, Shulman & Pilowsky, 1996). However, in these populations performance was correlated with non-verbal IQ, which suggests general cognitive factors like memory and attention can limit mental state understanding. This was not the case in our study, where general and verbal IQ did not explain group differences, nor did visual attention distribution during the task. A detailed analysis of looking behavior confirmed that failure was not due to “missing out” on important information, such as where the object had been placed/displaced or whether the person was attending to the scene or not. Therefore, success on this task does not appear to be due to where someone looks for information, but to how or whether the ‘looked at’ information is later used.

The mechanisms of mental state understanding have been subject to heated debates both within autism research and within developmental psychology. We believe that both fields will benefit from the study of younger siblings of children with autism. The greater variability of social, communicative and attentional abilities this group manifests will make it possible to identify necessary conditions for the development and online use of mental state understanding. For example, while succeeding in standard false belief task depends on language skills, this is not the case for our task. Being able to encode and maintain in memory action goals while events unfold seems to be the limiting factor when mental states are used for action anticipation. Moreover, the wider variety of clinical outcomes in this population will help settle debates about the specificity of mindreading difficulties to autism spectrum disorders.

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Table 1
Participants' Characteristics

GROUPS	Control (n = 39)	At-risk (n = 47)	At-risk Typical (n = 18)	At-risk Atypical (n = 12)	At-risk ASD (n = 17)
Age (months)					
M (SD)	39.5 (3.3)	38.0 (1.7)*	37.9 (1.5)	37.5 (1.7)	38.6 (2.0)
Range	36-52	33-42	34-40	33-40	35-42
Gender					
Male	21	27	12	9	6
Female	18	20	6	3	11
General IQ (Mullen) ^a					
M (SD)	114.7 (16.1)	104.2 (22.5)*	113.2 (15.0)	103.4 (18.9)	94.7 28.5 ++
Range	72-137	49-147	86-142	63-126	49-147
Verbal IQ (Mullen) ^b					
M (SD)	57.5 (8.5)	51.2 (12.0)*	55.9 (14.5)	49.8 (8.6)	47.2 (15.9)
Range	41-69	29-73	43-69	29-64	20-73
ADOS SC ^c					
M (SD)	5.5 (4.5)	8.8 (5.3)**	4.1 (2.0)	12.0 (4.0)++	11.4 (5.0)++
Range	0-21	0-19	0-7	5-18	1-19
ADI Social					
M (SD)	-	5.0 (5.4)	1.9 (1.7)	3.4 (4.9)	9.7 (5.5) ++
Range	-		0-5	0-18	1-19
ADI Communication					
M (SD)	-	4.5 (4.9)	2.2 (1.8)	3.5 (5.4)	8.2 (5.2)++
Range	-		0-6	0-20	1-16
Ethnicity					
Caucasian	33	41	17	9	14
Caucasian/Asian	2	2		1	1
Caucasian/Latino		1		1	1
Caucasian/Black	1	1			1
Asian	1				
Black	1	1		1	
Mixed	1	1	1		
Income ^d					
< 40 k	9	17	4	8	5
40-80 k	14	23	8	3	12
> 80 k	16	7	6	1	0
Education ^e					
M (SD)	3.0 (0.9)	2.7 (0.9)	2.8 (0.9)	3.0 (0.9)	2.4 (0.9)
Range	1-4	1-4	1-4	1-4	1-4

Note. Superscripts indicate differences between low and high risk (* $p < 0.05$; ** $p < 0.01$) and between the At-risk ASD or Atypical groups and at-risk TD (+ $p < 0.05$; ++ $p < 0.01$; Bonferroni correction). ^aMullen ELC score, Mean = 100, SD = 15; ^bVerbal ability T-score, Mean = 50, SD = 10 (based on average of Expressive and Receptive Language domains); ^cThe Social and Communication algorithm score of the ADOS; ^dOverall household income; ^eMother's education level (1= formal education to 16; 2= formal education to 18; 3= university degree or equivalent; 4 = postgraduate); Missing data for Education level (7 data points) were replaced by the average of the risk group.

Table 2
Looking Time Differential Scores in the True Belief and False Belief Trials

		Control	At-risk	At-risk Typical	At-risk Atypical	At-risk ASD
True Belief	M	.30*	.25*	.19	.32	.28
	SD	.62	.55	.61	.52	.52
	N	38	41	16	11	14
False Belief	M	.42*	.07	.18	.04	-.04
	SD	.49	.53	.64	.40	.46
	N	36	43	17	10	16

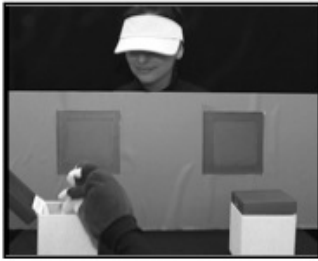
Note. The asterisk indicates significance of one-sample t-tests ($p < 0.05$) against a chance level of zero.

Figure Captions

Figure 1. Key events during the false belief trial. The last frame depicts the three areas of interest used for data extraction.

Figure 2. Looking time differential scores in the False Belief trial. The chance level is at zero.

Figure 3. a) Proportion of children looking at the face AOI during the FB trial. b) Proportion of children looking at the correct AOI (the right box) during the FB trial to illustrate all groups keeping track of where the banana was placed. At-risk ASD and At-risk Atypical data was pooled together for clarity (continuous grey line). At-risk Typical (dashed grey line) and Controls (black line). The onset of important events is indicated on the time line.



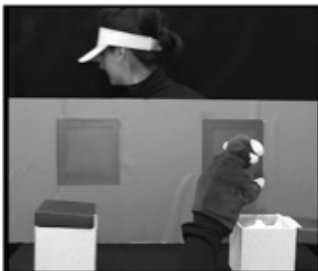
Monkey places
the banana in
the left box.



Monkey places
the banana in
the right box.



Monkey leaves.



Monkey steals
banana from
right box.



Monkey leaves.
Person turns
back.



Cue given.

