Psychophysiological perspectives on autism

Psychofysiologische aspecten van autisme (met een samenvatting in het Nederlands)

PROEFSCHRIFT

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Marco Rudolf Hoeksma
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Promotores:	Prof. dr. H. van Engeland Prof. dr. M.N. Verbaten
Co-promotor:	Mw. Dr. C. Kemner

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Chapter One

Introduction

In this thesis, electrophysiological studies on visual and auditory selective attention and processing capacity in autism are described. To date, the literature has suggested several abnormalities in different aspects of information processing. However, detailed electrophysiological studies specifically aiming at selective attention or processing capacity are still scarce.

Studying selective attention and processing capacity can provide more insight in the underlying aspects of the clinical presentation of autism, such as difficulties in social interaction, restricted interests and preoccupations. Electrophysiological methods give the opportunity of following neural phenomena related to stimulus processing in great temporal detail. This not only allows for inferences on what aspects of information processing are deficient in autism, but also when these abnormalities first occur. Furthermore, accurate source localizations of electrophysiological abnormalities may provide invaluable insights in where such abnormalities may be located in the brain. The ERP studies published to date have focused on groups of autistic patients of different ages. However, none have made a direct comparison of autistic patients of different ages. Therefore, no information is available on the stability of the reported ERP abnormalities over time. In this thesis, studies are reported in which autistic children as well as adolescents participated in the same experiments. Taken together, such information may help to establish a biological marker for autism, thus aiding the diagnosis of the disorder.

Overview of this thesis

The present chapter serves to provide a theoretical background for the following experimental chapters. Current literature on the autistic disorder will be described and the concepts of selective attention, processing capacity and electrical source localization will be explicated. The following chapters are written as separate papers, in which experimental work is described. A general discussion concludes this thesis.

Autism is a severe developmental neuropsychiatric disorder, with an onset in the first three years of life. It essentially affects aspects of behaviour which are generally regarded as specifically 'human'. The core characteristics of autism are abnormalities in language, communication and social interaction, narrowed interests and stereotyped behaviour. Epidemiological data, reviewed by Fombonne [Fombonne 1999], show that the best estimate for the prevalence of autism is about 5.5 per 10,000, with a male/female ratio of 3.8 to 1. There are no indications of a link between autism and socioeconomic class or ethnicity.

Autism is often associated with other conditions: 80 percent of the autistic population has mental retardation and 25 percent will develop epilepsy. Furthermore, about six percent of the autistic population has other medical conditions like cerebral palsy, tuberous sclerosis or fragile X [Fombonne 1999].

Diagnosis

Since there is currently no clear biological marker for autism, the classification of the disorder relies on strict behavioural criteria, formulated in the Diagnostic and Statistical Manual of Mental Disorders (DSM-IV) (table 1-1). Autism, or Autistic Disorder (AD), falls within the broader category of Pervasive Developmental Disorders (PDD). This category includes a group of psychiatric disorders which share impairments in social skills, language development, and behavioural repertoire. The category of Pervasive Developmental Disorder -Not Otherwise Specified (PDD-NOS) is reserved for patients who show such impairments, but who do not meet all criteria for autistic disorder or any other pervasive developmental disorder. There is still considerable debate on the exact boundaries between autistic disorder and PDD-NOS [Lord and Risi 1996]. PDD-NOS inevitably shares considerable clinical similarity with autistic disorder. The most common differences between the two disorders are age of onset and severity of symptoms[Ciaranello and Ciaranello 1995]. It could be argued that autistic disorder lies on the severe end of a continuum of autistic features, and PDD-NOS represents a milder, or sub-threshold form. The opposite and least severe end of the continuum may be represented by mild communicative and social deficits or stereotyped behaviours. Such milder deficits, often called the 'broader autism phenotype', are more frequently observed in families of autistic individuals than in the general population [Piven et al. 1997;Rutter 2000]. It may well be that the disorders on this continuum share underlying neurobiological deficits [Ciaranello and Ciaranello 1995].

In the absence of biological markers for autistic disorder the diagnosis remains difficult, despite the strict classification scheme as established in the DSM-IV. Since the onset of symptoms has to occur before 36 months of age, autism is often diagnosed in retrospect. A diagnostic aid, widely used in research and clinical settings is the Autism

Diagnostic Interview-Revised (ADI-R)[Lord et al. 1994]. It is a semi-structured interview for care givers of suspected autistic individuals, that closely follows the criteria of the DSM-IV. When used by investigators who are trained in administration and scoring, the instrument shows excellent validity and reliability for individuals with mental ages from 18 months into adulthood [Lord et al. 1994].

Although autism was first conceptualized as a neuropathological disorder because of its early onset, in the 1950's the focus shifted towards 'refrigerator parents' as the primary cause of autism. This was caused by the observation that parents of children with autism were often socially inept (an observation which is now linked to the 'broader autism phenotype'). However, especially in recent years, science has produced compelling evidence that the first view, of a neuropathological origin for autism, is correct [Folstein 1999]. Twin studies have indicated that monozygotic (MZ) twins have a much higher concordance rate for autism than do dizygotic (DZ) twins. Furthermore, family studies have shown that siblings of autistic individuals have a recurrence risk for autism that is 100 times higher than in the general population [Rutter 2000]. Because of the disparity between the concordance rates for MZ and DZ twins, autism is likely to be caused by interactions among several genes. Recently, several genetic susceptibility loci for autism have been identified [International Molecular Genetic Study of Autism Consortium 1998;Cook 1998]. Genetic studies suggest a heritability of autism of about 90%. Furthermore, variability of symptoms within MZ twin pairs appears to be just as variable as between pairs. Thus, although a strong genetic factor is involved in autism, there is room for environmental or teratogenic factors [Rodier and Hyman 1998;Juul-Dam et al. 2001].

Cognitive functions

Currently, there are three leading theories regarding the cognitive problems in persons with autism. First, autistic individuals may have difficulties with Theory of Mind (ToM), or mentalizing. That is, persons with autism show an impaired ability to infer what other persons are thinking, which leads to problems in explaining or predicting behaviour [Happe 1999]. Interestingly, autistic individuals may attempt to make such inferences as frequently as normal persons, but they fail to make the correct inference [Abell et al. 2000]. ToM deficits are not unique to autism; similar, or somewhat milder deficits have been found in schizophrenia [Pilowsky et al. 2000] or following acquired brain damage [Happe et al. 1999].

The second leading view is that autism is characterized by abnormalities in executive functions. 'Executive functions' is a generic term for higher-level cognitive abilities for the control of action. Such abilities include planning and monitoring of behaviour, inhibition of automatic actions, set-shifting or flexibility, and working memory. Persons with autism especially show impairments in flexibility, showing errors of perseveration in

Table 1-1: Diagnostic criteria for autism according to DSM-IV

A. A total of at least six items from (1), (2), and (3), with at least two from (1), and one each from (2) and (3):

1. Qualitative impairment in social interaction, as manifested by at least two of the following:
   - marked impairment in the use of multiple nonverbal behaviors such as eye-to-eye gaze, facial expression, body postures, and gestures to regulate social interaction.
   - failure to develop peer relationships appropriate to developmental level
   - a lack of spontaneous seeking to share enjoyment, interests, or achievements with other people (e.g., by a lack of showing, bringing, or pointing out objects of interest)
   - lack of social or emotional reciprocity

2. Qualitative impairments in communication as manifested by at least one of the following:
   - delay in, or total lack of, the development of spoken language (not accompanied by an attempt to compensate through alternative modes of communication such as gesture or mime)
   - in individuals with adequate speech, marked impairment in the ability to initiate or sustain a conversation with others
   - stereotyped and repetitive use of language or idiosyncratic language
   - lack of varied, spontaneous make-believe play or social imitative play appropriate to developmental level

3. Restricted repetitive and stereotyped patterns of behavior, interests, and activities, as manifested by at least one of the following:
   - encompassing preoccupation with one or more stereotyped and restricted patterns of interest that is abnormal either in intensity or focus
   - apparently inflexible adherence to specific, nonfunctional routines or rituals
   - stereotyped and repetitive motor mannerisms (e.g., hand or finger flapping or twisting, or complex whole body movements)
   - persistent preoccupation with parts of objects

B. Delays or abnormal functioning in at least one of the following areas, with onset prior to age 3 years: (1) social interaction, (2) language as used in social communication, or (3) symbolic or imaginative play.

C. The disturbance is not better accounted for by Rett's Disorder or Childhood Disintegrative Disorder.

go-nogo tasks [Ozonoff et al. 1994] and in incompatibility tasks, where a subject has to inhibit a cued response [Noterdaeme et al. 2001].

Although abnormalities in ToM and executive functions may explain some aspects of the clinical picture of autism, like aberrant social behaviour or excessive desire for sameness, these theories are less capable of explaining areas of superior functioning in autism. The third leading theory, Central Coherence (CC), is more suited to explain such preserved or superior skills [Frith 1997]. Central coherence is the term to characterize the normal tendency to process incoming information in its context. Autistic individuals tend not to do so, they focus more on detail and less on the whole picture, and thus are said to have 'weak central coherence'. Weak central coherence can be demonstrated at the perceptual, visuospatial and verbal-semantic level. Weak CC on the perceptual level has been demonstrated by superior performance of autistic individuals on the judgment of visual illusions. Autistic subjects tend to be less distracted by the illusion-inducing context, and thus make more accurate judgments [Happe 1999]. On the verbal-semantic level, weak CC can (among others) be demonstrated by the use of homographs. Homographs are words that are spelled the same, but have different meanings and pronunciations. The correct meaning and pronunciation of a homograph can only be inferred when the context of the sentence in which it occurs is taken into account. Autistic subjects often fail to pronounce the word "lead" correctly in sentences like these: "Ann wanted to take the dog for a walk, so she went to get the lead", as opposed to: " The suitcase was so heavy it seemed like it was filled with lead" [Jolliffe and Baron-Cohen 1999]. On the visuospatial level, weak CC has been demonstrated by superior performance on the Block Design test of the Wechsler intelligence scales and on the Embedded Figures Task [Frith 1997]. Brain imaging supports the idea of a more piecemeal fashion of information processing in the Embedded Figures task [Ring et al. 1999], in that autistic subjects showed more ventral occipital activations suggestive of feature processing, where controls showed more frontal activations related to working memory. Other evidence for a preference for local processing comes from studies which have used the Navon task, in which large letters are presented that are made up of smaller letters (for example, a large letter H made up of small d's). When no prior information was given regarding to the level of appearance of the target (divided attention), autistic children showed an advantage in the processing of the smaller letters, as opposed to controls. When subjects were instructed to attend only to one level of information (the selective attention condition), autistic children performed similar to controls [Plaisted et al. 1999]. This can be interpreted as an impairment in shifting of attention in autism. Apparently, autistic individuals do not covertly shift their attention between global and local detail, but only do so when they are specifically primed. Attention appears to be less automatic in autism, and seems to rely on external cues.

Deficiencies in attention may be an aspect common to the theories of Executive Functions, Theory of Mind and Central Coherence. Abnormalities in attention have been a rather common finding in autism and may contribute to the clinical features of

autism [Allen and Courchesne 2001]. For example, the clinical observation of heightened reactivity to seemingly meaningless stimuli may be a sign of increased distractibility, while the narrowed interests and repetitive behaviours may be a representation of a deficit in shifting of attention. Therefore, attention seems to be an important area of research in autism.

Attention in itself is a broad term, which needs refinement in order to clarify what aspects of attention are under discussion. The main subdivision can be made between selective and sustained attention. Selective attention pertains to the filtering of a particular stimulus from one or more streams of incoming information. Sustained attention, or vigilance, is related to alertness and a steady-state preparation to react to a certain stimulus.

Sustained attention seems to be largely normal in autism. Autistic individuals have been found to perform similar to controls on the Continuous Performance Test (CPT) [Casey et al. 1993;Buchsbaum et al. 1992].

The focus in this thesis is on selective attention, since impairments in selective attention in autism have been demonstrated in several studies. For example, Burack showed that autistic individuals' performance in a forced-choice selective attention task was impaired when target stimuli were presented in the presence of distractor stimuli [Burack 1994]. This seems in line with clinical observations of increased distractibility in autism. However, Plaisted and colleagues showed that autistic individuals may show superior performance on selective attention tasks. When subjects had to select a conjunctive target (sharing colour with one set of distractors and shape with another set), autistic persons were significantly faster to find the targets than their controls. In the feature search condition, reaction times were normal in autistic subjects, which indicates that the shorter reaction times in the conjunction search were not the result of a general tendency for faster responses [Plaisted et al. 1998].

An important aspect of selective attention is the ability to move our attention across visual or auditory space and thus to make a spatial attention shift. Spatial attention shifting in autism has been studied using the Posner task [Posner et al. 1984]. In this task a subject is instructed to fixate on a central fixation stimulus and to press a button as fast as possible when a target appears on the right or left side of the fixation. Before a target is presented, a cue appears that can either be valid (on the side of the cue) or invalid (opposite the target location). Such a task requires three attentional operations: one has to disengage from the point of fixation, move to a new focus of attention (i.e., the target), and to engage attention at that location [Posner and Petersen 1990]. Casey and colleagues used the Posner task in autistic savants (having an extraordinary calender-calculating skill) and found that the autistic participants were more hindered by invalid cues than controls, as evident in larger reaction time increases from the valid to the invalid trials. This was interpreted as an inability to disengage attention [Casey et al. 1993]. However, a recent study using the gap-overlap paradigm suggested that the deficit may be located at the level of engagement [van der Geest et al. 2001] . In this task, a

subject is instructed to focus on a fixation cross and to move his eyes as fast as possible to a peripheral target. In the overlap condition the fixation cross is still present when the target appears, while in the gap condition the fixation stimulus is removed some time before the appearance of the target stimulus. Thus, saccadic reaction times are expected to be shorter in the gap condition because attention is already disengaged from the fixation location. The smaller difference in saccadic reaction time between the gap- and overlap condition in the autistic group as compared to controls was interpreted as autistic children showing a lower level of attentional engagement [van der Geest et al. 2001]. Courchesne and colleagues have proposed abnormalities in the cerebellum to lie at the basis of abnormalities in shifting or focusing of attention in autism [Courchesne et al. 1995;Harris et al. 1999]. Also, they found that patients with cerebellar damage and autistic patients may show similar spatial attention deficits [Townsend et al. 1999].

The studies mentioned above indicate that the attentional deficit in autism is at the level of selective attention or attention shifting. There are several steps in information processing that can be influenced by attention, or may be under 'attentional control'. Attention may already operate very early, at the level of perception, but also on working memory or response selection [Luck et al. 2000]. Because of their high time resolution, ERPs are very well suited for the study of such early, as well as the late aspects of attention.

The basic comparison to identify the effects of selective attention in ERP research is to compare the ERP waveform elicited by an attended stimulus to the waveform evoked by an identical stimulus when it is ignored. In many cases, these waveforms are subtracted, such that a difference wave remains. Whenever this wave significantly deviates from zero, this can be interpreted as an effect of attention. In both the visual and the auditory modality, the earliest effects of attention can be observed around 60 milliseconds (ms) after stimulus presentation. The timing of attention related peaks is dependent on the nature of the task involved. This early attentional processing occurs in modality specific areas of the brain [Luck et al. 2000;Woods et al. 1993].

In the auditory modality, two attentional components can be discerned. The earliest negative difference, Nde, occurs at latencies around 60 ms. Ndl, or late negative difference, is observed at latencies from 300-500 ms. Both components are thought to represent different stages of attentional processing, and to have distinct neuronal generators in temporal and temporo-parietal cortex. The early Nde is thought to represent feature selection, where Ndl is thought to be a correlate of the comparison of features against the target stimulus[Woods et al. 1993].

In the visual modality, three attention related differences can be observed in the ERP when selection has to be based on the co-occurrence of two (non-spatial) selection cues (or 'conjunctions'), like colour, shape, or spatial frequency. First, a frontal selection positivity (FSP) can be observed with an onset around 140-160 ms after stimulus presentation[Kenemans et al. 1993]. Second, an occipital selection negativity (SN) occurs, with an onset similar to FSP [Smid et al. 1997]. Third, around 200-250 ms a

centro-frontal N2b occurs [Lange et al. 1998]. These components are thought to reflect the activity of distinct functional mechanisms. FSP may be related to the selective association of a relevant stimulus with a relevant response [Kenemans et al. 1993;Smid et al. 1997]. SN would be more concerned with feature integration and selective analysis of the visual precept [Smid et al. 1997]. Finally, N2b is seen as a reflection of covert orienting of attention to stimuli that are classified as relevant by earlier attentional processes [Rugg et al. 1987].

In both the visual and the auditory modality, a large positivity or P3 can be observed from 300 ms onward, whenever a target stimulus is correctly detected [Picton 1992]. Two types of P3 responses have been identified: P3a, which is maximal over the frontocentral scalp, and P3b, which is maximal over the parietal scalp. The first, P3a, has often been associated with the identification of novel, highly intrusive information. However, there is evidence that P3a amplitude is dependent on the difficulty in target discrimination and not so much on novelty [Polich 1998]. P3b, the second P3 component, is associated with target detection in a general sense, and is thought to reflect attentional and memory-related operations. The amplitude of P3b increases with decreasing stimulus probability. Thus, rare target stimuli will elicit larger P3b than standard stimuli. Temporal probability also influences P3b amplitude, such that longer intervals between stimuli result in larger P3b amplitudes [Polich 1998]. In the remainder of this thesis, no formal distinction between P3a and P3b will be made. However, since the focus of this work is on the parietal/posterior P3, the insisting reader may read this as meaning P3b.

An important aspect of P3 amplitude is that it is proportional to the amount of attentional resources invested in a task. It is evident that human beings have a limited ability to process multiple streams of information. There appears to be a trading relationship, where extra resources needed for one process need to be borrowed from another. One can influence processing capacity and the amplitude of P3 experimentally by letting a person perform two tasks at the same time. Introduction of a second task reduces the P3 amplitudes on the first task [Isreal et al. 1980;Sirevaag et al. 1993;Kramer et al. 1987]. Also, when two tasks are performed, increasing the difficulty of one task increases the P3 amplitudes to that task at the expense of those in the secondary task. Similar results can be obtained when the second task does not require an explicit response. In such a task, irrelevant 'probe' stimuli are presented in stead of a secondary task. Difficulty of the primary task is manipulated, and the irrelevant stimuli are used to probe the amount of attentional capacity that is left. The more difficult the primary task, the higher the processing load, which means larger P3 amplitudes. For the probe stimuli, the reverse is true.

The most consistent results with such irrelevant probe tasks have been found when visual probe stimuli were used [Kok 1997]; auditory probes seem less susceptible to variations in task difficulty [Kramer et al. 1995].

To date, a number of ERP studies in autism have been reported. In the first study of event-related potentials (ERP) in autism, autistic subjects were presented with a train of visual or auditory stimuli, in which, at irregular intervals, a stimulus was deleted [Novick

et al. 1979]. The authors found that although autistic subjects were able to detect such omissions, they did not exhibit the same brain potentials as controls did, following a deleted stimulus. Although this study had some methodological problems, like a very small sample size (N=3), it was the incentive for many other ERP studies to follow. Most of these studies have followed a similar oddball design, that is they have presented a train of frequent stimuli with occasional intermittent deviant stimuli.

Studies that have focused on the auditory domain have produced the most consistent results. There are indications from auditory (brainstem) evoked potential studies that abnormalities in the auditory system may be present at a very early stage in autism, within several milliseconds after stimulus presentation [Thivierge et al. 1990;Buchwald et al. 1992;Bruneau et al. 1999]. No abnormalities were found at a somewhat longer latency, in a recent study of P50 gating [Kemner et al. 2002]. Auditory ERP studies have shown that abnormalities may also occur at a later stage in stimulus processing, mainly in the P3.

In a task similar to the one used by Novick et al (1979), Courchesne and colleagues found smaller P3 amplitudes on electrode Pz in autistic individuals when occasional targets where either present or absent in a train of ongoing stimuli [Courchesne et al. 1989]. Other oddball studies have reported similar smaller P3 amplitudes on Cz [Courchesne et al. 1985] [Dawson et al. 1988] and Pz [Lincoln et al. 1993]. Some investigators have added rare and highly deviant stimuli to the oddball design, thus making it a three stimulus paradigm. Abnormal ERPs to these highly unexpected stimuli, or novels, have revealed that autistic subjects process novel auditory information differently from controls, as indicated by abnormally small central A/Pcz/300 amplitudes [Courchesne et al. 1984;Courchesne et al. 1985;Kemner et al. 1995]. In a task in which subjects were required to pay attention only to auditory stimuli, which where presented concurrently with visual stimuli, autistic subjects not only showed smaller P3 amplitudes on Pz, but also an absence of attention related negativity (Nde and Ndl) [Ciesielski et al. 1990]. However, in a later report using the same task, none of these effects were replicated [Ciesielski et al. 1995].

For the visual modality, the evidence is less widespread. Similar to what was reported in the auditory modality, Courchesne et al [Courchesne et al. 1989] found smaller P3 amplitudes on Pz in an autistic group. Another study reported smaller frontal (Nc) amplitudes to novel and target stimuli in autism [Courchesne et al. 1985]. Furthermore, large P3 amplitude reductions in autism have been observed in oddball studies over central occipital electrode Oz in children [Verbaten et al. 1991;Kemner et al. 1994]. Similar to what was reported in the auditory modality, Ciesielski et al reported abnormalities in visual selective attention components in autism (a smaller N270) and parietal P3 [Ciesielski et al. 1990]. Again, none of these effects proved to be present in a later study using the same task [Ciesielski et al. 1995].

The ERP studies in autism reviewed above have for the most part found abnormally small P3 amplitudes over the parietal and occipital scalp. Abnormalities in P3 ampli-

tude are consistently reported in both adult and paediatric autistic groups. Most of these studies have used an oddball paradigm, in which the subject is only required to detect stimulus deviance in a single stream of information. Although oddball studies in autism have resulted in fairly consistent P3 abnormalities, none have found abnormalities in attentional components preceding the P3. This might be related to the fact that the oddball manipulation is not sensitive enough to detect abnormalities in selective attention. Indeed, the study by Ciesielski and others [Ciesielski et al. 1990], which is the only study to use some form of a selective attention paradigm, proved that there may be attention related abnormalities preceding the P3. Thus, a detailed study of selective attention in the auditory and the visual modality may help to clarify the nature of the often found P3 abnormalities in autism.

Brain structure in autism

It is reasonable to assume that attentional and other cognitive deficits in autism are not only related to abnormal brain function, but also to abnormal anatomy or neuronal make-up. There is wide evidence of abnormalities in the structure of the brains of autistic individuals. A limited number of post-mortem studies have shown increased numbers of neurons per unit volume and reduced cell size in the hippocampal area and the amygdala. On the other hand, a recent study by Casanova and colleagues [Casanova, Buxhoeveden, et al. 2002] suggested that frontal and temporal brain areas of autistic patients may show cortical cell columns of decreased width in which cells are more dispersed than in controls. However, the overall cell density in the gray matter of autistic patients seemed to be comparable to controls. In the cerebellum, reduced numbers of Purkinje cells have been noted [Bauman 1991]. On the macroscopic level, autistic individuals often show a head circumference that is larger than normal [Woodhouse et al. 1996;Fidler et al. 2000], or larger brains post-mortem [Bailey et al. 1998]. These observations have been substantiated by in-vivo Magnetic Resonance Imaging (MRI) studies, showing enlargement of the entire brain [Piven et al. 1995] or occipito-parietal and temporal regions [Piven et al. 1996a]. A recent MRI study suggested that autistic patients may show abnormal brain development with early overgrowth followed by an abnormally attenuated growth after the age of four [Courchesne et al. 2001]. Studies of specific regions of the brain have produced heterogeneous results, including indications of abnormalities in the parietal lobes [Courchesne et al. 1993], the amygdala and temporal gyri [Abell et al. 1999], caudate nucleus [Sears et al. 1999], hippocampus [Saitoh et al. 2001], the corpus callosum [Hardan et al. 2000] and the brain stem [Hashimoto et al. 1992].

The cerebellum has been cited as an important site of abnormalities in autism, especially by the group of Courchesne [Courchesne et al. 1988;Courchesne et al. 1995;Courchesne 1995;Saitoh et al. 1995;Courchesne 1997;Carper and Courchesne 2000]. However, the findings of hypo- or hyperplasia of the cerebellar vermis by the

Courchesne group have failed to be replicated [Holttum et al. 1992;Piven et al. 1992]or have been questioned by others [Filipek 1995;Peterson 1995].

Recent functional neuroimaging studies have provided the opportunity to make a direct link between brain function and anatomy. Many such studies have been carried out in autism, and they will be reviewed below.

Functional neuroimaging in autism

Functional neuroimaging studies using Positron Emission Tomography (PET) and Single Photon Emitting Tomography (SPECT) have indicated a diverse pattern of abnormalities in cerebral blood flow and metabolism in autistic individuals. Lower perfusion rates or glucose metabolism has been identified in the temporal lobes [George et al. 1992;Mountz et al. 1995;Ryu et al. 1999;Ohnishi et al. 2000;Starkstein et al. 2000;Zilbovicius et al. 2000], frontal areas [George et al. 1992;Siegel et al. 1995;Ohnishi et al. 2000;Hashimoto et al. 2000] and the parietal lobes [Mountz et al. 1995;Ryu et al. 1999]. Other regions showing abnormalities include the thalamus [Buchsbaum et al. 1992;Ryu et al. 1999;Starkstein et al. 2000] and the cingulate gyrus [Haznedar et al. 2000;Haznedar et al. 1997]. Furthermore, a number of studies have shown that autistic subjects may show an altered asymmetry in brain activation [Buchsbaum et al. 1992] [Chiron et al. 1995;Hashimoto et al. 2000]. Abnormalities in cerebral blood flow may be found in the absence of anatomical brain abnormalities [Chiron et al. 1995;Ryu et al. 1999]. Illustrative of the diverse findings in functional neuroimaging in autism are strongly contrasting findings of lower total brain perfusion [George et al. 1992] as opposed to a total absence of abnormalities [Zilbovicius et al. 1992].

Since PET and SPECT make use of radioactive tracers, the ethical constraints for such studies are very strict. This is especially a problem in control group selection; many control groups are comprised of patients referred for other medical conditions. Still, other studies are performed without a control group. Functional MRI (fMRI) can be performed without tracer substances, and thus does not suffer from these limitations. fMRI studies are typically performed in conditions where a subject has to perform a task. This has led to findings of weakened or absent ventral temporal activations in face processing tasks [Critchley et al. 2000;Schultz et al. 2000;Pierce et al. 2001], stronger activation of occipitotemporal regions in the Embedded Figures Task [Ring et al. 1999] and scattered activations when autistic patients performed a simple motor task [Muller et al. 2001].

Taken together, functional neuroimaging studies have provided an intricate picture of abnormalities. This may be due to the wide methodological differences between studies and the selection of control groups. With the advent of fMRI, results begin to converge, but this methodology poses limitations on available tasks. For instance, it is very difficult to perform auditory studies in the noisy environment of the MRI scanner. Furthermore, the time resolution of fMRI and of the other functional imaging methods is still limited. This may be the reason that none of the reported studies have focused

primarily on selective attention, although some studies have used cognitive tasks (for example the CPT [Buchsbaum, Siegel, et al. 1992]. Electrophysiological studies do not suffer from temporal limitations, and more complex study designs can be used. The spatial resolution of ERPs is limited, but this can be overcome by using source localization methods, which are described below.

Dipole analyses and Current Density reconstructions

Event-related potentials (ERPs) are derived from the electroencephalogram (EEG) and are electrical manifestations of brain activity following a certain stimulus event. EEG is recorded through scalp electrodes and are reflections of temporal and spatial summation of synchronized postsynaptic cortical potentials [Schaul 1998]. ERPs are computed from the ongoing EEG by averaging time-locked segments of the EEG to repeated stimuli. Averaging reduces background EEG (not time-locked to the stimulus) and noise, thus enhancing the ERP.

The advantages of ERPs over other neuroimaging methods are a very high temporal resolution (in the order of milliseconds), non-invasiveness, experimental flexibility and low cost. However, due to volume conduction effects, or the blurring of potentials by poorly conductive intermediate layers (i.e., the skull and skin), their spatial resolution is limited without the use of advanced source localization techniques.

Because of the poor conductivity of the skull, the potentials measured on the scalp electrodes will be attenuated and spread out. Therefore, there is no straightforward relationship between the recording electrodes and underlying cortical generators. This means that when a certain electrode shows a maximum potential amplitude, this can not be interpreted as this electrode lying directly over the cortical generator which produces this potential. Neuronal activations and inhibition are mainly mediated through current flow through the neuronal membrane. This way, one can think of a neuron as a small battery or dipole, with a positive (source) and negative (sink) side. There is as much current flowing out of the neuron as there is flowing in. When a patch of neurons oriented in parallel direction is concurrently active, this activity can be described with an equivalent dipole, reflecting the compound activity of the microscopic neuronal current fields. The aim of source localization techniques is then to calculate such a source configuration from the potential field measured on the scalp, a procedure which is also known as the inverse problem. However, one also needs to have information on the degree of attenuation and spreading of potentials measured on the scalp. Thus, information is needed as to what the potential field on the scalp would be given a certain intracranial source configuration. This is known as the forward problem. For solving the forward problem, a model of the head is needed, taking into account the conductivities of the layers lying between the generating surface (the cortex) and the recording site (the electrode). Classical methods have used a sphere in order to model the head, with a number of shells with different conductivities to account for the electrical properties of the scalp, skull and the brain.

The conductivity of the skull is about 80 times less than the conductivity of brain tissue, and thus accounts most for the spreading of the measured potentials [Scherg 1990].

The solution of the inverse problem has some limitations. First, the degrees of freedom for solving the solution is limited by the number of recording electrodes. The number of parameters that has to be estimated for one dipole source is six (3 for location, 3 for orientation and source strength). Thus, given the number of recording sites, only a limited number of independent sources can be computed. Furthermore, the computed location of a dipole does not have to correspond to the actual location of the activated brain structures, when in actuality more sources are responsible for the measured potential field. Therefore, the inverse problem is non-unique. These problems can be partly overcome by using a priori knowledge, for example on the number of sources or presumed hemispheric symmetry (using mirrored dipole pairs), for constraining the solution.

In dipole models, only a few dipole sources are computed. As long as the number of estimated dipoles is small and data are measured from an adequate number of electrodes, the reconstruction problem is overdetermined, i.e., there are more known than unknown parameters in the solution. The opposite is true for Current Source Density (CSD) reconstructions, a more advanced localization technique. Here, a very large number of current sources are placed on fixed locations, covering the entire spatial extent where activity may occur. A set of simultaneously active sources is computed by an optimization process. In this situation there are many more parameters to be estimated than there are measurements. Therefore, the model needs additional assumptions on the source model in order to make it solvable. Regularization techniques are used in the weighting between measured data and the source model. The simplest source model is the Minimum Norm assumption, where the sum of squares of the dipole strengths of the fixed current sources needs to be minimal for an adequate solution [Wagner 1998]. The physiological basis for this assumption lies in the idea that when the active brain always uses as little energy as possible. CSD methods have the advantage over dipole modelling that they require less knowledge beforehand on the possible number of active sources or on assumed source locations.

Recently, it has become possible to make individual head models derived from MR images, in stead of the spherical approximation used in traditional dipole modelling [Wagner 1998;Fuchs et al. 2001]. Such individual models allow for more detailed interpretation of the reconstructed sources, since they are directly linked to the anatomy. Furthermore, solutions can be constrained on the basis of anatomical information. For CSD reconstructions for example, the fixed sources may be distributed solely on (and/or perpendicular to) the cortical surface.

Conclusions

This chapter started with the notion that the study of selective attention and processing capacity in autism with electrophysiological measures can provide insight in

the underlying aspects of the clinical presentation of autism. Not only can such studies provide information on what aspects of information processing are abnormal, but also on when such abnormalities occur. When extended with accurate source localization methods, inferences can also be made about where these abnormalities in information processing are located in the brain.

Currently, Theory of Mind, Central Coherence and Executive functions are the three predominant cognitive theories that try to explain the cognitive abnormalities and, indirectly, the clinical presentation of autism. These three theories are rather elaborate frameworks covering many aspects of cognition. A deficiency in attention may be an important aspect that is shared between all three theories. Indeed, there are a number of behavioural studies that indicate that attention may be deficient in autism. These abnormalities have primarily been demonstrated in the field of selective attention and attention shifting.

Neuroimaging studies have provided a complicated and complex picture of functional abnormalities in the autistic brain. Although some of these studies have coupled cognitive tasks with functional imaging methods, none have focused primarily on selective attention. Moreover, attentional processes may act very fast and may consequently be imperceptible for relatively slow imaging methods such as PET, SPECT and fMRI.

In comparison to these imaging methods, ERPs have a superior time resolution. The spatial resolution of ERPs is limited without advanced source localization techniques. ERP studies in autism have shown that autistic individuals consistently show reduced P3 amplitudes compared to control groups. Such diminished P3 amplitudes are encountered across age groups; they are found in adults, adolescents and children. Furthermore, smaller P3 amplitudes are seen in the auditory and visual modality.

The P3 is related to target detection, and thus reflects the end stage of attentional processes. However, earlier attentional processes related to filtering and selection of relevant information precede the P3. Up until today, only one study [Ciesielski et al. 1990] has attempted to study these earlier selective aspects of attention in detail. Therefore, a systematic study of selective attention in autism seems warranted, in order to examine whether the abnormal P3 amplitudes in autism are a corollary of earlier deficits in attention. Since abnormal P3 has been found in different modalities and different age groups, the following chapters will present studies of selective attention in visual and auditory tasks in children as well as in adolescents. Based on the consistency in the current literature regarding the P3 in autism, one would not expect to find differences between these age groups. On the other hand, a recent structural MRI study by Courchesne and colleagues [Courchesne, Karns, et al. 2001] indicated there may abnormalities in brain growth in autism that vary with age. If such age differences do exist, they could ultimately lead to differences in brain potentials measured on the scalp. However, to date no ERP studies that directly compare two age groups of autistic patients in one and the same design have been published.

Attention and processing capacity are strongly linked and both have a strong effect on P3 amplitude. In order to examine whether the abnormally small P3 amplitudes

observed in autism are related to a deficiency in processing capacity, one of the following chapters will describe a study in which task difficulty is manipulated in a so-called probe task.

In the current chapter, the remark has been made several times that ERPs have a limited spatial resolution without the use of advanced source localization methods. Therefore, the ERPs from the visual selective attention task described in this thesis are subjected to high-resolution source analysis techniques. These techniques take advantage of the high temporal resolution of ERPs and the fine spatial resolution of structural MRI scans to answer the question of when and where in the brain attention related abnormalities occur in autism.

Chapter Two

The Development of Visual Event-Related Potentials in Autism

Marco R. Hoeksma
Chantal Kemner
Cisca Aerts
Marinus N. Verbaten
Herman van Engeland

Based on earlier findings of abnormal P3 amplitudes in autism, it was hypothesized that these abnormalities may be related to abnormalities in attentional processes preceding this processing stage.

Methods: Eighteen autistic children of about 10 years of age and 14 healthy control children and 10 autistic adolescents and 13 controls of about 19 years of age, performed a visual selective attention task during which the electroencephalogram was measured. Subjects were required to respond with a button press whenever a designated target stimulus was presented. The resulting Event-related Potentials (ERPs) were compared on three attention related peaks (FSP, SN and N2b) and were entered in an overall analysis to test for other effects, not related to selective attention.

Results: It was found that young autistic patients showed smaller occipital P1 and much smaller posterior P3 amplitudes than their matched controls. Adolescent autistic subjects mainly showed larger N2b amplitudes than autistic children.

Conclusions: Early abnormalities in visual processing seem to be present in childhood autism and the most prominent effects found in this study are not related to selective attention. Furthermore, the electrophysiological abnormalities observed in childhood autism seem to differ from those seen at adolescence.

Autism is a severe pervasive developmental disorder, which is characterized by disturbances in social interaction, language and speech development and stereotyped behaviour. The autistic syndrome has been extensively studied in many fields of research. An important number of studies suggest a neurobiological basis for autism [Bailey et al. 1996]. It is thought that important deficits in autism are those in perception and cognitive processing of sensory information [Minshew 1996]. Neuroimaging methods like Positron Emission Tomography (PET), Single Photon Emitting Tomography (SPECT), functional Magnetic Resonance Imaging (fMRI) and Event-Related brain Potentials (ERPs) provide valuable insight in how this processing takes place in the brain. Despite their relatively low spatial resolution, ERPs have the advantage over the other methods of having a superior temporal resolution and of being a direct measure of neural activity. This allows for following the time course of information processing in great detail.

Many ERP studies in autism have focused on the P3, a large positive wave starting around 300 milliseconds after a stimulus is presented. Abnormal P3 responses have been found in both the visual [Ciesielski et al. 1990;Courchesne et al. 1989;Kemner et al. 1994;Verbaten et al. 1991] and auditory modalities [Ciesielski et al. 1990;Courchesne et al. 1984;Courchesne et al. 1985;Courchesne et al. 1989;Kemner et al. 1995]. Except for Kemner et al., (1994, 1995) and Verbaten et al. (1991), these studies were carried out in adults or adolescents.

P3 abnormalities could serve as an index for the abnormalities in later stages of cognitive processing of sensory information. However, other defects in processes on the sensory or attentional level preceding the P3 could be responsible for such a P3 abnormality. Courchesne [Courchesne 1987] argued that there might be an active process interfering with earlier stages of processing, attenuating a normal flow of sensory information to later stages of processing. These early stages of information processing can be studied using a selective attention paradigm.

In selective attention tasks, a subject typically has to attend to one of two channels of information. Within these channels, frequently presented (standard) stimuli and rarely presented (deviant) stimuli are present. The subject is required to respond only to deviant stimuli within the attended channel. Selective visual attention to specific stimulus features, like colour selection, is characterized by a number of typical peaks in the ERP, which are thought to be functionally distinct. In adults, four different attention related components are distinguished in the difference wave resulting from subtracting unattended from attended standard stimuli. First, a frontal positivity, known as the frontal selection positivity (FSP) with an onset between 140 and 160ms post-stimulus [Kenemans et al. 1993]. Second, an occipital selection negativity (SN) is observed [Hillyard and Anllo-Vento 1998; Smid et al. 1997] with an onset latency of around 150-200ms. Third, around 200-250ms a central-frontal N2b occurs [Wijers et al. 1996]. Finally, a P3 (or P3b) occurs which is maximal over the parietal scalp and has an onset latency of 300-700ms [Picton 1992].

In autism, one selective attention study in adult autistics has been reported [Ciesielski et al. 1990]. This study used a combined visual and auditory task, in which the subjects had to attend to the visual and ignore the auditory stimuli or vice versa. In the visual modality, no attention effects were found on the N270 (comparable to the N2b) in the autistic group, while they were present in the control group. The authors did a separate test on the N270 of the four best performing autistic subjects who had task performances comparable to those of controls, but this test yielded the same results. Thus, the absence of attention effects on the N270 in autistic subjects could not be explained by differences in task performance. However, the interpretation of these effects are problematic, since attended and unattended channels stem from different experimental blocks and the study followed a cross modal design.

In the present study, we tested for the presence of diagnosis related differences in FSP, N2b and SN in children and adolescents, using a visual selective attention task as described by Jonkman [Jonkman et al. 1997]. To our knowledge, the present study is the first to study the developmental course of differences pertaining to selective attention between control and autistic groups. Furthermore, a more elaborate statistical design was used to test for the presence of additional differences between controls and autistic subjects.

Method

Subjects: School age children

The total initial sample consisted of 19 controls and 25 autistic children. From the control group, four children were excluded because of poor task performance (>50% omissions in three cases and 67% false alarms for the unattended deviant in one case) and one because of poor EEG quality. From the autistic subjects, three were excluded because of poor task performance (50% omissions in two cases, one subject with 47% omissions), one child was excluded on the basis of a known medical condition, one because of unclear diagnosis and two children were unable to complete the EEG recording session. The final clinical and control groups consisted of 18 and 14 subjects, respectively. The controls were all boys; one girl was included in the clinical group. There was no significant age difference between groups, mean ages 10.96 (sd 1.86, range 8-14.5) and 10.44 (sd 1.11, range 9.2-12.2) years for the clinical and control groups, respectively. The clinical subjects were recruited from the Department of Child and Adolescent Psychiatry at the Utrecht Academic Hospital. Controls were recruited from elementary schools in and around Utrecht.

All subjects were administered the Wechsler Intelligence Scale for Children, revised Dutch edition (WISC-RN). For autistic subjects, all diagnoses were based upon DSM-IV criteria and were made by a child psychiatrist (HvE) after extensive diagnostic evaluation, including a review of prior records (developmental history, child psychiatric and psychological observations and tests and neurological investigations). Furthermore, all autistic subjects were administered the Autism Diagnostic Interview Revised (ADI-R) [Lord et al. 1994] by a trained rater. Five subjects did not meet all the ADI-R cutoff

criteria for autism, but they did however meet the criteria for PDD-NOS as indicated by the psychiatrist. All subjects were medication free and had no significant neurological history.

The study was approved by the medical ethical committee of the Academic Hospital and all parents or caretakers gave written informed consent prior to participation. Furthermore, the child's assent was obtained and it was pointed out that participation in the experiment could be stopped at any time and for any reason by the child or the accompanying adult.

Table 2-1: Mean IQ scores for autistic and control groups. Standard deviations in parentheses, range in italics.

	Total	Performance	Verbal
young groups			
control	98.2 (9.68) ; 81-116	103.2 (13.15); 73-120	93.3 (9.04); 81-114
autistic	97.2 (14.03); 62-119	100.6 (19.88); 59-133	95.5 (14.26); 68-118
adolescent groups			
control	109.5 (8.19); 96-120	115.7 (9.76); 94-126	103.6 (7.99); 89-114
autistic	96.9 (10.91); 80-112	100.7 (13.25); 78-118	95.1 (11.13); 77-107

Subjects: Adolescents

Clinical subjects were recruited from a residential institution for autistic adolescents (the Dr. Leo Kanner house). Control subjects were recruited from a secondary school in Utrecht. The total initial sample consisted of 13 subjects in both clinical and control groups. From the autistic group, 3 subjects were excluded on the basis of poor task performance or because of technical problems. Thus, the final sample consisted of 10 autistic subjects and 13 controls. One female was included in the clinical group; the controls were all males. There were no significant differences in age between the groups; mean ages 19.0 (sd 3.37, range 15.2-24.5) and 18.2 (sd 0.74, range 17.2-19.6) years for clinical subjects and controls. All controls were administered the Wechsler Adult Intelligence Scale (WAIS), Dutch edition. For one autistic subject, the Wechsler Intelligence Scale revised Dutch edition (WISC-RN) was used. All autistic subjects were extensively diag-

nosed by psychiatrists at the Leo Kanner house. The ADI-R was administered to all subjects in the clinical group by a skilled rater and all subjects met the ADI-R criteria for autism. All subjects were extensively informed about the experimental procedures prior to participation. All subjects gave written informed consent. For subjects who were not of legal age, parents or caretakers were also asked to give written consent.

Although care was taken to match IQ scores between clinical and control groups, the autistic groups had significantly lower total and performance IQ than controls (TIQ; $F(1,53)=4.28$, $p=0.043$: PIQ; $F(1,53)=4.54$, $p=0.038$). The means suggest that the differences are especially large in the adolescent groups (see table 2-1). Post-hoc, analyses of IQ scores for each age group separately confirmed that the differences in total and performance IQ were significantly different in the adolescent groups (TIQ; $F(1,21)=10.1$, $p=0.005$: PIQ; ($F(1,21)=9.79$, $p=0.005$), but not in the young groups (all $F<.26$).

EEG and EOG recordings

Electroencephalic activity was recorded from 62 tin electrodes by means of an electrocap. Electrodes were placed on the scalp according to the 10% system of the American Electroencephalographic Society [1991]. An electrode attached to the left mastoid was used as reference. Horizontal EOG was recorded from tin electrodes attached to the outer canthus of each eye by means of adhesive rings. Vertical EOG was measured from infra- and supraorbitally placed electrodes at the left eye. A ground electrode was placed at the middle of the forehead. Impedances of the ground and reference electrodes were kept below 5 kOhms. All signals were amplified with a time constant of 10 seconds by a Sensorium EPA-5 amplifier (Sensorium inc., Charlotte, VT, USA). All signals were digitized on-line by a computer at a rate of 256 Hz and stored as a continuous signal. After sampling, signals were epoched off-line starting 100 ms before stimulus onset, and lasting for 1s. After epoching, all signals were filtered with a 30Hz, 24dB/octave digital low pass filter.

Task

The task consisted of 300 stimuli, 150 red and 150 yellow rectangles. The rectangles subtended a length of 4.5 degrees of arc and a width of 3.7 degrees of arc. They were presented for 50 ms each, with an inter-stimulus interval (ISI) randomized between 1750 and 2150 ms. Total task duration was about 10 minutes. During each recording session, three other tasks were presented, which will be discussed elsewhere. The order of presentation was balanced across subjects.

The stimulus attribute to define relevant and irrelevant channels was colour (i.e., yellow or red). Standards or deviant stimuli were defined by the orientation (to upper left \\ or upper right //, respectively) of thin, black diagonal bars in the rectangles. Within each colour, 20% were deviant and 80% were standard stimuli. Which orientation was deviant or standard was balanced across subjects, as was the relevant colour. Stimuli were presented in the centre of the visual field on a computer monitor positioned approximately 70 cm from the subject's eyes. The instruction was to press a button, which was

held in the preferred hand, as fast as possible to rectangles of one colour in which the orientation of the bars was deviant.

Procedure

A parent or caretaker always accompanied children who participated in the study. The autistic adolescents were in most cases accompanied by a supervisor. On arrival, they were familiarized with the procedure. After the electrocap and EOG electrodes were attached, a teeth mould was made which was used in the measurement of electrode positions after EEG recording (see below). The subject was then seated in a dentist's chair in an acoustically shielded room. The chair was adjusted so that the subject's head was approximately parallel to a computer monitor, positioned slightly above and in front of the subject. After attachment of the electrocap to the amplifiers and a check of the signals, the test session was started. Instructions for the visual task were given orally, and the subject had to perform a short practice series during which the experimenter gave feedback. When the experimenter was convinced that task requirements were met, the subject was instructed to move as little as possible during the task and to keep his eyes fixed on a fixation cross on the computer screen. The experimenter then left the room, closed the door and dimmed the lights. During the task, EEG was monitored on a computer screen. With children, in most cases the accompanying person was seated behind the child during recordings.

After the recording session was completed, the electrode positions were digitized by means of a Polhemus IsoTrak digitizer. When all experimental procedures were completed, children were rewarded with a toy, while adolescents were paid for their participation.

Signal analysis

EEG and EOG data were analysed off-line using the SCAN software package (Neuroscan Inc, El Paso TX, USA). All signals were baseline corrected on the basis of the 100ms pre stimulus interval. All epochs containing artifacts like saturation of the A/D converter, flat lines or amplitudes larger than +/-125 μV were removed. The EEG was corrected for EOG artifacts by subtracting vertical and horizontal EOG from EEG epochs by a regression method in the time domain [Kenemans et al. 1991].

ERPs were computed by averaging all remaining trials with correct performance for each subject in four stimulus categories (attended deviants, attended standards, unattended deviants and unattended standards) per lead. Thus, only attended deviants with hits and standards and unattended deviants with no response were included (see table 2-2). The resulting waveforms are shown in figure 2-1.

Statistical analysis

Repeated measures MANOVAs were performed for ERPs and task performance (proportions of omissions and false alarms and reaction times) separately. The significance level for all tests was set at $p < 5\%$, two-tailed.

Table 2-2: Means and standard deviations (in parentheses) of behavioural measures for all groups. Upper row schoolaged groups, bottom row adolescents. The adolescent groups made no errors in the unattended channel. RT = Reaction Time (in milliseconds); FA = False Alarms (proportion); Att Std = Attended Standards; Unatt Dev = Unattended Deviants; Unatt Std = Unattended Standards.

	RT	Omissions	FA Att Std	FA Unatt Dev	FA Unatt Std
Young groups					
Autistic	752 (128)	0.10 (0.07)	0.07 (0.10)	0.004 (0.01)	0.004 (0.008)
Control	705 (148)	0.06 (0.07)	0.03 (0.05)	0.005 (0.01)	0.0006 (0.002)
Adolescent groups					
Autistic	571 (165)	0.02 (0.03)	0.02 (0.02)	- (-)	- (-)
Control	530 (77)	0.02 (0.03)	0.005 (0.007)	- (-)	- (-)

For FSP, N2b and SN, analyses were done on difference waves constructed by subtracting the ERPs to unattended standards from those to attended standards. These analyses included a factor Diagnosis (autistics vs. controls) and Age (school age vs. adolescent) as between-subjects factors. In the school age group, the time windows used for these tests were 175-275ms for FSP, 250-450 ms for N2b and 150-200 ms for SN. In adolescents, the windows were adjusted to 150-250 ms for FSP, 200-350 ms for N2b and 150-300 ms for SN. Mean area amplitudes were scored in these windows. FSP was scored on electrode Fz, N2b was scored on Cz and SN on Oz. Since these comparisons are also a test of the efficacy of our manipulation of attention, attention effects are also reported when they do not interact with diagnosis. Difference waves representing FSP, N2b and SN are shown in figure 2-2.

From 50 to 350 ms the ERPs were divided in 12 segments of 25 ms in which the mean area amplitude was computed. In the 350-750 ms time range, four 100 ms segments were used. The mean amplitudes in these segments on electrodes Fz, Cz, Pz and Oz were entered in an overall analysis which was done for each time segment separately, with Diagnosis and Age as between-subjects factors and Channel (attended and unattended), Stimuli (standards and deviants) and Leads (Fz, Cz, Pz and Oz) as within-subjects factors. This analysis was included to test for any other differences between the groups present in the data. Since the effects of attention are already covered by the FSP, SN and N2b analyses, and to limit the number of tests, only interactions with diagnosis and age x diagnosis are now considered for further analysis. Also, from 50-350 ms, sub-

sequent tests were only done when two or more adjacent segments showed significant interactions. These measures were taken as protection against the increased probability of type I errors when performing multiple ANOVA.

Results

Performance

Three performance measures were obtained for each age group: percentage omissions, percentage false alarms and mean reaction time to hits. False alarms were divided into three categories; false alarms to attended standards, unattended deviants and attended standards. The young groups showed significantly longer reaction times ($F(1,53)=26.1$, $p=0.000$), more omissions ($F(1,53)=16.35$; $p=0.000$) and false alarms to the attended standard ($F(1,53)=5.96$, $p=0.018$) than the adolescent groups. Means and standard deviations per group for these measures are given in table 2-2. No effects of Diagnosis were found.

FSP, N2b and SN

All groups showed a significant FSP ($F(1,51)=13.39$, $p=0.001$) and N2b ($F(1,51)=28.35$, $p=0.000$), thus demonstrating the efficacy of our manipulation of selective attention. An Age x Diagnosis effect for N2b ($F(1,51)=4.97$, $p=0.03$) indicated that adolescent autistic subjects showed larger N2b amplitudes than young autistics ($F(1,26)=4.343$, $p=0.047$). The effect for SN was less straightforward: SN showed a main effect of Diagnosis ($F(1,51)=5.13$, $p=0.028$). Further analysis indicated that the autistic groups showed a significant SN ($t(27) = -3.449$, $p= 0.002$), whereas controls did not.

Overall analysis

From 75-125 ms significant Leads x Age x Diagnosis interactions were found ($F(3, 49)=3.942$, $p=0.01$; $F(3, 49)=7.547$, $p=0.000$), the young autistic groups showed smaller P1 amplitudes than controls on electrode Oz ($F(1,30)= 6.16$, $p=.019$; $F=8.27$, $p=0.007$). In the segment from 100-125 ms, this group also showed smaller amplitudes on electrodes Fz ($F= 5.74$, $p=0.023$) and Cz ($F=7.17$, $p=0.012$).

Next, from 225-650 ms the autistic groups as a whole showed smaller amplitudes on electrode Oz (all $F(1,53) > 7$, all $p<0.01$), as indicated by significant Leads x Diagnosis interactions (all $F(3,49) < 3.212$, all $p <0.031$) . From 450-650 ms, smaller amplitudes in the autistic group were also noted on electrode Pz ($F=8.93$, $p=.004$; $F=7.49$, $p=0.008$).

Discussion

Attention effects

In the present study a visual selective attention task was used in which subjects had to identify target stimuli based on a conjunction of colour and orientation of a superimposed grating. In adults, such a task usually elicits three different attention related peaks: A frontal selection positivity (FSP), a central selection negativity (N2b) and an occipital

selection negativity (SN). In the present study, significant main effects were found for FSP and N2b in both age groups, indicating a successful manipulation of attention. No overall significant SN was found. However, we did find an effect of Diagnosis on SN, which is discussed below.

Group Comparison

No group differences were found with respect tot the FSP, but there were group differences in N2b and SN. Adolescent autistic subjects showed larger N2b amplitudes than young autistic subjects. A similar age difference was not observed in the control groups. Although little is known about the functional process associated with N2b, it has been suggested that it reflects a feature non-specific attention process which is similar for attention to colour, location or conjunctions [Lange et al. 1998]. N2b would reflect covert orienting of attention to stimuli classified as relevant by earlier attentional processes [Rugg et al. 1987].

At first sight, the difference in SN between controls and autistics seems quite remarkable, since controls do not show a significant SN whereas autistic subjects do. Figure 2-2 shows that the young controls group even shows a positivity in the SN time-range. However, the particular relevance of this finding is unclear. It could be that in autistic subjects this stage of information processing has matured more rapidly or differently, since SN is usually reliably found in older age groups only [van der Stelt et al. 1998]. However, it then remains puzzling that the adolescent control groups do not show SN. Another explanation might be that the presence of SN in autistic subjects, and the absence thereof in controls, reflects a different selection strategy in autism. In a recent functional Magnetic Resonance Imaging (fMRI) study [Ring et al. 1999] using the Embedded Figures Task, it was found that where normal control subjects showed elevated activation of prefrontal brain regions associated with working memory, autistic subjects showed more ventral occipitotemporal activation associated with object feature analysis. The authors concluded that in autism, a different cognitive strategy is used to solve the task. A final possible explanation for the absence of SN in controls could be latency jitter in these groups.

In the overall analysis, apart from two isolated effects on Fz and Cz, the most notable group effects were seen on the posterior Pz and Oz leads in the autistic groups. The young autistic group showed a smaller P1 amplitude on Oz (75-125ms), independent of attention or stimulus type (figure 2-2). No such difference was found in the adolescent groups. P1 is a sensory-evoked, exogenous component which originates bilaterally in extrastriate cortex [Ossenblok et al. 1994], most likely from the fusiform gyrus [Heinze et al. 1998]. From flash-visual evoked potentials (FVEPs), it is known that the developmental course of the P1 amplitude shows an U shape, with amplitudes decreasing from 4-16 years of age and again increasing from 60 years onward [Dustman et al. 1996]. It has been suggested that this increase of P1 amplitude in older age might be a consequence of a decrease in inhibitory interneurons in the visual cortex [Diaz and Amenedo

Figure 2-1: Event-related potentials for all stimulus types, diagnostic- and age groups at four midline leads. Att dev = Attended deviant; Att std = Attended standard; Unatt dev = Unattended deviant; Unatt std = Unattended standard.

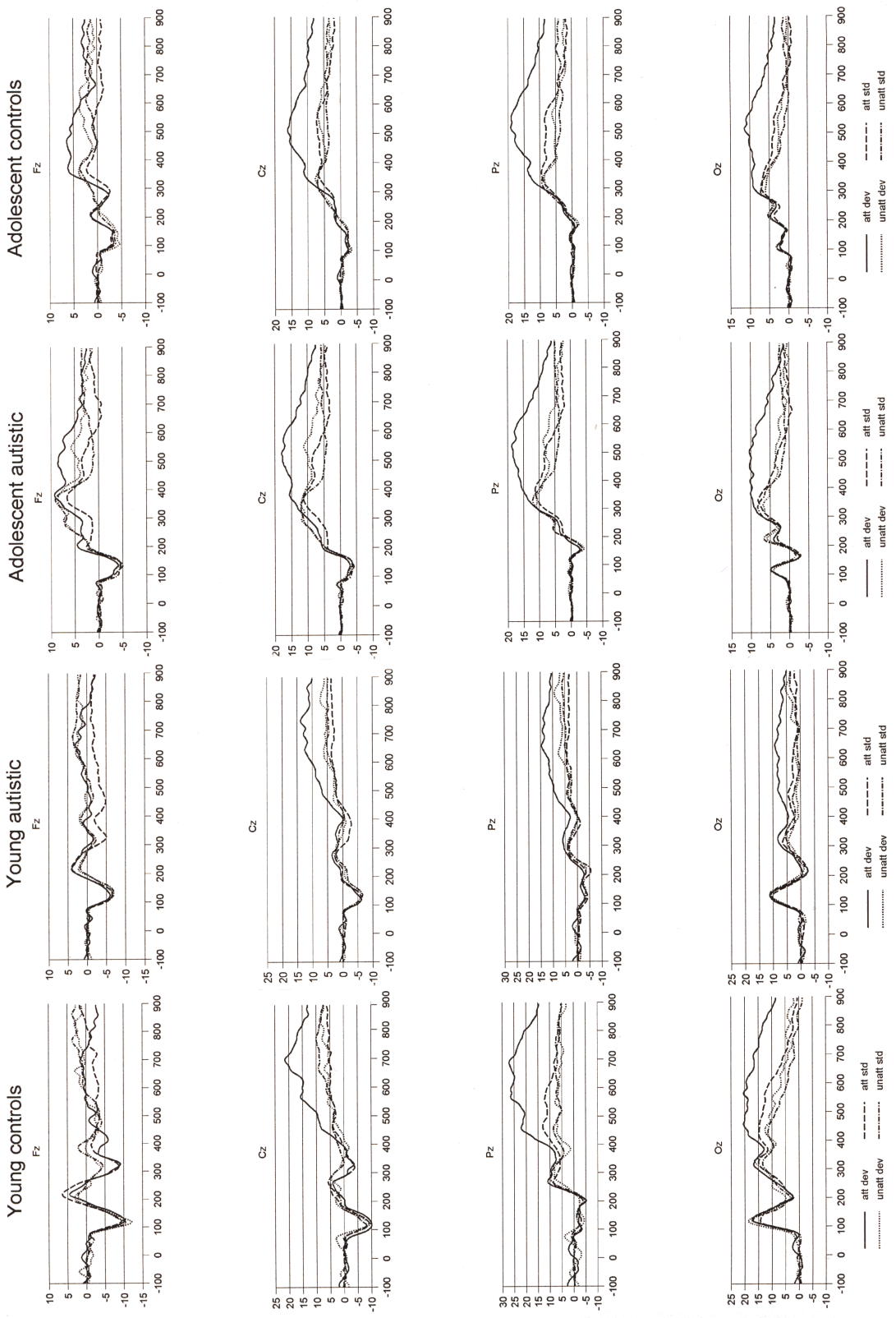
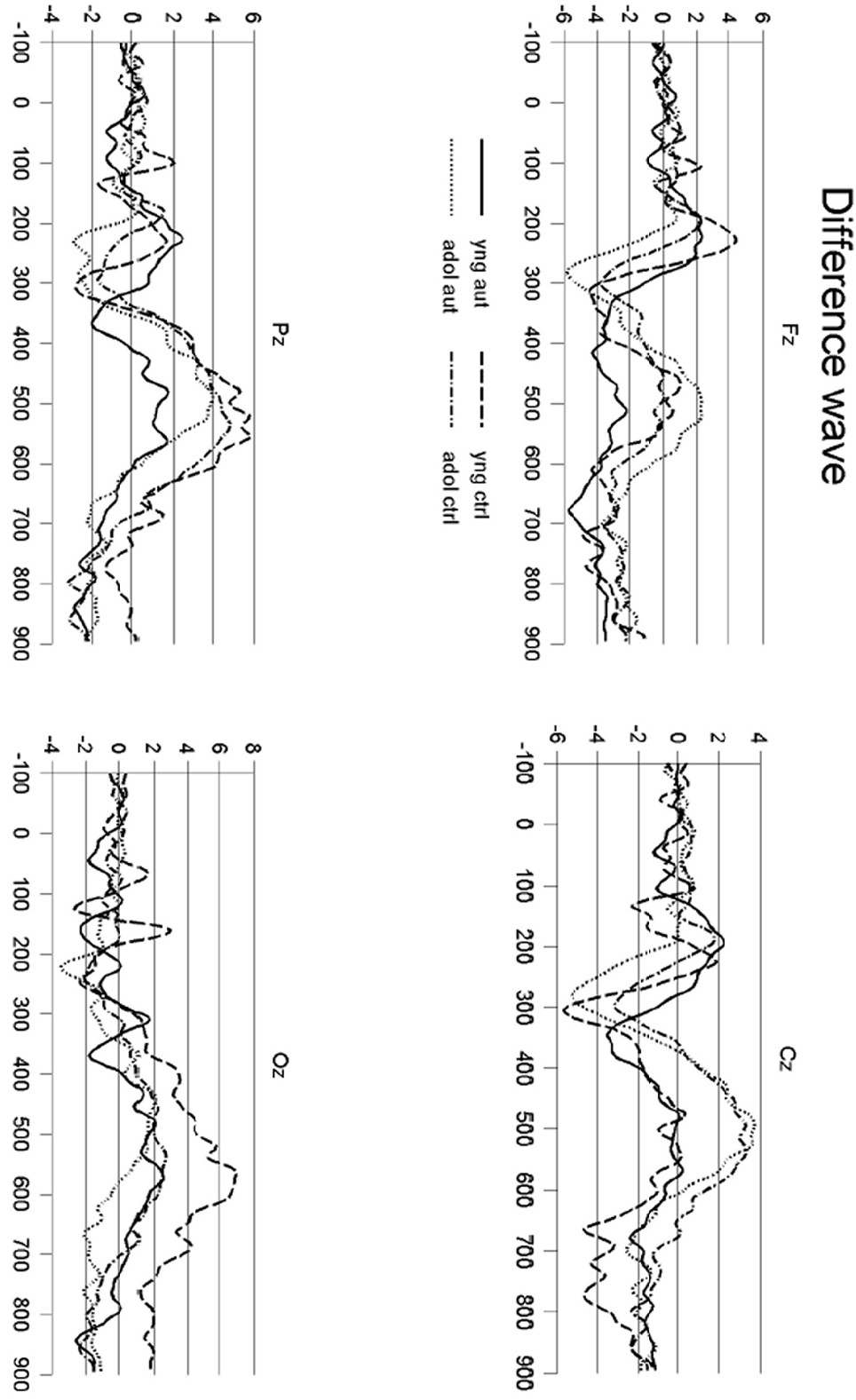


Figure 2-2: Difference wave of attended minus unattended standards at the four midline leads for all groups.



1998;Man'kovskii et al. 1978]. Extending this suggestion to childhood, the smaller P1 amplitudes in autistic children could be an indication of larger numbers of interneurons in autism and/or a more developed inhibitory function. Larger numbers of neurons could be compatible with reports of larger brain size in autism [Piven et al. 1995] [Piven et al. 1996a;Woodhouse et al. 1996]. Interestingly, Courchesne et al reported a study, which indicated that in autism brain size was larger in childhood only; at early adolescence the volumes were comparable to controls [Courchesne et al. 2001]. Since the origins of P1 in healthy subjects are known and since P1 is a rather focused and circumscribed peak, localization of its electrical sources will be very feasible. Hopefully, this will enable us to shed light on the question whether the P1 in autism originates from the same or different brain structures as in healthy subjects. It could be that the locations of the sources of P1 in autism are normal, but that the amplitudes are reduced as a result of poorer propagation of electrical signals due to different skull thickness, larger brain size or disordered neuronal organization. Electrical source localizations by means of realistic individual head models take some of these differences into account and may help to answer these questions.

A very striking result emerges when the P3 amplitude is considered (figure 2-1). The autistic groups, especially the young autistic subjects, show a marked reduction of P3 amplitude on electrodes Pz and Oz. [Ciesielski et al. 1990] and [Courchesne et al. 1989] also found reduced P3 amplitudes to visual stimuli (although smaller P3s were not reported in an earlier study [Courchesne et al. 1985]). However, these studies differed from the present study in terms of the task used. Also, none of these studies considered the Oz electrode in their analyses. This site proved to be the electrode with the most prominent abnormalities in the present study, as well as in the studies by [Kenemans et al. 1991] and [Kemner et al. 1995]. As can be seen in figure 2-1, the P3 amplitude reduction is much larger in the young autistic group than in the adolescents. On the other hand, autistic adolescents showed larger N2b amplitudes. It may be that this enlarged N2b reflects a compensatory process, normalizing the P3 abnormalities present from childhood. Since P3 amplitudes are most dramatically reduced in autistic children, paired with normal amplitudes of peaks related to selective attention in this group, we conclude that these reduced P3 amplitudes are not the result of abnormal selective attention.

To our knowledge, the present paper represents the first study concerning the development of visual selective attention in autism. Due to the use of ERPs, the flow of information could be followed with high temporal resolution. This high temporal resolution allowed the demonstration of abnormal information processing in childhood autism, first occurring around 100ms after stimulus onset. The finding of smaller occipital P3s in autism is consistent with the findings of Verbaten et al. (1991) and Kemner et al. (1995) with visual oddballs. To date, reports on visual ERPs in autistic adolescents have been less consistent [Novick et al. 1979; Courchesne et al., 1989, 1985; Ciesielski et al., 1990]. Our finding of clear P1 and P3 abnormalities in childhood autism and abnormal

N2b in autistic adolescents provides new and exciting insights in the pathophysiology of autism. In autistic children, abnormalities seem to originate before 100ms after stimulus processing, which underlines the value of the high temporal resolution of ERPs and indicates basic abnormalities in visual processing. These early abnormalities need to be corroborated by further research aimed at dissecting the circumstances in which these phenomena occur. One next step will be the accurate anatomical localization of the abnormal P1 by means of advanced source localization techniques. More definitive answers regarding the apparent dissociation between ERP amplitudes in childhood and adolescence can only be found in subsequent longitudinal studies.

Acknowledgements

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Chapter Three

Developmental Course of Auditory Event-Related Potentials in Autism

Marco R. Hoeksma
Chantal Kemner
Cisca Aerts
Marinus N. Verbaten
Herman van Engeland

Previous studies of auditory Event-related Potentials (ERPs) in autism have mainly focused on so-called oddball tasks and found smaller P3 amplitudes in autistic subjects. Most, but not all of these studies were conducted in autistic adolescents. The present study was designed to focus on true auditory selective attention and to study any age-related changes with respect to autism. ERPs were recorded in an auditory selective attention task in autistic children (mean age 10.6 yr.) and adolescents (mean age 19.1 yr.) and their matched controls. Autistic adolescents show a frontal processing negativity which starts earlier than in controls. Autistic children do not show such an abnormality. Furthermore, autistic children as well as adolescents show no significant differentiation in P3 amplitudes to attended and unattended deviants. Different abnormalities are found in autistic children and adolescents. The difference in processing negativity develops rather late, which is possibly related to a different pattern of brain growth in autism, and might reflect a compensatory process.

Autism is a severe pervasive developmental disorder, which is characterised by disturbances in social interaction, language and speech development and stereotyped behaviour. An important number of studies suggest a neurobiological basis for autism [Bailey et al. 1996]. Several event-related potential (ERP) studies in autism have indicated that autistic subjects show abnormal electrophysiological responses. The most consistent results in ERP studies are found in the auditory modality, especially with regard to the parietal P3 in response to target stimuli ([Courchesne et al. 1984;Courchesne et al. 1985;Courchesne et al. 1989] [Lincoln et al. 1993;Ciesielski et al. 1990]; but see [Kemner et al. 1995]). Most of these studies found smaller P3s in autistic subjects when compared to normal controls, in oddball tasks in which the subject had to react to the stimuli. However, in oddball tasks where no active response is required (a so-called passive condition), [Courchesne et al. 1984;Courchesne et al. 1985] and [Kemner et al. 1995] did not find these P3 differences.

The finding of the abnormal P3 responses in autistic subjects may be preceded in time by abnormalities in earlier ERP components, such as those related to selective attention. Also, from clinical experience it is known that autistic subjects may show hyper- or hyposensitivity to noises [O'Neill and Jones 1997]. This might be a behavioural reflection of deficient attentional processes. In a typical selective attention task, a subject is required to attend to one of two channels of information. Within these channels, frequently presented (standard) stimuli and rarely presented (deviant) stimuli are present. The subject has to respond only to deviant stimuli in the attended channel. Usually, a frontally distributed negative deflection with an onset around 200 ms (often called processing negativity, or PN) is seen in response to stimuli in the attended channel, as compared to stimuli presented in the unattended channel, reflecting selective attention or between channel selection. Selection of stimuli within channels is reflected by the P3.

Ciesielski et al. reported an absence of attention related activity at Fz in adult autistic subjects [Ciesielski et al. 1990]. These authors presented a task in which auditory as well as visual stimuli were included, with the active condition alternating between modalities. Thus, in one condition subjects had to attend to the auditory stimuli, while in the other condition they had to attend to the visual stimuli. However, because of the combination of visual and auditory stimuli (and because the activity in response to stimuli from different experimental blocks was compared), interpretation of these results with respect to auditory selective attention is difficult. Therefore, in the present study a selective attention task with only auditory stimuli is presented.

In the present paper auditory selective attention will be studied in two different age groups, namely school age children and adolescents. The inclusion of the two age groups will shed light on the stability of the ERP effects over time, as there are indications that behavioural abnormalities in autism may improve with age [Piven et al. 1996b] and that cortical areas in autistic subjects may show a delayed maturation (e.g. Zilbovicius et al. 1995). A recent Magnetic Resonance Imaging (MRI) study by Courchesne et al [Courchesne et al. 2001] indicated that autistic patients might show normal brain

volumes at birth, larger than normal volumes at 2-4 years, and again normal volumes at late childhood and adolescence. One would expect such volumetric differences to be associated with measurable functional differences.

The present study is the first to investigate the developmental course of event-related potential activity in persons with autism.

Methods

Subjects: School age children

The total initial samples consisted of 18 controls and 25 autistic children. From the autistic subjects, five were excluded because of poor task performance. Additionally, one child was excluded on the basis of a known medical condition, one because of unclear diagnosis and two children were unable to complete the EEG recording session. Six children from the control group were excluded on the basis of poor task performance and one because of poor EEG quality. The final clinical- and control groups consisted of 16 and 11 subjects, respectively. The controls were all boys; one girl was included in the clinical group. There was no significant age difference between groups, mean ages were 10.6 (range 8.0-13.6; sd 1.66) and 10.5 (range 9.2-12.2; sd 1.11) for the clinical and control groups, respectively. The clinical subjects were recruited from the department of Child and Adolescent Psychiatry at the Academic Hospital Utrecht. Controls were recruited from elementary schools in and around Utrecht.

All subjects were administered the Wechsler Intelligence Scale for Children, revised Dutch edition (WISC-RN). There were no differences in IQ scores between groups (for IQ scores, see table 3-1). For autistic subjects, all diagnoses were based upon DSM-IV criteria and were made by a child psychiatrist (HvE) after extensive diagnostic evaluation, including a review of prior records (developmental history, child psychiatric

Table 3-1: Mean IQ scores for autistic and control groups. Standard deviations in parentheses, range in italics.

	Total	Performance	Verbal
Young groups			
Autistic	96.9 (13.7); 62-119	98.8 (19.9); 59-133	96.3 (15.3); 68-116
Control	101.0 (8.7); 84-116	106.1 (10.0); 85-120	96.6 (8.7); 85-114
Adolescent groups			
Autistic	103.7 (10.7); 91-125	108.6 (12.3); 94-132	100.1 (11.3); 77-116
Control	109.5 (8.2); 96-120	115.7 (9.8) ;94-126	103.6 (8.0); 89-114

and psychological observations and tests and neurological investigations). Furthermore, all autistic subjects were administered the Autism Diagnostic Interview Revised (ADI-R) [Lord et al. 1994] by a trained rater. Five subjects did not meet the ADI-R criteria for autism, but they did however meet the criteria for PDD-NOS as indicated by the psychiatrist. All subjects were medication free and had no significant neurological history.

The study was approved by the medical ethical committee of the Academic Hospital and all parents or caretakers gave written informed consent prior to participation. Furthermore, the child's assent was obtained and it was pointed out that participation in the experiment could be stopped at any time and for any reason by the child or the accompanying adult.

Subjects: Adolescents

Clinical subjects were recruited from a residential institution for autistic adolescents (the Dr. Leo Kanner huis). Control subjects were recruited from a secondary school in Utrecht. The total initial sample consisted of 13 subjects in both clinical and control groups. From the autistic group, two subjects were excluded on the basis of poor task performance, and two because of technical problems. Thus, the final sample consisted of 9 autistic subjects and 13 controls. There were no significant differences in age between the groups (mean ages 19.1 (range 15.2-24.6; sd 3.43) and 18.2 (range 17.2-19.6; sd 0.74) for clinical subjects and controls). All controls were administered the Wechsler Adult Intelligence Scale (WAIS), Dutch edition. For one autistic subject, the Wechsler Intelligence Scale revised Dutch edition (WISC-RN) was used. There were no significant differences in IQ measures between groups (table 3-1). All autistic subjects were extensively diagnosed by psychiatrists at the Dr. Leo Kanner huis. The ADI-R was administered to all subjects in the clinical group by a skilled rater. All subjects met the ADI-R criteria for autism.

All subjects were extensively informed about the experimental procedures prior to participation. All subjects gave written informed consent. For subjects who were not of legal age, parents or caretakers were also asked to give written consent.

Although no significant IQ differences between diagnostic groups were found, significant IQ x Age effects showed that the young groups had significantly lower total ($F(1,47)=5.24$, $p=0.027$) and performance ($F=4.45$, $p=0.04$) IQ scores (young groups: TIQ 99.2 (sd 11.9), PIQ 102.7 (sd 16.9); adolescent groups: TIQ 107.1 (sd 9.5), PIQ 112.7 (sd 11.2)). Since no significant IQ differences between diagnostic groups exist, any diagnosis related effects in the electrophysiological measures or in task performance could not be contributed to IQ.

EEG and EOG recordings

The electroencephalogram (EEG) was recorded from 62 tin electrodes by means of an electrocap. Electrodes were placed on the scalp according to the 10% system of the American Electroencephalographic Society. From this array, four midline electrodes (Fz, Cz, Pz, Oz) were used in the statistical analysis. An electrode attached to the left

mastoid was used as reference. Horizontal EOG was recorded from tin electrodes, which were attached to the outer canthus of each eye by means of adhesive rings. Vertical EOG was measured from infra- and supraorbitally placed electrodes at the left eye. A ground electrode was placed at the middle of the forehead. All electrodes were filled with electrolyte paste (ECI inc.). Impedances of the ground and reference electrodes were kept below 5 kOhms. All signals were amplified with a time constant of 10s by a Sensorium EPA-5 amplifier (Sensorium inc., Charlotte, VT). All signals were digitised on-line by a computer at a rate of 256 Hz and stored as a continuous signal. After sampling, signals were epoched off-line starting 100 ms before stimulus onset, and lasting for 1 s. After epoching, all signals were filtered with a 30Hz, 24dB/octave digital low pass filter.

Task

The auditory selective attention task consisted of 300 stimuli, 150 presented in the left and 150 in the right ear. Within each ear (or channel), 20% were deviant and 80% were standard stimuli. The stimuli were sine waves of 1000 Hz and 1100 Hz with a duration of 50 ms each, with an inter-stimulus interval (ISI) randomised between 1750 and 2150 ms. Stimuli were presented through stereo in-ear headphones at 95dB sound pressure level. Total task duration was about 10 minutes. During each recording session, three other tasks were presented, which will be discussed elsewhere. The order of presentation was balanced across subjects.

The stimulus attribute to define relevant and irrelevant channels was the ear in which stimuli were presented (i.e., left or right). Standards or deviant stimuli were defined by the frequency (1000 Hz vs. 1100 Hz) of the tones. Which frequency was deviant or standard was balanced across subjects, as was the relevant ear. The instruction was to press a hand held button as fast as possible to tones with a deviant frequency in the attended ear.

Procedure

A parent or caretaker always accompanied children who participated in the study. The autistic adolescents were in most cases accompanied by a supervisor. On arrival, they were familiarised with the procedure. After the electrocap and EOG electrodes were attached, a teeth mould was made which was used in the measurement of electrode positions after EEG recording (see below). The subject was then seated in a dentist's chair in an acoustically shielded room. The chair was adjusted so that the subject's head was approximately parallel to a computer monitor, positioned slightly above and in front of the subject. After attachment of the electrocap to the amplifiers and a check of the signals, the test session was started. Instructions for the task were given orally, and the experimenter checked whether the subject was able to hear the subtle difference in frequency by letting them listen to a random set of stimuli which had to be classified (with an oral response) as 'high' or 'low' by the subject. After that, a short practice series was presented during which the experimenter gave feedback. When the experimenter was convinced that task requirements were met, the subject was instructed to move as little

as possible during the task and to keep his eyes fixed on a fixation cross on the computer screen. The experimenter then left the room, closed the door and dimmed the lights. During the task, EEG was monitored on a computer screen. With children, in most cases the accompanying person was seated behind the child during recordings.

After the recording session was completed, the teeth mould was used in the digitisation of electrode positions by means of a Polhemus IsoTrak digitizer. Subjects were then transferred to the MRI department, where whole-head MRI scans were made for use in future high-resolution electrical source imaging of the present data and volumetric analyses, which will be reported elsewhere. When all experimental procedures were completed, children were rewarded with a toy, while adolescents were paid for their participation.

Signal analysis

EEG and EOG data were analysed off-line using the SCAN software package (Neuroscan inc., El Paso TX). All signals were baseline corrected on the basis of the 100ms pre-stimulus interval. All epochs containing artefacts like saturation of the A/D converter, flat lines or amplitudes larger than ±125 (V were removed. After that, the EEG was corrected for vertical EOG artefacts by subtracting vertical and horizontal EOG from EEG epochs by a regression method in the time domain [Kenemans et al. 1991].

ERPs were computed by averaging all remaining trials with correct performance (hits and correct rejections) for each subject in four categories (attended deviants, attended standards, unattended deviants and unattended standards) per lead. The resulting waves are presented in figure 3-1. (For performance measures, see table 3-2).

Table 3-2: Mean error proportions and reaction times (in ms) in control and autistic groups (standard deviations in parentheses). FA = false alarms; as = attended standard; ud = unattended deviant; us = unattended standard; RT = reaction time.

	Omissions	FA as	FA ud	FA us	RT
Young groups					
Autistic	0.15 (0.12)	0.04 (0.05)	0.03 (0.05)	0.0005 (0.002)	710 (98)
Control	0.28 (0.24)	0.05 (0.06)	0.02 (0.03)	0.004 (0.01)	675 (104)
Adolescent groups					
Autistic	0.05 (0.06)	0.02 (0.03)	0	0.001 (0.003)	530 (144)
Control	0.08 (0.1)	0.03 (0.06)	0.008 (0.01)	0	514 (92)

Statistical analysis

Repeated measures MANOVAs were performed for ERPs and task performance (proportions of omissions and false alarms and reaction times) separately. The significance level for all tests was set at $p < 5\%$, two-tailed, Greenhouse-Geisser corrections for degrees of freedom were used where applicable. From 50 to 350 ms the ERPs were divided in 12 segments of 25 ms in which the mean area amplitude was computed. In the 350-750 ms time range, four 100 ms segments were used.

The mean amplitudes in all segments on electrodes Fz, Cz, Pz and Oz were entered in an overall analysis which was done for each time segment separately, with Diagnosis and Age as between-subjects factors and Channel (attended and unattended), Stimuli (standards and deviants) and Leads (Fz, Cz, Pz and Oz) as within-subjects factors. To limit the number of tests, only interactions that included the factor diagnosis were considered for further analysis. Also, from 50-350ms only, subsequent tests were only done when two or more adjacent 25 ms segments showed significant interactions. This measure was taken as protection against the increased probability for Type I errors when performing multiple ANOVA.

Results

Performance

Four variables were defined as measures of response accuracy: omissions (not pressing a button to a target) and three types of false alarms (to unattended deviants and to attended and unattended standards). No differences involving Diagnosis were present in the data. However, the young groups showed longer reaction times than the adolescents did ($F(1,48)=31.16$, $p=0.000$). Also, the young groups produced more omissions ($F(1,30)=10.37$, $p=0.002$) and false alarms to unattended deviants ($F(1,30)=4.55$, $p=0.038$). Performance measures are presented in table 3-2.

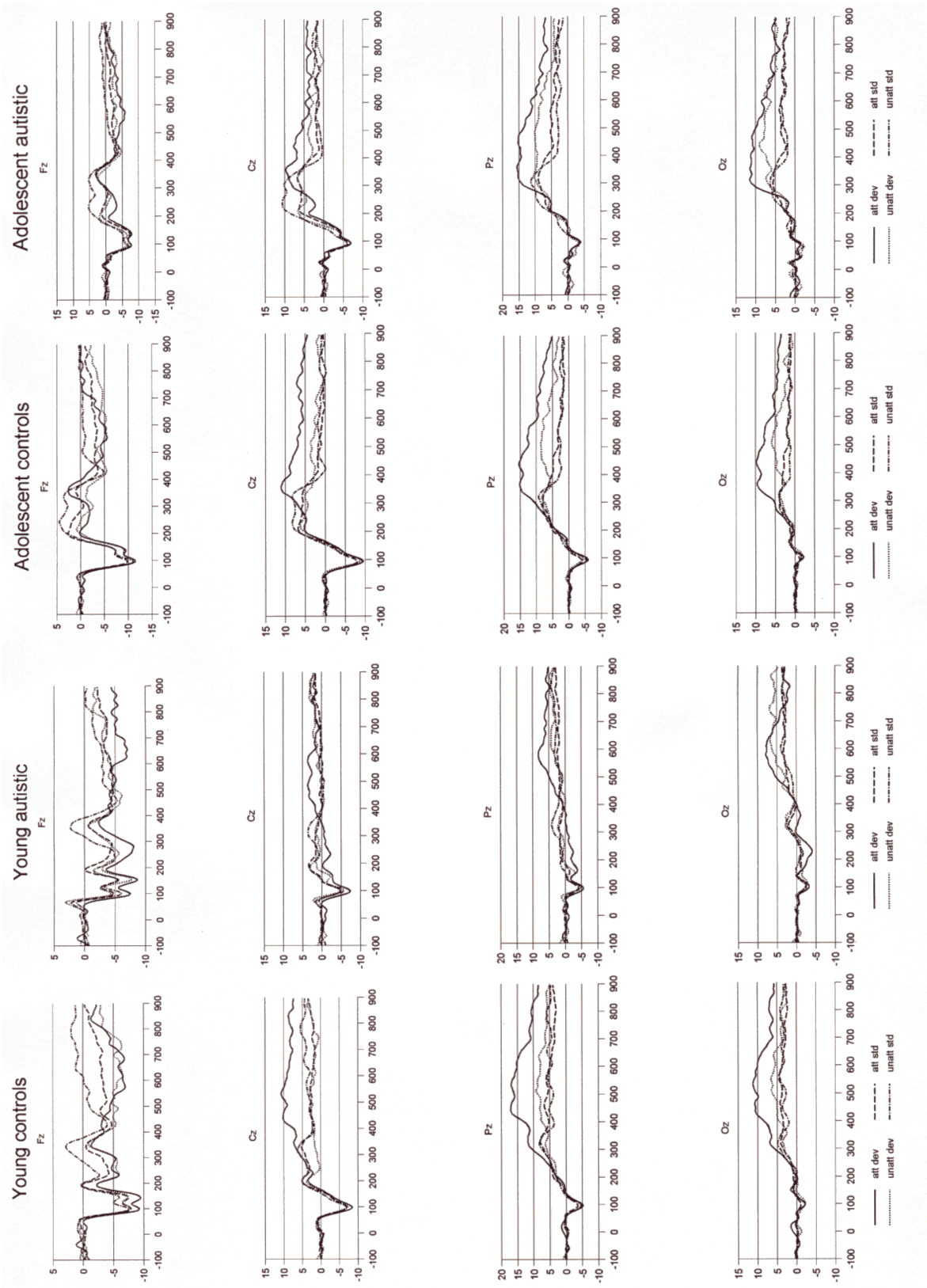
ERPs

A Channels x Leads interaction extending from 75-175ms and from 250-750ms indicated that our manipulation of attention was successful. When this interaction was tested per lead, the effect of Channel was mainly located at frontocentral leads from 50-350ms and at posterior leads from 350-750ms (fig. 3-2).

In the two segments from 150-200 ms the adolescent autistic subjects showed a significant attention effect (attended vs. unattended channel) on electrode Fz ($F(1,8)=16.79$, $p=0.003$; $F=23.75$, $p=0.001$) while their controls did not. From 200-275 ms, the adolescent autistic and control groups both showed an effect of attention on Fz ($F(1,21)=4.772$, $p=0.04$; $F=8.856$, $p=0.011$; $F=10.528$, $p=0.004$) (figure 3-3).

Next, from 350-450 ms the young autistic groups showed smaller overall amplitudes than their controls on electrode Pz ($F(1,25)=7.47$, $p=0.011$) and Oz ($F(1,25)=4.79$, $p=0.038$).

Figure 3-1: Event-related potentials for all stimulus types, diagnostic- and age groups at four midline leads. Att dev = attended deviant; Att std = attended standard; Unatt dev = unattended deviant; Unatt std = unattended standard.



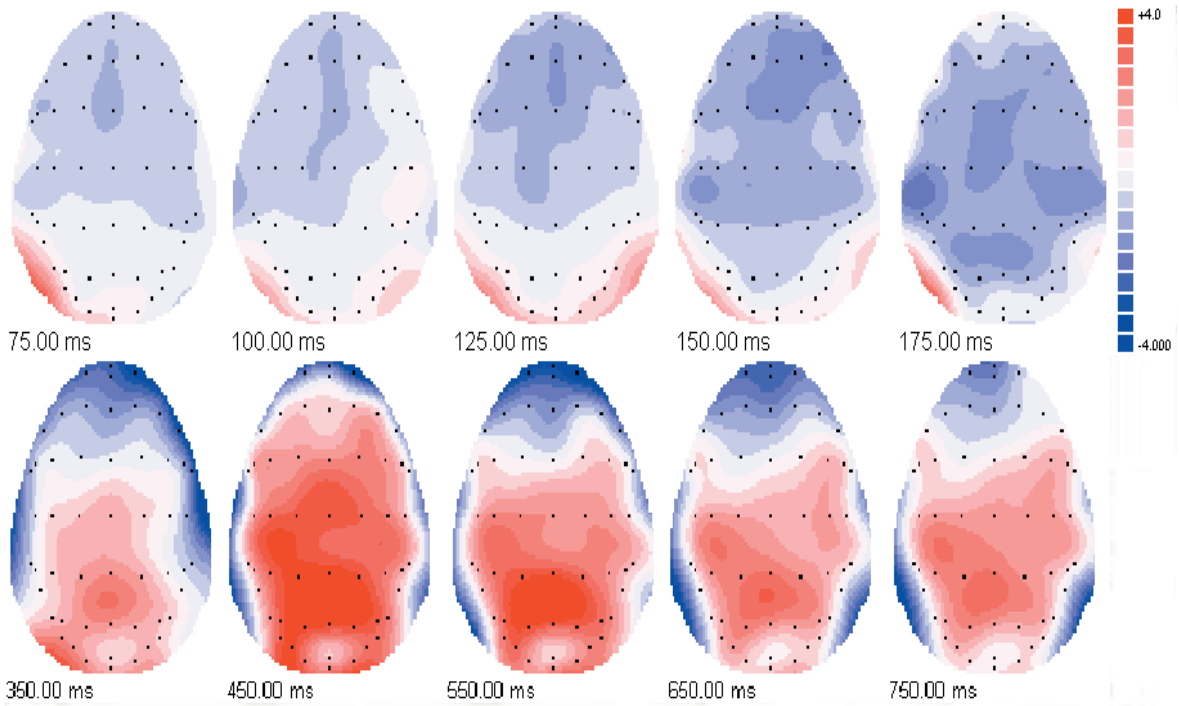
Finally, from 450-750 ms the control groups showed significantly larger amplitudes to attended than to unattended deviants ($F(1,23)=16.67$, $p=0.001$; $F=9.81$, $p=0.005$; $F=8.88$, $p=0.007$). From 650-750 ms, this was also true for attended and unattended standards ($F(1,23)=5.00$, $p=0.035$). The autistic groups did not show these differences (see figure 3-1).

Discussion

The aim of our study was to examine whether electrophysiological processes related to auditory selective attention differ between autistic individuals and controls, and to search for any age related differences in these processes between diagnostic groups. Although IQ was matched between controls and autistic subjects, the young groups showed lower IQ scores than the adolescents. This however, does not influence any diagnosis-related findings in our study.

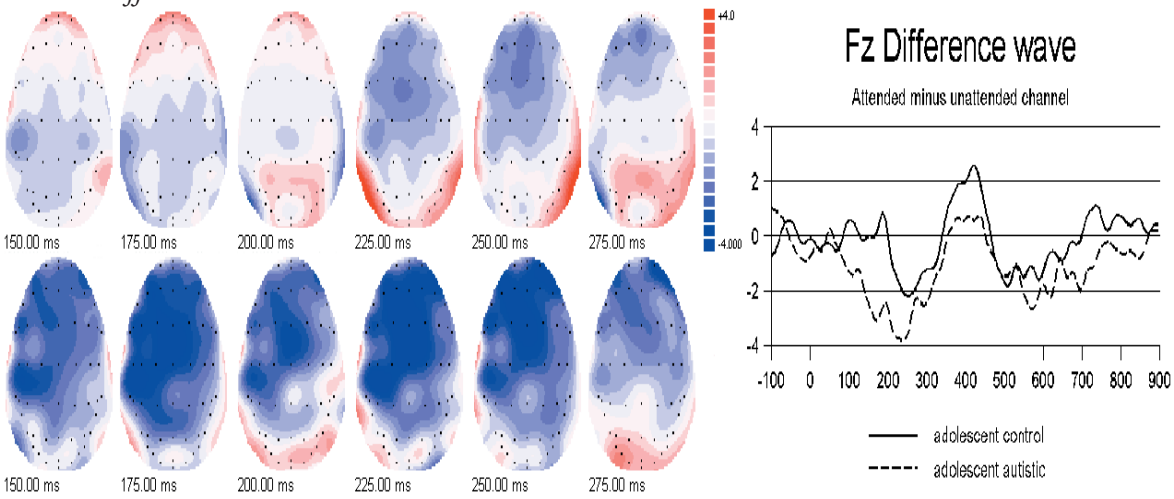
Behavioural measures showed that autistic subjects were as efficient in identifying the target from the non-targets as controls. No effects of diagnosis were present on either reaction times, percentage of omissions or false alarms. However, the young age groups showed longer reaction times, more omissions and more false alarms to unattended deviants. Thus, it seems that the task was harder for the school age groups, since they were both slower and less accurate in their responses.

Figure 3-2: Isopotential maps of the difference wave of attended minus unattended channel. From 350ms onward, the effect of attention takes the form of a posterior positivity. Before 350ms a frontocentral negativity is observed. (Scale in microvolts)



In the ERP data, the manipulation of attention proved to be successful, since significant channel x leads interactions were found from 75-175 ms and from 250-750 ms. In the segments between 75 and 350 ms, this attention effect was found to be significant mainly on Fz and Cz in the form of a negativity. From 350 ms onward, significant posterior positivities were found (electrodes Pz and Oz) (figure 3-2). These effects are comparable to the processing negativity (PN) and P3b effects as reported by [Jonkman et al. 1997].

Figure 3-3: Difference wave of attended minus unattended channel on electrode Fz and isopotential maps for adolescent groups. Top row controls; bottom row autistic. From 150-200ms, the negativity is only significant in the autistic group. From 200-275ms both diagnostic groups show a significant attention effect.



The first difference between age- and diagnostic groups was observed from 150-200 ms (figure 3-3). Here, the adolescent autistic group showed a significant attention effect on Fz while their controls did not. In the autistic group, the attention effect was sustained until 275 ms. For adolescent controls, the attention effect lasted from 200-275ms. Thus, the data suggest that in autistic adolescents the differentiation between relevant and irrelevant channels starts some 50 ms earlier than in controls but ends in the same time range. Figure 3-3 could suggest that the attention related negativity in the autistic group starts to differ from the controls as early as 75 ms after stimulus presentation, but the difference only becomes significant from 150 ms onward.

From 350-450 ms after stimulus presentation, the young autistic group showed smaller amplitudes to all stimuli on electrodes Pz and Oz, regardless of attention. Next, from 450-750 ms the attended deviants produced larger positivities than unattended deviants in the control groups of both ages, but the difference in the autistic groups did not reach significance. From 650-750 ms this was also true for standards.

Ciesielski et al reported absence of the auditory difference wave on Fz to standards (termed Nd in this paper) in autistic subjects with comparable ages as the present adolescent group [Ciesielski et al. 1990]. In particular the early phase of the Nd (Nde),

measured as the average amplitude over a window from 100-300 ms, was found to be absent in autistic subjects. It has to be noted that the task in this study differed considerably: there was an auditory and a visual channel, the pitch difference between standard and deviant stimuli was much larger (1000 Hz) and stimulus intensity was lower. In our data, adolescent autistic subjects show a negativity on Fz in this time range which is comparable in amplitude to controls, but starts approximately 50 ms earlier. Thus, where the data by Ciesielski were interpreted as an absence of the neurophysiological manifestation of interchannel selection, our data seem to indicate the contrary: the autistic adolescents show a broader negativity on Fz than controls. Furthermore, when this negativity overlaps in time with that in controls, the amplitude is comparable.

The broader negativity could be viewed in terms of overselectivity, but the behavioural data do not reflect this. Although no performance gains are seen when compared to controls, which could be due to the relative ease of the task, it could be that autistic subjects are overly selective in between channel discrimination in order to compensate for the less efficient within channel selection as reflected by the P3. The broader Fz negativity in adolescent autistics could thus be interpreted as a compensatory between-channel selection mechanism to balance subsequent abnormal within channel selection.

As for age-related differences between autistic groups, the present data suggest that autistic subjects develop differently with regard to early between channel selection. While being neurophysiologically similar to controls at early age in this study, at adolescence an attention-related negativity is observed which starts earlier and lasts longer than in controls. Furthermore, autistic subjects show no significant differentiation between P3 amplitudes to attended and unattended deviants.

It is known from clinical observations that autistic subjects may show an overreactivity to auditory, visual or tactile stimulation, which seems to be irrelevant for the bystander. Our data could be a reflection of the neurophysiological mechanism behind this phenomenon. Although in young autistic subjects irrelevant information is classified by early selection processes similar to controls, the abnormality arises in later stages of information processing when irrelevant, deviant information attracts a disproportionate amount of processing resources as indicated by absent P3 difference.

It should be noted that autistic subjects did not differ from controls on behavioural measures of task performance. Thus, the observed neurophysiological abnormalities did not have detrimental effects on task performance in a controlled laboratory environment. Yet, in situations where the flow of information is less structured, behavioural consequences from such a deficit may arise. Also, the error rate in performance scores of the adolescent groups was very low. It might be that the present task was too easy for these groups to produce performance differences, given the neurophysiological differences between controls and autistic subjects.

Courchesne et al reported a MRI study of autistic subjects in which brain development is investigated between 2-26 years of age. The authors concluded that newborns, which are later found to be autistic, have a head circumferences indicative of normal brain volumes at birth, whereas autistic 2-4 year olds have larger cerebral and cerebellar volumes than controls. In later childhood and adolescence, autistic patients again show normal brain volumes [Courchesne et al. 2001]. These findings suggest that autistic individuals show a pattern of brain development, which is entirely different from normal; it is characterised by an early phase of excessive growth, followed by a developmental arrest. Courchesne and colleagues speculated that this excessive growth occurs "without guidance" of functional experiences and adaptive learning [Courchesne et al. 2001]. It is likely that the abnormal brain growth results in abnormal functional processes, and this might be reflected in the abnormal P3 activity. After termination of abnormal growth, experience and adaptive learning shape the functional processes into a form which is different from normal, as indicated by the abnormal frontal processing negativity seen in the autistic adolescents. Indeed, this specific abnormality is not seen in autistic children, which suggests that it develops rather late. [Berman and Friedman 1995] reported that the development of auditory selective attention to pure tones from childhood to young adulthood is mainly characterised by shortening of the Nd latency. In our data, the latency of the frontal negativity was markedly reduced in autistic adolescents, which may be a sign of a more rapid maturation than in controls. Taken together, our data may reflect functional abnormalities in autism, which go together with the structural abnormalities as described by Courchesne et al[Courchesne et al. 2001].

The present paper, which is the first to study the developmental course of ERP responses in autism, suggests that autistic adolescents show early selection processes on electrode Fz which are different from autistic children and from normal subjects. Furthermore, autistic individuals show intra-channel processing which is different from normal, as evidenced by abnormal P3. Of course, an important limitation of the present study is its cross-sectional design in samples of fairly limited size. Longitudinal studies are needed to give more definitive data on the development of attentional processes in autism. In how far the functional abnormalities in our sample are paralleled by differences in brain morphology similar to those reported by Courchesne et al (2001) remains to be established. Future MRI-based high-resolution electrical source analyses on the present ERP data and volumetric studies on the MRIs from the present sample will yield more insight in this interesting question.

Acknowledgement

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Chapter Four

Processing Capacity in Autistic Children and Adolescents

Marco R. Hoeksma
Chantal Kemner
Marinus N. Verbaten
Herman van Engeland

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This study sought to investigate whether P3 abnormalities in autism are related to differences in processing capacity. Autistic children and adolescents and their control groups participated in the study. Visual probe stimuli were presented during the presentation of an auditory task with two levels of difficulty. Event-related potentials were measured from 62 electrodes during task performance. All groups showed amplitude increases to auditory stimuli with increasing task load. Controls showed smaller P3 amplitudes to visual probes, whereas autistic subjects did not. Furthermore, adolescent autistic subjects showed normal smaller P2 amplitudes on probe stimuli with increased load, where autistic children showed no effect. The results suggest that autistic subjects show abnormal capacity allocation. Some of these abnormalities may dissolve over time, while others remain into adolescence.

In a number of event-related potential (ERP) studies, autistic subjects have demonstrated smaller P3 amplitudes, both in visual [Courchesne et al. 1989] [Ciesielski et al. 1990] and auditory oddball studies [Courchesne et al. 1984;Courchesne et al. 1985;Courchesne et al. 1989]. Interestingly, such abnormalities have been reported for the parietal (Pz), as well as for the occipital (Oz) lead [Verbaten et al. 1991] [Kemner et al. 1994]. In two studies in which a visual and auditory selective attention paradigm was used, such smaller Pz and Oz amplitudes were also reported (Hoeksma et al, submitted).

There are, however, also some negative studies as far as the smaller Pz P3 amplitudes in autistic subjects are concerned. Verbaten et al (1991) reported smaller Pz and Oz P3 waves in a visual oddball, but only smaller Oz P3 in a (non-task) habituation series. Kemner et al (1994) also did not find smaller Pz P3 amplitudes in a passive visual oddball, but again found smaller Oz P3s in this non-task condition, in particular to standard stimuli. In the latter two cases stimuli were presented passively, not requiring a response. It might be that in these cases no effects on Pz P3 are seen because these conditions hardly draw on processing capacity. The frequently reported smaller Pz P3 waves in task conditions (both in oddball and selective attention paradigms) might point to a lack of processing capacity or a deficient allocation thereof in autistic subjects.

The idea that the P3 amplitude can be seen as an index of processing capacity stems from so-called dual task studies, in which subjects have to perform a secondary task against the background of an enduring primary task. Usually, dual tasks are comprised of a primary and a secondary task which both require responses from the subject. Additionally, the subject is usually required to keep his performance on the primary task constant, regardless of the introduction of the secondary task or of any manipulations of primary task difficulty. Isreal , Wickens, Chesney and Donchin (1980) used a visual detection task as primary task and an auditory secondary task. The visual detection task consisted of a display monitoring task, in which stimuli either changed in intensity (flash) or changed in the course they travelled across the screen (course change). The difficulty of the visual task was manipulated by increasing the number of visual elements on the screen. Introduction of the visual task reduced the parietal P3 amplitudes to the auditory task stimuli, but this amplitude was not further reduced when the display load in the primary task was increased. On the behavioural level, task performance improved with increasing task difficulty. Kramer, Sirevaag and Braune (1987) note that there is an inverse relationship between the P3s elicited by the primary and the secondary task, such that increases in the cognitive and/or perceptual difficulty of the primary task result in decreases of P3s elicited by the secondary task, while increasing the P3 amplitudes to the primary task. However, Kok (1997) points out that in probe tasks, in which no response is required for the secondary task stimuli, results with auditory probes have been small. With visual probes, more promising results have been reported, e.g. [Trejo et al. 1995]; [Sirevaag et al. 1993], but: "Strikingly, in these studies only the amplitude of the probe-evoked P3 was shown to decrease with an increase of the load of the primary task" [Kok

1997]. Thus, it seems that when the secondary task requires no response but is presented passively, the inverse relationship between primary and secondary task P3s does not hold.

Jonkman et al. (2000) used the irrelevant probe technique in a visual-visual setup with ADHD and control children. The authors claimed that the irrelevant probe technique is suitable to measure capacity trade-off when the probes are salient enough to attract attention and to allow for P3 processing. Therefore, secondary visual stimuli were divided into standards, deviants and novel stimuli. The novel stimuli were highly deviant from the standards and deviants and occurred only rarely. Furthermore, task load was manipulated which resulted in an increase in Pz P3 amplitude to primary task stimuli in the hard task versus the easy task in controls only. A planned comparison of P3 amplitudes on Pz to novels showed a decrease with increased difficulty in both groups, but this effect was only small (p=.06). The present study is based on the tasks used in the study by Jonkman et al (2000), and was modified so that the primary task stimuli were auditory and the irrelevant probes were visual (see below).

Although in adults, effects of task load are usually only seen at Pz (see e.g. [Verbaten et al. 1997]), it is less clear how load effects manifest themselves in children. In the study by Jonkman et al (2000) the reported load effects were all the result of planned comparisons on Pz. However, careful inspection of their figures suggests that such effects were also present at Oz, but unfortunately no tests are reported to substantiate this observation. Maximal visual P3 amplitudes are usually seen at Oz in children (also in Jonkman et al, 2000), and at Pz in adults. It might be that in children load effects also have a broader topographic range, including Oz. If so, this might explain the earlier reported smaller Pz and Oz P3 amplitudes under task conditions in autism [Verbaten et al. 1991]. A lack of processing capacity (the first hypothesis which is investigated in this study) may not explain the occurrence of smaller Oz P3 amplitudes under non-task conditions, since in such conditions a dissociation between Pz P3 (no difference between autistic and control) and Oz P3 (smaller in autistics) has been reported.

An alternative explanation for the abnormal Oz P3 in autism was proposed by Kemner et al. (1995), based on their findings of smaller Oz P3 amplitudes to passive visual standard stimuli [Kemner et al. 1994], and larger occipital P3s to auditory deviants in an active oddball [Kemner et al. 1995]. The authors argued that the larger occipital P3 amplitudes in autistic subjects for deviant auditory stimuli, combined with a smaller Oz-P3 to visual stimuli could reflect a deficiency in functioning of a parieto-occipital source with regard to visual processing. One explanation for this deficiency follows the line of what has been found in blind subjects. Following an enduring understimulation of posterior brain areas normally allocated to the visual system, the available 'overhead capacity' in these areas is reallocated to adjacent systems, i.c. the auditory system. In essence, this could mean that there is less processing capacity available for the visual system and more for the auditory system, as indexed by the P3 amplitudes in the studies by Kemner et al (1994, 1995).

Previous dual-task or irrelevant probe task studies have found the most prominent effects of dual task interference and task load on electrode Pz [Verbaten et al. 1997;Jonkman et al. 2000]. Here, we expect the amplitude of the P3 to auditory task stimuli to increase with increasing task load. Furthermore, we expect the amplitude of the P3 to visual probes on Pz to decrease with increasing task difficulty. Based on previous studies of the occipital P3 in autism [Verbaten et al. 1991]; [Kemner et al. 1995] Hoeksma et al, submitted) we expect visual probes to elicit smaller occipital P3s in autistic children when compared to controls. Since all auditory stimuli in the present task are relevant we could expect larger occipital P3s in autistic children, in line with the findings of Kemner et al (1995). Furthermore, when the smaller Oz P3 amplitudes to visual stimuli are indeed the result of a reallocation of visual processing capacity to the auditory modality, we would expect a more detrimental effect of task load on the visual probes. Thus, task load effects on visual probes on Oz would be more prominent in autistic subjects than in controls.

The expectations with regard to the occipital P3 as outlined above are only applicable to autistic children, since they are based on findings of abnormal occipital P3 amplitudes in autistic children [Verbaten et al. 1991]; [Kemner et al. 1994];[Kemner et al. 1995] Hoeksma et al submitted). In a visual selective attention task, Hoeksma et al (submitted) found markedly reduced occipital P3 amplitudes in autistic children, but the abnormalities were much smaller in autistic adolescents. Based on these findings, we would not expect any differences in task load effects in the latter age group.

Table 4-1: Mean IQ scores for autistic and control groups. Standard deviations in parentheses, range in italics.

	Total	Performance	Verbal
Young groups			
Autistic	96.3 (15.6) 62-119	99.9 (21.9) 59-135	93.1 (15.1) 68-116
Control	97.9 (8.8) 81-116	103.7 (11.8) 73-120	93.5 (8.4) 83-114
Adolescent groups			
Autistic	99.7 (12.6) 80-125	103.2 (15.1) 78-132	97.6 (11.4) 77-116
Control	109.5 (8.2) 96-120	115.7 (9.8) 94-126	103.6 (7.9) 89-114

Methods

Subjects: School age children

The initial groups consisted of 18 controls and 25 autistic children. From the control group, one child was excluded because of bad EEG data. From the autistic group,

one child was excluded because of a known medical condition, one because of unclear diagnosis and two children did not complete all decision tasks. Thus, the final control and clinical groups consisted of 17 and 21 subjects, respectively. Controls were all boys, one girl was included in the clinical group. There was no significant age difference between groups, with mean ages 10.2 (sd 1.6, range 7-13) for the controls and 10.7 (sd 1.86, range 7-14) for the autistic group. The autistic subjects were recruited from the Department of Child and Adolescent Psychiatry at the Utrecht Academic Hospital. Controls were recruited from elementary schools in and around Utrecht.

All subjects were administered the Wechsler Intelligence Scale for Children, revised Dutch edition (WISC-RN) (see table 4-1). All diagnoses were based upon DSM-IV criteria and were made by a child psychiatrist (HvE) after extensive diagnostic evaluation, including a review of prior records (developmental history, child psychiatric and neurological observations and tests and neurological investigations). Furthermore, all autistic subjects were administered the Autism Diagnostic Interview Revised (ADI-R) [Lord et al. 1994] by a trained rater. Five subjects did not meet the strict criteria for autism, but they did however meet the criteria for PDD-NOS. All subjects were medication free and had no significant neurological history. The study was approved by the medical ethical committee of the Academic Hospital and all parents or caretakers gave written informed consent prior to participation. Furthermore, the child's assent was obtained and it was pointed out that participation in the experiment could be stopped at any time and for any reason by the child or the accompanying adult.

Subjects: Adolescents

Clinical subjects were recruited from the dr. Leo Kanner house, a residential institution for autistic adolescents. Control subjects were recruited from a secondary school in Utrecht. The total sample consisted of 13 subjects in both the clinical and control groups. One female was included in the clinical group, the controls were all males. There were no significant differences in age between the groups (mean ages 19.6 (sd 3.2, range 15-24) and 18.2 (sd 0.7, range 17-19) for clinical and control subjects). All controls were administered the Wechsler Adult Intelligence Scale (WAIS), dutch edition. For one autistic subject, the WISC-RN was used (see table 4-1). All autistic subjects were extensively diagnosed by psychiatrists at the dr. Leo Kanner house. The ADI-R was administered to all clinical subjects by a skilled rater. All subjects met ADI-R criteria for autism.

All subjects were extensively informed about the experimental procedures prior to participation. All subjects gave written informed consent. For subjects who were not of legal age, consent was obtained from parents or caretakers as well.

There was a main effect of Age on total IQ ($F(1,60)=5.856$, $p=0.019$) and verbal IQ ($F=6.078$, $p=0.017$). The young groups showed lower scores on these measures than adolescents (mean TIQ young: 97.03 (sd 12.9), adolescent 104.6 (sd 11.5); mean VIQ young: 93.3 (sd 12.4), adolescent 100.6 (sd 10.1)). Since no effects of Diagnosis were

present on the IQ scores, any diagnosis related differences in ERPs or behavioural data can not be explained by differences in IQ.

EEG and EOG recordings

Electroencephalographic (EEG) activity was recorded from 62 pure tin electrodes, applied to the scalp by means of an electrocap. An electrode attached to the left mastoid was used as reference. The horizontal and vertical electro-oculogram (EOG) were measured from electrodes attached to the outer canthus of each eye and from infra- and supraorbital electrodes at the left eye. EOG electrodes were attached by means of adhesive rings. A ground electrode was attached to the middle of the forehead. All electrodes were filled with electrolytic gel. Electrode impedances of reference and ground electrodes were kept below 5kOhms. All signals were amplified with a time constant of 10s by a Sensorium EPA-5 amplifier (Sensorium Inc., Charlotte, VT, USA). All signals were digitized on-line by a computer at a rate of 256 Hz and stored as a continuous signal. After sampling, signals were epoched off-line, starting 100ms before stimulus onset and lasting for 1s. After epoching, all signals were filtered with a 30Hz, 24dB/octave low pass filter.

Tasks

During the experimental session, four tasks were presented. Two tasks were visual and auditory selective attention tasks, which will be described elsewhere. The other two tasks, and the focus of this paper, were an easy and a hard auditory decision task. In these tasks, the level of difficulty of the auditory task was manipulated, while visual probes were presented (see below). The order of presentation of hard and easy conditions was balanced across subjects. The tasks were based on the those used in the study by Jonkman et al (2000). The tasks were adapted so that the primary task stimuli were auditory and the irrelevant probes (or the 'passive secondary task') were visual. Apart from this modification the novel stimuli were altered, such that every novel stimulus was presented only once (which was not the case in the study by Jonkman et al). Another modification was made with respect to the ISIs, which were longer in the present task than in the study by Jonkman et al. Stimulus duration was 0.3s, and interstimulus intervals (ISIs) between primary (auditory) task stimuli was randomised between 4.2 and 5.4s. The easy and hard condition both consisted of two blocks. In the easy task, each block contained 70 task stimuli, which were animal sounds of a cat, dog, sheep and pig. These stimuli were presented binaurally through in-ear phones, at a level of 95 dB. In the easy task, the subject's instruction was to press a right hand button whenever a cat sound was presented and a left hand button for every other sound. In this condition there were 34 cat sounds and 36 non-cat sounds for each block to ensure approximately as many right- as left-hand button presses. In the hard task, the animal sounds were evenly divided. In this condition, the subject's task was to compare each sound that was presented with the preceding sound. When the sound was the same as the preceding sound, the subject was instructed to press the right hand button. When the sounds were dissimilar, the left hand

button was to be pressed. To ensure as many right- as left-hand button presses, the probability that two successive stimuli were (dis-)similar was 50%. Thus, for proper execution of this task condition, subjects had to keep a running memory of presented stimuli. In both the easy and hard condition all auditory task stimuli were task relevant, since they all required either a right or left hand button press.

Visual probes were presented in the centre of a computer screen, in between presentations of auditory task stimuli. There were 70 probe stimuli per block, divided into three stimulus types. Standards (60%) consisted of abstract black-and-white Attneave inf-80 stimuli (Attneave, 1954). Deviants (20%) were adapted from the standard stimuli, but the stimuli were divided into four evenly large quadrants which were each mirrored and rotated 180 degrees. Novels (20%) were unique abstract coloured patterns. Each novel occurred only once in the entire experimental session. All visual stimuli were presented on a computer monitor which was positioned at about one meter from the subject's eyes. Standard and deviant probes subtended a height of 5.7 degrees of arc and a width of 11 degrees of arc. Novels subtended a height of 10.5 degrees of arc and a width of 11.75 degrees of arc. Probe stimuli were presented with a duration of 0.3s, following a task stimulus with an ISI varying randomly between 1.95 and 2.55s. Subjects were told that in between task stimuli, pictures would be presented on the screen, which were to be attended but not acted upon.

Procedure

Children were accompanied by a parent or caretaker, all adolescent patients were accompanied by either a parent or a supervisor from the dr. Leo Kanner house. On arrival, they were familiarized with the procedure. After all electrodes were attached, a teeth mould was made which was used in the measurement of electrode positions. The subject was then seated in a dentist's chair in an acoustically shielded room. The chair was adjusted so that the subject's head was approximately parallel to a computer monitor, positioned slightly above and in front of the subject. After attachment of the electrodes to the amplifier and a check of the signals, the test session was started. Instructions for the tasks were given orally, and the subject had to perform a short practice series during which the experimenter followed the subject's responses on-line. When the experimenter was convinced that task requirements were met, the subject was instructed to move as little as possible during the task and to keep his eyes fixed on a fixation cross on the centre of the computer screen. The experimenter then dimmed the lights, left the room and closed the door. After each task block, the experimenter entered the room and gave instructions for the next block. During the task, EEG was monitored on a computer screen. With children, in most cases the accompanying person was seated behind the child during recordings.

After the recording session was completed, the positions of the scalp electrodes were digitized by means of a Polhemus Isotrak digitizer. When all experimental procedures

were completed, children were rewarded with a toy, while adolescents were paid for their participation.

Signal analysis

EEG and EOG data were analysed off-line using the SCAN software package (Neuroscan Inc, El Paso TX, USA). All signals were baseline corrected on the basis of the 100ms pre-stimulus interval. All epochs containing artifacts like saturation of the A/D converter, flat lines or amplitudes larger than +/- 125 μV were removed. EEG was corrected for vertical and horizontal eye movements by subtracting the EOG signals from the EEG epochs by means of a regression method in the time domain [Kenemans et al. 1991]. ERPs were computed by averaging all remaining trials with correct performance in every stimulus category. ERPs to auditory task stimuli were subsequently pooled, since they were all task relevant. For the probes, separate ERPs were computed for standards, deviants and novels. For the latter ERPs, only trials which were not responded to were included in the average. For statistical analyses, segment scores were computed for all ERPs, which are described below.

Table 4-2: Mean error proportions and reaction times for all groups. Standard deviations in parentheses. Omiss = omissions; RT = reaction time (ms); E = easy condition; H = hard condition.

	Omiss E	Omiss H	Errors E	Errors H	RT E	RT H
Young groups						
Autistic	0.042 (0.025)	0.017 (0.017)	0.05 (0.022)	0.067 (0.047)	1055 (228.4)	1297 (187.7)
Control	0.077 (0.049)	0.036 (0.036)	0.054 (0.033)	0.082 (0.05)	942 (313.3)	1164 (372)
Adolescent groups						
Autistic	0.094 (0.075)	0.046 (0.048)	0.048 (0.031)	0.059 (0.041)	811 (293.8)	1000 (372.7)
Control	0.115 (0.066)	0.047 (0.036)	0.044 (0.024)	0.039 (0.022)	725 (174.2)	918 (211.5)

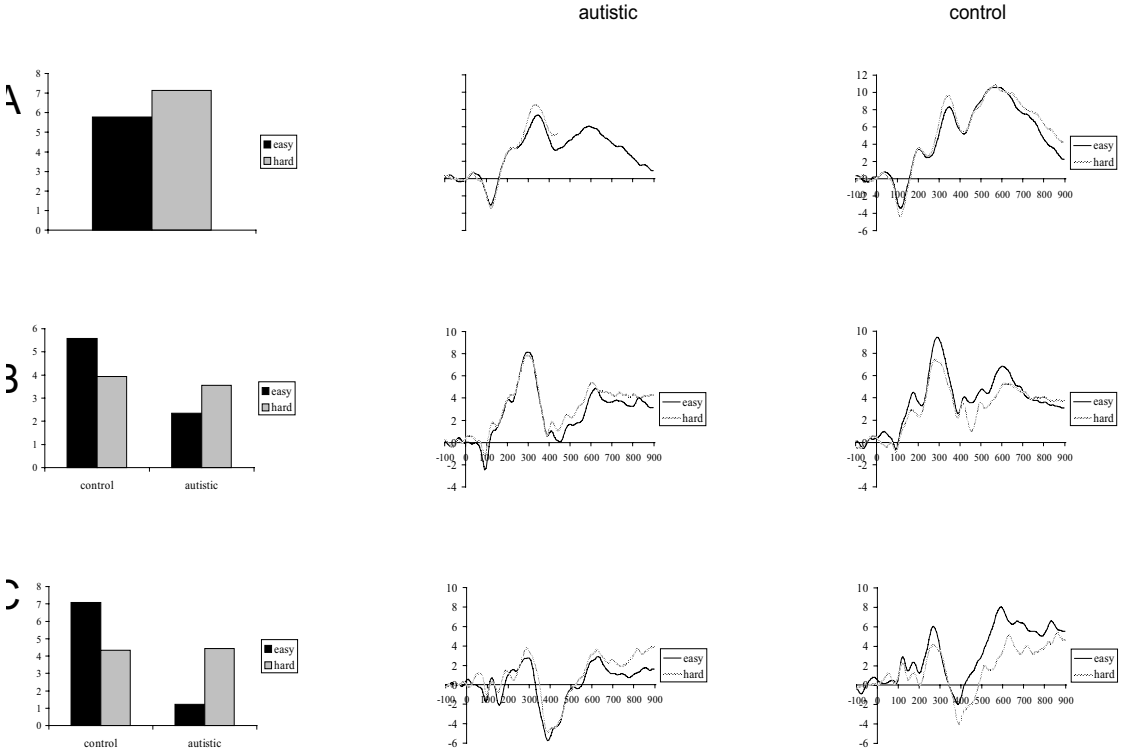
Statistical analyses

For behavioural data, errors, omissions and reaction times were separately entered in analyses with the factor Load as within variable (easy vs hard) and Diagnosis and Age as between variables. Reaction times were expected to increase with increasing task difficulty. For errors and omissions, a similar expectation seems plausible, although the data by Isreal et al (1980) indicated that when subjects are instructed to keep their performance optimal under increased task difficulty, the number of errors and omissions may even improve with increased task load.

From 50 to 350 ms the ERPs to visual probes were segmented into 12 segments of 25 ms in which the mean area amplitude was computed. From 350-750 ms four windows of 100 ms were used.

A prominent finding in previous research was the smaller (overall) Oz P3 in autistic children as compared to controls [Verbaten et al. 1991; Kemner et al. 1994; Hoeksma et al, submitted]. This finding, (and a single finding of a larger auditory Oz P3 [Kemner et al. 1995] led to the current question as to how the visual and auditory modality interact in autism, given the hypothetical underutilisation of visual areas [Kemner et al. 1994]. Thus, to answer this question a prerequisite is that we find a smaller overall Oz P3 to visual stimuli, at least in autistic children. Therefore, we tested the pooled amplitudes of visual probes and of standards only on Oz between groups, from 200-650ms. To test for larger occipital P3s to auditory task stimuli, similar to the finding by Kemner et al (1995), we also tested the pooled auditory stimuli on Oz in the same window, but for children only.

Figure 4-1: ERPs to auditory task stimuli, pooled standard and deviant probes and novel probes in the easy and hard condition on electrode Pz. Bars represent the mean amplitudes in the time windows in which significant effects were present (see text). ***A:*** *auditory task;* ***B:*** *pooled standard and deviant probes;* ***C:*** *Novel probes.*



Since we expected the most prominent effects of our difficulty manipulation (i.e., the effects of Load) on electrode Pz, we performed separate repeated measures analyses on auditory task stimuli, visual novels and pooled visual standards and deviants on this electrode from 200-650ms in a Load (hard, easy) x Diagnosis x Age design. We expected a reciprocal relationship between auditory task stimuli and visual probes, such that where the amplitudes to task stimuli would increase with increasing difficulty, the amplitudes to visual probes would decrease. This decrease was expected to be greatest for novels.

To test for any other effects present in the data, we performed an overall analysis for visual probes only, since a review by Kok (1997) suggested that effects of task load are usually most pronounced on probe stimuli only and not necessarily present on task stimuli. In this overall analysis, the mean amplitudes in each of the 16 windows on electrodes Fz, Cz, Pz and Oz were then entered into separate repeated measures MANOVAs for each window. This MANOVA contained the factors Diagnosis and Age as between-subjects factor and Load (easy and hard), Stimuli (standards, deviants and novels) and Leads (Fz, Cz, Pz and Oz) as within-subjects factors. For all analyses, to limit the number of tests only interactions with Diagnosis, Age x Diagnosis and Load were considered for further analysis. Also, subsequent tests were done only when two or more adjacent segments showed significant interactions. These measures were taken as protection against the increased likelihood of type I error when performing multiple MANOVA. The significance level for all tests was set at p<5%, two tailed.

Figure 4-2: Amplitude effects on electrode Oz from 450-550ms in children. Bars represent the mean amplitude from 450-550ms for standard probe stimuli and for all probes pooled. Means are computed from the plotted ERPs.

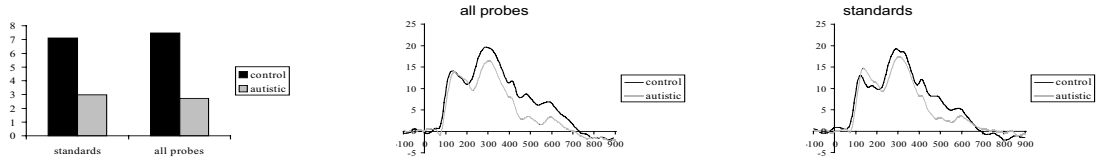
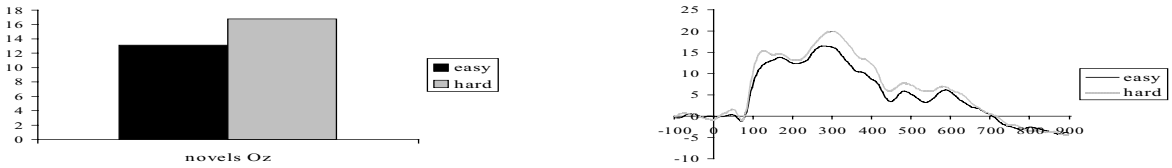


Figure 4-3: Novels elicit larger amplitudes in the hard condition in children from 275-450ms on electrode Oz. Bars represent the mean amplitude to novels in the easy and hard condition in this time window. Right panel: ERP to novels pooled over young autistic and control groups on electrode Oz.



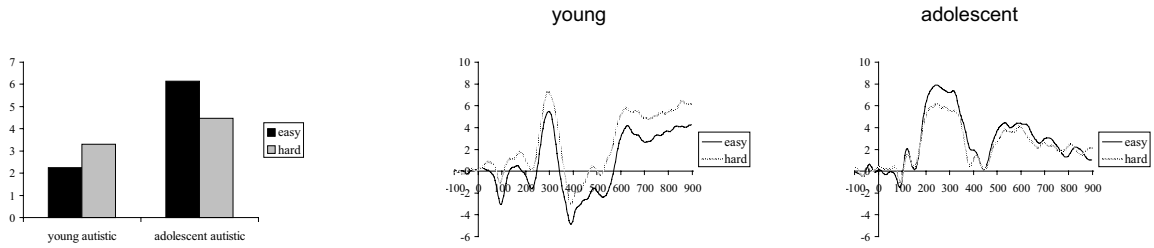
Results

Behavioural measures

Main effects of Load were present on all three behavioural measures. The number of errors increased with higher task load (F(1,58)=7.052);p=0.01), as did reaction times (F(1,58)=61.895);p=0.000). The reverse was true for the number of omissions, which showed a decrease when task load was increased (F(1,58)=65.41);p=0.000; Load x Age interaction F(1,58)=5.182, p=0.027). Furthermore, there was a larger increase in the number of errors with task load in the young groups (F(1,36)=9.947; p=0.003; Load x Age

interaction F(1,58)=4.343, p=0.042). The decrease in number of omissions with increased task load was larger for the adolescent groups (mean decrease 0.0579: F(1,24)=31.638; p=0.000)) than for the young groups (mean decrease 0.0317: F(1,36=26.937; p=0.000)). Behavioural measures are given in table 4-2.

Figure 4-4: On electrode Pz, the adolescent autistic subjects (right panel) show significantly smaller amplitudes in the hard condition than in the easy condition to all probes from 200-275ms. The young autistic groups (middle panel) show an insignificant increase in amplitudes. Bars represent mean amplitudes to probes in the easy and hard condition from 200-275ms.



Load Effects on Pz

There was a significant main effect of Load on auditory task stimuli from 275-350ms (all F(1,60)<4.9). All groups showed larger positivities in the hard condition (figure 4-1A). No further effects were noted for task stimuli.

For the pooled standard and deviant probes, there were significant Load x Diagnosis interactions from 450-650ms. Controls showed smaller amplitudes in the hard than in the easy condition (F(1,29)=5.842, p=.022; F=4.355, p=0.046). Autistic subjects showed insignificant increases in amplitude in the hard condition (F(1,33)=2.958, p=0.095; F=1.666, p=0.206)(Figure 4-1B). A similar effect was found on novel stimuli from 550-650 ms, where controls showed the expected smaller amplitudes with increased task load (F(1,29)=4.868, p=0.035). Again, autistic subjects showed small an insignificant increases in amplitude with increased load (F(1,33)=1.366, p=0.251)(figure 4-1C).

OZ P3 to visual probes

In children, we tested for all visual probes pooled in each Load condition, which revealed a significantly smaller Oz P3 between 450-550ms in the easy condition only (F(1,36)=6.08, p=0.019). A separate analysis for standards only (which showed the largest effect in the study by Kemner et al (1994)) also revealed the same effect (F (1,36)=4.58, p=0.039) (figure 4-2).

Oz P3 to auditory task stimuli

On electrode Oz, no indications were found for larger P3 amplitudes in autistic children, neither in the easy nor in the hard condition (all p>.10).

Load effects on Oz

On electrode Oz, no effects of Load were present in children on auditory task stimuli (all p>.08). For visual probes, the analysis (standards and deviants pooled and novels) resulted in Stimulus x Diagnosis interactions from 200-250ms. When this interaction was broken up, the remaining effect indicated that the autistic group showed smaller amplitudes to novels from 225-250ms ($F(1,36)=5.80$, $p=.021$).

From 275-450 ms significant Load x Stimulus interactions were found. Further analysis of these interactions indicated that both groups showed larger amplitudes to novels in the hard condition compared to the easy condition (all $p< .03$). These effects are displayed in figure 4-3.

Overall analysis

The overall analysis revealed significant interactions of Load x Diagnosis x Age from 200-300ms (all $F(1,36) > 4.5$, all $p<0.04$). When these interactions were broken up, they revealed that in the windows from 200-275 ms the adolescent autistic groups showed smaller amplitudes to probes in the hard than in the easy condition (all $F(1,12) > 5.0$, all $p<0.05$), whereas the young autistic groups did not show a load effect (figure 4-4).

Discussion

The present study was set up to investigate processing capacity in autism. To this end, a probe-task was used with two levels of difficulty. We expected the increased demand for processing capacity in the hard task to have a detrimental effect on behavioural data and to be reflected in P3 amplitudes to task and/or probe stimuli. On the behavioural level, reaction times as well as the number of errors were increased in the hard condition all groups. In the young groups there was a larger increase in the number of committed errors with increasing task load. There was a significant decrease in the number of omissions when task load was increased. The decrease in omissions was larger for autistic adolescents than for autistic children, which may indicate that the task was relatively easier for the adolescent group. The prolonged reaction times and increased number of errors can be seen as support for a successful manipulation of task difficulty. The decrease in the number of omissions might reflect an increased task engagement. Improvement of behavioural performance with increasing task load has also been reported by others [Isreal et al. 1980]. Thus, from our behavioural data we conclude that our manipulation of task difficulty or -load was successful and we therefore expected this effect to be visible in the ERP data as well.

Previous dual-task studies or studies using the irrelevant probe technique have found the most prominent effects of task interference on electrode Pz [Verbaten et al. 1997]; [Jonkman et al. 2000]. Also in the present study, the effect of task load on auditory task stimuli was present as a main effect of Load at Pz from 275-350 ms; all groups showed

increased Pz amplitudes in the hard task. Since the auditory stimuli required more elaborate processing in the hard condition, this increase was expected.

For visual probe stimuli, we expected the Pz amplitudes to decrease in the hard condition, relative to the easy condition. However, in this respect we found a significant difference between autistic subjects and controls, and between autistic children and adolescents. In the control groups, the effect of task load was present as expected; from 450-650 ms the control group showed smaller amplitudes in the hard than in the easy condition on standard and deviant probe stimuli. The same effect was found in controls on novel stimuli from 650-750 ms. The autistic subjects did not show a decrease in amplitudes with increasing task load. These groups showed small but insignificant increases in amplitudes to probe stimuli with increasing load. The overall analysis pointed out that from 200-275 ms, the adolescent autistic groups showed amplitude decreases to probe stimuli when task load was increased, while autistic children did not.

The absence of a reciprocal relationship between amplitudes to auditory task stimuli and visual probes in autistic subjects suggests that autistic subjects are abnormal in the utilization of processing capacity. Since autistic subjects showed small but insignificant increases in amplitudes to visual probes with increasing load, we can conclude that autistic subjects do not suffer from a shortage of processing capacity, but rather from a deficient allocation thereof. Interestingly, from 200-275 ms the adolescent autistic groups did show a load effect in the expected direction on probe stimuli, but this effect was not present in autistic children. Thus, deficient allocation of processing capacity in autistic subjects does not seem to be stable over time, and seems to develop more in the normal direction with increased age.

Kemner et al (1995) suggested that the smaller Oz P3 amplitudes to visual stimuli in autistic children might be the result of an understimulation of visual areas in autism, and that their larger Oz P3 to auditory stimuli [Kemner et al. 1995] might be a reflection of auditory processing drawing on processing capacity that is not used by the visual system. To gain insight in this notion, we used visual probe stimuli presented against the background of an auditory task. We hypothesized that any effects of task load on electrode Oz would be more detrimental in young autistic subjects than in controls. For this hypothesis to be tenable, we expected to find smaller Oz P3s to visual probes and larger Oz P3s to auditory task stimuli in autistic subjects when compared to controls. There was some indication of smaller Oz P3s to visual probes from 450-550 ms, but this effect was only small. No support was found for larger P3s to auditory task stimuli on Oz. Subsequently, on Oz we found a main effect of Load on novel stimuli only from 275-400 ms, with larger amplitudes in the hard than easy task, but no interactions with Diagnosis. Possibly, the larger amplitudes on novels on Pz in the hard task are a result of smearing of these larger occipital amplitudes. In sum, in the present data nothing seems to support the idea of reallocation of processing capacity from the visual to the auditory system, as put forward by Kemner et al (1995).

The data from the present study suggest that autistic subjects allocate processing capacity to visual probe stimuli in a different way than healthy controls. This is reflected by the phenomenon that autistic subjects do not show the usual trade-off between P3s to task- and probe stimuli as seen in controls. Surprisingly, autistic subjects show a tendency towards increased amplitudes with increasing task load. Furthermore, from 200-275 ms a dissociation is seen between young and adolescent autistic subjects, which suggests that these event-related potential abnormalities normalize with age. Further characterization of these abnormalities and the extent of the apparent normalization will be possible by means of accurate localizations of the sources of the measured ERPs. Such localizations will give insight in the brain structures involved in these processes, making it a logical next step in our research.

Chapter Five

Localization of Visual Event-Related Potential Abnormalites in Autism in MRI-based Individual Head Models

Marco R. Hoeksma
Chantal Kemner
Marinus N. Verbaten
Herman van Engeland

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Abnormal event-related potentials are a common finding in autism. However, to date no attempts have been made to localize the brain correlates of such abnormalities. Here, we report a study in which we used high resolution EEG and realistic individual head models to localize visual P1 and P3 abnormalities in autistic children and adolescents.

Methods: We used high-resolution visual event-related potential data and MRI based structural scans of the head in order to localize visual P1 and P3 peaks in autistic children, adolescents and their controls. Dipole locations were transformed to standardized space to allow for statistical comparisons.

Results: We found that dipoles for P1 were located more superiorly in autistic children when compared to controls. No abnormalities were found in adolescent groups. Cortical Current Source Density (CSD) maps suggested that patients with autism exhibit a more complex pattern of activation than controls.

Conclusions: These results demonstrate that with accurate source localization techniques, differences in the locus of cortical activation can be observed in childhood autism, as early as 100 ms after a visual stimulus is presented.

Abnormalities in Event-related potentials (ERPs) have been a rather common finding in autism research. Abnormal P3 amplitudes have been found in both visual [Ciesielski et al. 1990;Courchesne et al. 1989;Kemner et al. 1994;Verbaten et al. 1991;Novick et al. 1979]and auditory modalities [Ciesielski et al. 1990;Courchesne et al. 1984;Courchesne et al. 1985;Courchesne et al. 1989]. Although ERP abnormalities have been observed on many occasions in autism, little is known about the sources of these abnormalities. While an advantage of ERPs is a very high temporal resolution (in the order of milliseconds), the spatial resolution is limited. For accurate localization of the generators in the brain which produce the measured scalp potentials, one needs to sample the scalp with an extensive set of electrodes. Most of the studies to date have only sampled from a very limited number of electrode sites. The data in our recent experiments were sampled from 62 scalp electrodes, which makes localization feasible.

ERPs have the advantage over other imaging methods of having a very high temporal resolution, but lack spatial detail. On the other hand, imaging methods like functional magnetic resonance imaging (fMRI), single photon emission tomography (SPECT) and positron emitting tomography (PET) have better spatial resolution, but their temporal resolution is very limited compared to ERPs. PET and SPECT studies have been carried out in autism in task [Siegel et al. 1995;Buchsbaum et al. 1992;Muller et al. 1999] and non-task conditions [Chiron et al. 1995;Ohnishi et al. 2000;Hashimoto et al. 2000;Haznedar et al. 1997;Horwitz et al. 1988;Mountz et al. 1995;Ryu et al. 1999;Starkstein et al. 2000;Zilbovicius et al. 2000;Zilbovicius et al. 1992;Zilbovicius et al. 1995] and have produced indications of abnormal hemispheric asymmetries in metabolism, delayed frontal maturation and decreased metabolism in frontal, temporal and parietal areas as well as in subcortical structures. fMRI studies using face processing tasks [Pierce et al. 2001;Schultz et al. 2000], the Embedded Figures task [Ring et al. 1999] and a simple motor task [Muller et al. 2001] suggest that autistic subjects activate different brain areas than controls under identical controlled task conditions. Since altered patterns of activity are observed in non-task and very simple task conditions [Muller et al. 2001], cognitive operations are not a necessary condition to evoke abnormal activation patterns in autism. Taken together, functional imaging studies suggest two ways in which the brain may be organized differently in autism. The first is a global decrease in the level of cerebral activation, as evidenced by the studies using a resting condition. Second, studies using task conditions indicate that autistic individuals may not activate areas associated with normal task performance. Similarly, the abnormal ERP amplitudes as measured on the scalp could be the result of differences in the locations of the generating sources. Another possibility is that the source locations are equal for autistic subjects and controls, but that these are less active in autism (i.e., have a lower source strength). These questions will be addressed in the present paper.

To answer these questions, we employed high resolution MRI-based source localization techniques to characterize the sources of the P1 and P3 components stemming from a visual selective attention task [Hoeksma et al, submitted]. In these data, both P1 and

P3 were markedly reduced in amplitude in autistic subjects, but this amplitude reduction was not related to effects of selective attention. The marked amplitude reduction found for P1 indicates that functional abnormalities are present at a very early processing stage in autism. The temporal resolution of functional imaging modalities other than ERPs may be too low to localize such early abnormalities.

To our knowledge, the present study is the first to employ source localization techniques in autism. Source localization greatly enhances the spatial detail of ERPs, since conductivities of the head are accounted for in a head model, modelling the effects of volume conduction. Traditionally, these techniques modelled the head as a layered sphere. Here we use accurate, realistic Boundary Element models (BEM) of individual MR images. Not only will this improve the accuracy of localizations over traditional techniques, it also provides a better link to the individual brain anatomy. Furthermore, the high temporal resolution of ERPs is preserved using this method. A closely matching BEM model of the cortical surface further enables us to apply Cortical Current Density (CSD) reconstructions to the measured data. CSD is the method of choice for modelling large, complex potential fields with little a-priori information about the location of sources, like the P3.

*Table 5-1: Age and IQ data of the final groups used in the source analyses of P1 and P3. Ctrl = control group; Aut = autistic group; VIQ = verbal IQ; PIQ = performance IQ; TIQ = total IQ. Standard deviations in parentheses. Sample sizes (N) for P1 and P3 differ within diagnostic groups because subjects with inadequate dipole fits were excluded. *Autistic < Control (F(1,20)>9,9; p<0.005). †Autistic < Control (F(1,20)>10,0; p<0.005).*

P1	**N**	**AGE**	**VIQ**	**PIQ**	**TIQ**
Young ctrl	11	10.5 (1.08)	94.2 (9.8)	105.9 (9.8)	99.6 (8.8)
Young aut	10	10.6 (1.56)	101.5 (14.4)	105.1 (16.7)	103.1 (9.8)
Adol ctrl	8	18.2 (0.73)	100.1 (7.4)	112.3 (10.1)	105.8 (7.1)
Adol aut	7	18.6 (1.75)	95.9 (11.9)	103.6 (12.9)	98.0 (11.6)
P3					
Young ctrl	12	10.4 (1.13)	93.4 (9.3)	102.6 (13.4)	97.3 (9.9)
Young aut	10	10.6 (1.56)	101.5 (14.4)	105.1 (16.7)	103.1 (9.8)
Adol ctrl	13	18.2 (0.74)	103.6 (8.0)	115.7 (9.8)	109.5 (8.2)
Adol aut	9	19.9 (2.9)	95.4 (11.3)	99.7 (14.2)†	96.1 (11.7)*

Methods

Task

The visual selective attention task consisted of 300 stimuli, 150 red and 150 yellow rectangles subtending a length of 4.5 degrees of arc and a width of 3.7 degrees of arc. Stimulus duration was 50 ms, inter stimulus intervals (ISIs) were randomized between 1750 and 2150 ms. Between stimuli, a fixation cross was presented in the centre of the screen. Total task duration was about 10 minutes. Relevant and irrelevant stimuli were defined by stimulus colour (i.e., yellow or red). Standards and deviants were defined by the orientation (to upper left \\ or upper right //) of thin, black diagonal bars superimposed on the rectangles. Within each colour, 20% were deviant and 80% were standard stimuli. Which orientation was standard or deviant was balanced across subjects, as was the relevant colour. Stimuli were presented in the centre of a computer monitor positioned approximately 70 cm from the subject's eyes. The subject was required to press a button, held in the preferred hand, as fast as possible whenever a rectangle of the relevant colour was presented in which the orientation of the bars was deviant.

Subjects

Autistic children and adolescents and IQ and age matched controls participated in the study. Autistic children were patients of the Department of Child and Adolescent Psychiatry of the Utrecht Academic Hospital. Control children came from elementary schools in and around Utrecht. Autistic adolescents were recruited from a residential institution for autistic patients (the dr. Leo Kanner house). Control subjects were recruited from a secondary school in Utrecht. All subjects were free of medication and had no significant neurological history. All clinical subjects were administered the Autism Diagnostic Interview Revised (ADI-R) [Lord et al. 1994]. All adolescent patients met all ADI-R criteria for autism. Five children did not meet all cutoff criteria for autism, but they did however meet criteria for PDD-NOS as indicated by a psychiatrist. For children, the initial samples consisted of 25 autistic subjects and 19 controls. Initial adolescent groups consisted of 13 subjects in both clinical and control groups. Subjects with large numbers of incorrect responses and omissions were excluded from the samples, as were subjects of whom no MRI data were available. Details on the final groups are presented in table 5-1. None of the controls or their family members had a significant history of neurological or psychiatric problems. Furthermore, all participants showed normal brain MRI as indicated by a radiologist. The study was approved by the medical ethical committee of the Utrecht Academic Hospital and all participants or their parents or caretakers gave prior written informed consent.

EEG recording and signal analysis

While subjects were performing the task, electroencephalic activity was measured from 62 tin electrodes placed on the scalp by means of an electrocap. A reference electrode was attached to the left mastoid, and a ground electrode was placed in the middle of the forehead. Impedances of ground and reference electrodes were kept below 5kOhms.

Horizontal EOG was recorded from electrodes attached to the outer canthus of each eye. Vertical EOG was measured from infra- and supraorbitally placed electrodes at the left eye. All signals were amplified with a time constant of 10 s by Sensorium EPA-5 amplifiers, digitized on-line by a computer at 256 Hz and stored as a continuous signal. Signals were epoched off-line starting 100 ms before stimulus onset and lasting for 1 second. Epochs were filtered with a 30 Hz, 24 dB/octave digital low pass filter and baseline corrected on the basis of the 100 ms pre-stimulus interval. Epochs containing artifacts like flat lines, saturation of the A/D converter and amplitudes larger than +/- 125 μV were removed. EOG artifacts were removed from the EEG by subtracting vertical and horizontal EOG from the EEG epochs by a regression method in the time domain [Kenemans et al. 1991]. ERPs were computed by averaging all remaining trials with correct performance for each subject in four stimulus categories (attended deviants and standards and unattended deviants and standards) per lead. For localizations of the P1, the ERPs to unattended stimuli were averaged for each subject.

MRI

After completion of the task, electrode positions and five fiducial marker positions (nasion, left and right pre-auricular points and mastoids) were digitized by means of a Polhemus Isotrak digitizer for coregistration with individual MRI scans. MR images were acquired on a 1.5 Tesla scanner (Philips Gyroscan NT). Three dimensional T1-weighted, coronal spoiled gradient echo (FFE) scans of the whole head were obtained (TE = 4.6ms, TR = 30 ms, flip angle 30°, contiguous slices. 1x1x1.2mm voxels for adolescents, 1x1x1.5mm voxels for children). Oil-filled capsules were placed on the digitized marker positions to make them clearly visible in the MR images.

All MR scans were resampled to isotropic volumes with 256 1x1x1mm voxels in every direction and automatically corrected for intensity nonuniformity [Sled et al. 1998]. All scans were registered to standardized space by an automated procedure [Collins et al. 1994]. The individual registration parameters from this procedure were used to convert dipole locations to standardized space for statistical analyses.

Source localizations

All source localizations were done with the Curry program (Neuroscan inc.). Individual realistic boundary element (BEM) models of the head were constructed by an automated procedure. The unregistered (i.e., non-normalized) scans were used for this procedure. Using the scaled scans for dipole localizations would result in a localization error, since the scalp potentials are not scaled accordingly. Therefore, we chose to use the unregistered scans for localization purposes and to convert the resulting dipole localizations to standardized space afterwards. For each subject, the digitized electrode cloud was fitted to the head model through manual identification of the marker positions visible in the MR images, which corresponded to the digitized fiducial marker positions.

Localizations of P1 were based on the averaged ERPs for unattended stimuli, localizations of P3 were done on ERPs for attended deviants in order to have maximal signal

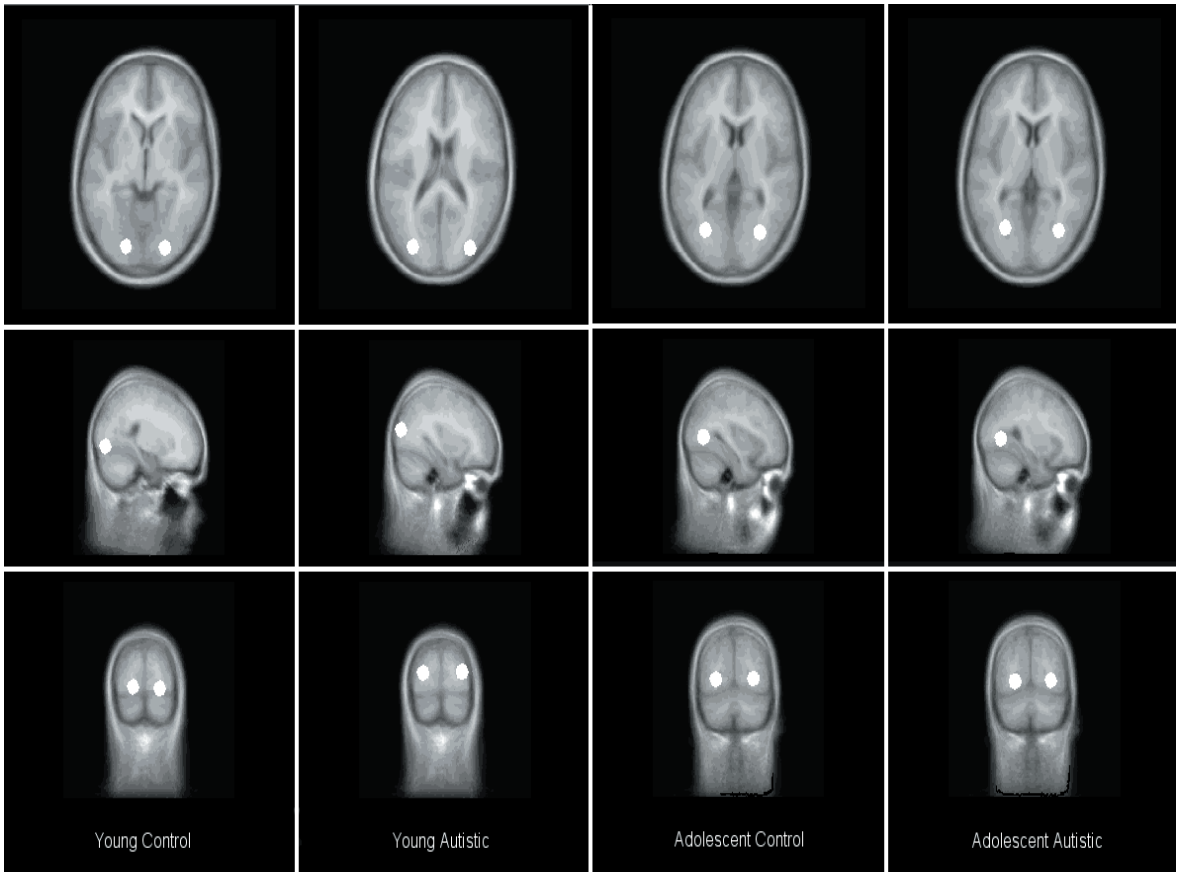
to noise ratios. For the purpose of source localization, all leads were re-referenced to a common average reference. P1 was modelled with a mirrored fixed dipole pair.

Initial attempts to characterize P3 by two or three sources did not produce satisfactory results. These solutions were unstable and had very high residual variances. Therefore, the single dipole approach was chosen for P3. This approach is not likely to produce a physiologically plausible location for the P3 generator, but it is rather a form of data reduction to quantify the entire scalp recorded potential field. Thus, any significant differences in dipole P3 location between groups could be interpreted as an indication of qualitative differences measured scalp potentials.

For P1 and P3, an initial dipole solution was produced based on the grand average ERPs for each group on an averaged head model (n=50). For P1, the timepoint for this solution was chosen as a 25 ms window around the peak of the global field power in the P1 latency (approximately 100-125ms). For P3, the reconstruction window was set as 50 ms around the maximum global field power in the 350-650 ms timerange.

The grand average solutions for P1 and P3 were entered as start locations for individual fits. First, a fit was performed with the fixed grand average dipole locations to

Figure 5-1: Averaged dipole locations for P1 projected on an averaged brain (n=50) in axial, sagittal and coronal directions. In the autistic group, dipoles were located significantly more dorsal and had a tendency to have a more lateral location. There were no significant differences in dipole parameters in the adolescent groups.



compute the explained variance of this source configuration. For individual fits, the timepoint for final reconstructions were chosen similar to the grand average fit described above, but now the point where the explained variance was maximal was taken in stead of the maximum global field power. Finally, all source parameters were optimized.

The locations of the dipoles were transformed with the registration parameters resulting from the MR registration in order to register them into a standardized coordinate system, enabling between subjects comparisons. No transformations were done on dipole orientations, since they were not affected by the registration procedure.

In order to gain insight into the cortical locations of P3 generators, cortical CSD maps were calculated for visual inspection. For comparison with the computed dipole locations, CSD maps were also computed for P1. CSD reconstructions (minimum norm, chi-square regularization) on grand-average data were done on an averaged brain (n=50), with 9864 sources with fixed orientations, placed on the cortex using surface normals.

Statistical analyses

Separate ANOVAs for dipoles were done for each of the three location (xyz) and orientation (xyz) parameters and mean dipole strengths. Since we expected a priori differences in dipole locations between young and adolescent groups e.g. [Friedman et al. 1997], we performed separate analyses for the two age groups.

Figure 5-2: Averaged dipole locations for P3 projected on an averaged brain (n=50) in axial, sagittal and coronal directions. No significant differences were present in the location parameters.

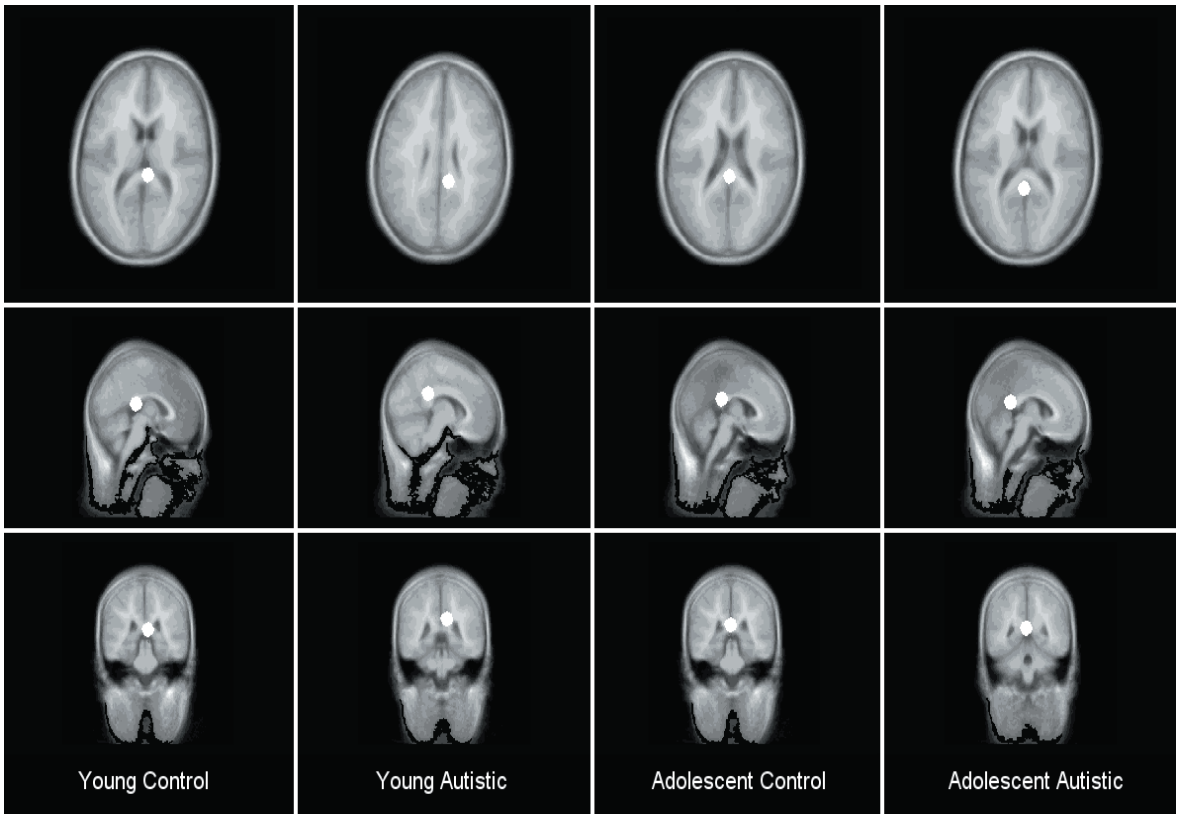
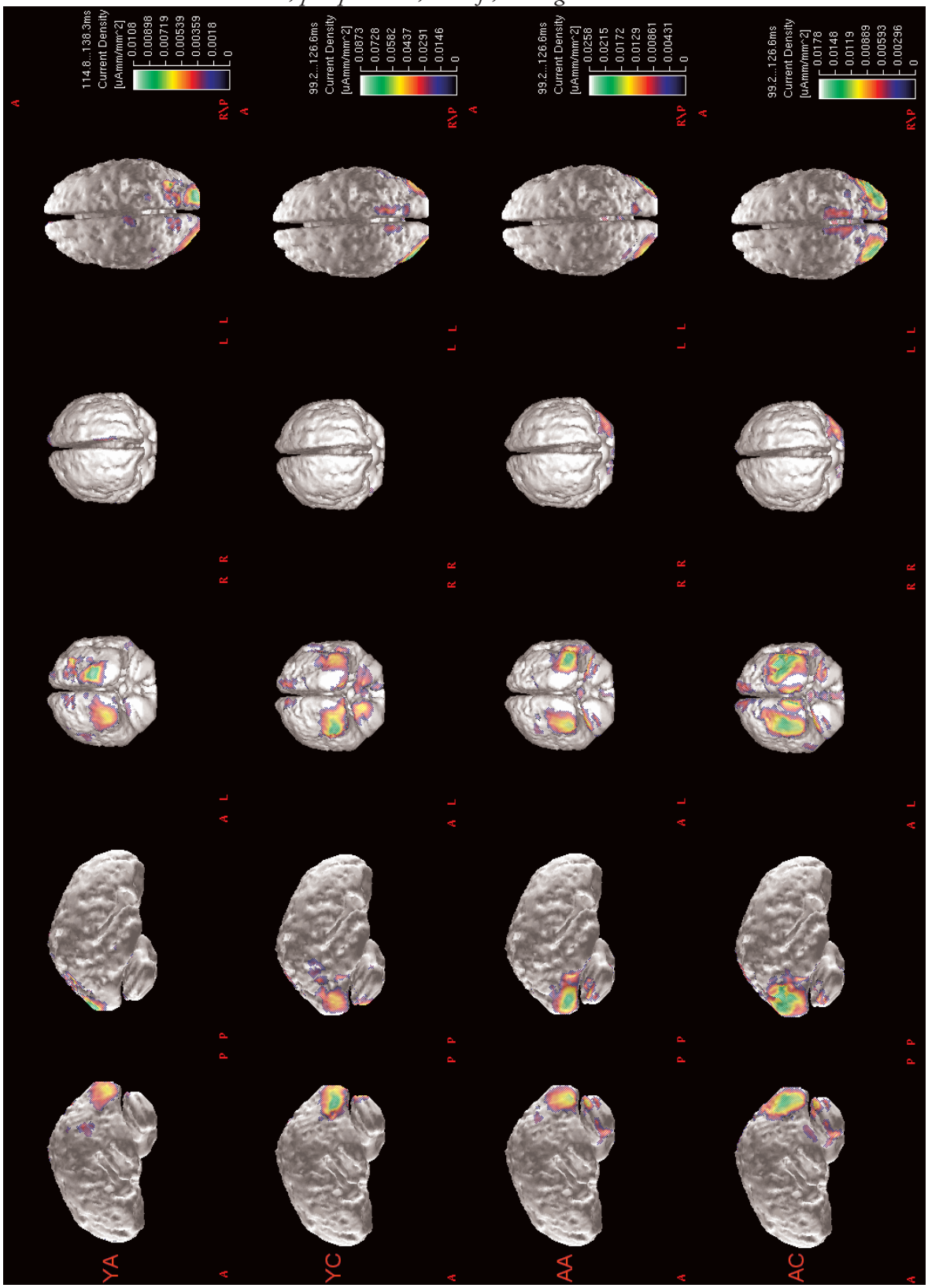


Figure 5-3: Grand average CSD maps for P1. The loci of activation correspond closely to the dipole locations (fig. 5-1). YA = young autistic; YC = young control; AA = adolescent autistic; AC = adolescent control. a = anterior; p = posterior; l = left; r = right.



Results

P1 dipoles

Results of the P1 localization are shown in figure 5-1. In the young groups, autistic subjects showed a significant difference in z-location for both the left ($F(1,19) = 5.609$, $p = 0.029$) and right ($F(1,19) = 6.902$, $p = 0.017$) dipoles, indicating that dipoles in the autistic group were located superiorly to those in the control group. No further significant effects were present.

For adolescent groups, no significant effects on location or dipole strength were present in the data. There was a difference in orientation, as indicated by an effect for the y-orientation ($F(1,13) = 5.081$, $p = 0.042$) (autistic 0.8329 vs 0.6763 for controls).

P3 dipoles

The dipoles for the P3 localization are shown in figure 5-2. Young autistic subjects showed a significantly weaker dipole strength than their controls ($F(1,20) = 4.513$, $p = 0.046$). No further effects were present in the data ($0.4 < F(1,20) > 2.11$; $0.162 < p > 0.534$). In the adolescent groups, no differences were noted for the P3 parameters ($0.15 < F(1,20) > 1.546$; $0.171 < p > 0.902$).

Quality of fit

To check for differences in the quality of fit between autistic and control subjects, the residual variances of P1 and P3 fits were tested per age group. No significant differences were noted (all $F < 1$).

CSD reconstructions

The CSD reconstruction of P1 (fig. 5-3) resulted in a clear bilateral occipital focus for all groups. The young autistic group showed a right sided activation that was located superior to that in controls, which agrees with the differences in dipole locations. Autistic adolescents show bilateral occipital foci; the adolescent controls show additional medial activations, which are not seen in autistic adolescents.

For P3, the CSD maps are much more complex (fig. 5-4). Especially in the young groups, the CSD localizations are suggestive of many sources in medial and central parietal, occipitoparietal, as well as bilateral frontal areas. In contrast to the young control group, the young autistic subjects show no activation in the occipital lobes but a more central parietal focus. Also, the frontal activity seems to be more lateral in the autistic subjects.

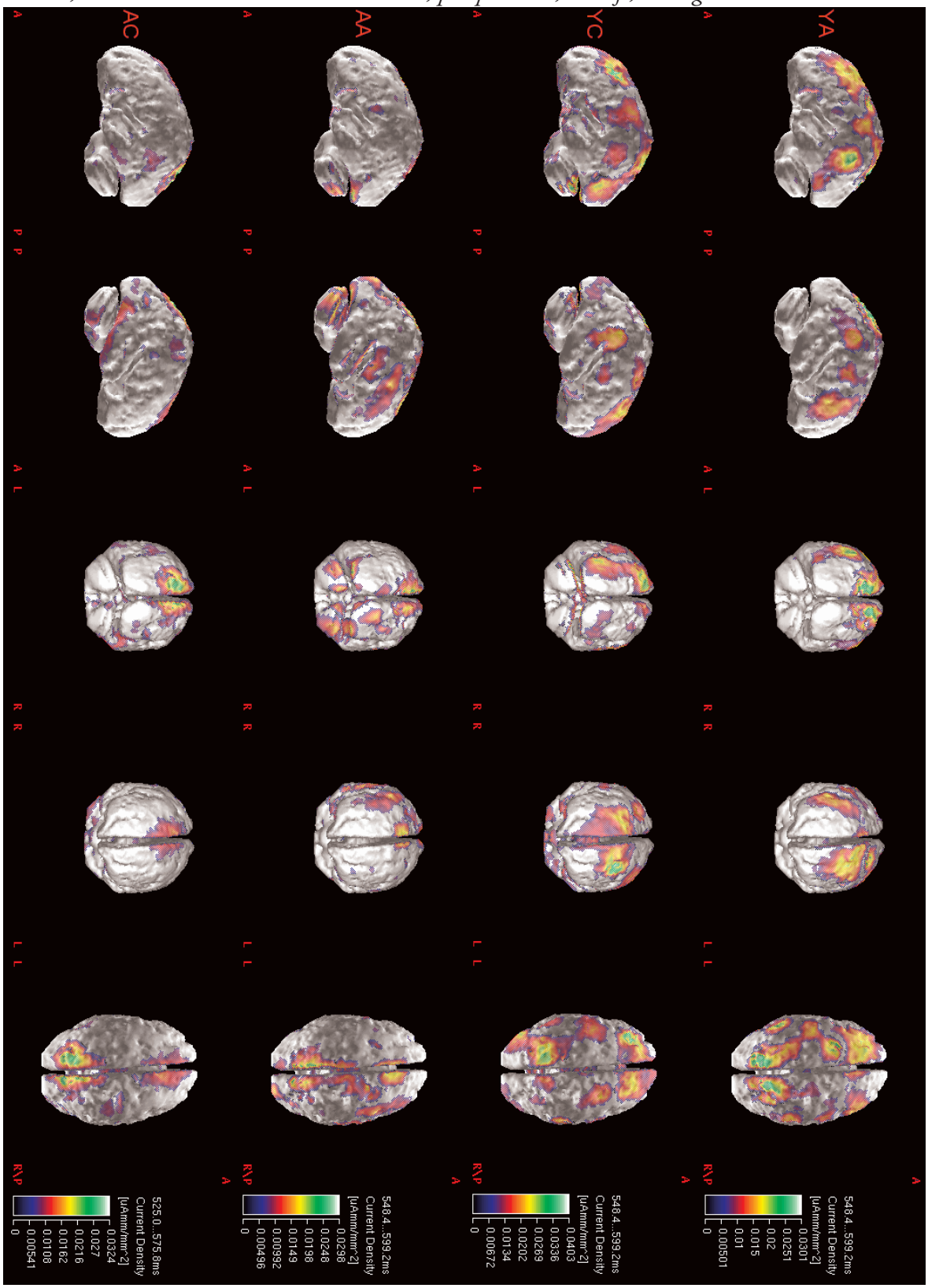
In the adolescent control group, the P3 is characterized as central parietal and frontal foci. These areas also seem to be activated in the adolescent autistic group, but many more areas seem to be implicated, especially in the occipital and right lateral frontal areas.

Discussion

The present study sought to identify differences in the generators of visual P1 and P3 in autistic children and adolescents, compared to control groups. ERPs were measured in a visual selective attention task, of which the data are described in detail elsewhere [Hoeksma et al, submitted]. The main effects in those data were a marked decrease in P1 and P3 amplitude in autistic children. These diagnosis effects were not related to selective attention (i.e., did not interact with the attention manipulation). Here, we wanted to study whether these amplitude abnormalities were associated with differences in the generating sources of these peaks. In principle, the amplitude abnormalities in autism could be the result of different locations of the generating sources. However, another possibility is that the locations of the generators are normal, but that these generators produce less energy (i.e., have a smaller dipole strength) compared to controls. Furthermore, we wanted to investigate whether the absence of amplitude abnormalities in autistic adolescents was coupled with normal cortical generators. To this end, we performed high resolution source analyses based on realistic individual head models. Dipole models for P1 were chosen as mirrored, bilateral dipole pairs. The sensory evoked P1 has been localized by means of electrophysiology and PET in extrastriate cortex, most likely the fusiform gyrus [Heinze et al. 1998;Ossenblok et al. 1994]. When present averaged dipole locations were transformed to Talairach coordinates [Talairach and Tournoux 1988], the locations of the P1 dipoles in young controls could be localized in the lingual gyrus, whereas the dipoles in the autistic group were located in the middle occipital gyrus. Although the Talairach coordinate system is based on the adult brain, transformations of child images to Talairach space results in only small locations errors (<5mm), which seem negligible compared to the resolution of source localizations (~10mm) [Burgund et al. 2001]. The lingual gyrus has been associated with discrimination of colour and spatial frequency [Corbetta et al. 1991] or object identity [Kohler et al. 1998], whereas the middle occipital gyrus has been implicated in perception of form and location [Braddick et al. 2000] [Kohler et al. 1998]. Thus, it seems like autistic children use areas of the visual system that are spatially and functionally distinct from those used by controls.

Similar observations have been made in fMRI experiments with face processing, in which autistic subjects showed significantly less activation of ventral temporal areas (in particular the fusiform gyrus) [Pierce et al. 2001;Schultz et al. 2000]. Although these studies relate their findings exclusively to face processing, our data suggest that abnormal cortical activations related to more general visual processing seem to occur already at a very early stage, as early as 100 ms after a stimulus is presented. It could be that the time resolution of fMRI is too low to detect such early abnormalities, since Schultz and colleagues did not find any abnormalities with object processing [Schultz et al. 2000]. It should be noted that recent fMRI studies indicate that activation patterns in autism show a very high individual variability [Pierce et al. 2001;Muller et al. 2001], and as such the averaged dipole locations of P1 in the current study may be an oversimplification of the true location of activity. However, inferring that autistic children activate different brain areas than controls in the present task is still justified.

Figure 5-4: Grand average CSD maps for P3. autistic subjects seem to exhibit a more complex pattern of activation than controls. YA = young autistic; YC = young control; AA = adolescent autistic; AC = adolescent control. a = anterior; p = posterior; l = left; r = right.



In autistic adolescents, no differences were observed in the locations of the P1 generators. Thus, the apparent normalization of the P1 abnormality in autistic adolescents seems to be paralleled by normal underlying generators in the brain. The CSD maps of the P1 were in close agreement with the dipole solutions, and indicate that our choice of two mirrored dipoles for P1 was appropriate. With respect to the locations of P1, it can be concluded that autistic children activate different areas of the visual system than their controls. Furthermore, the absence of any effects on dipole parameters in adolescent groups parallels the apparent normalization of P1 in autistic adolescents.

With regard to the P3, our localization approach does not justify any inferences on the anatomical location of the sources. The dipoles must only be viewed as a form of data reduction. Still, such data reduction has to be interpreted with caution, since smeared potential fields, possibly different numbers of sources [Goto et al. 1996], as well as measurement noise are all incorporated in the dipole parameters of one source. Bearing this in mind, no differences were seen in the locations or orientations of the P3 dipoles. However, the young autistic groups showed a significantly smaller dipole strength, which is in line with the smaller P3 amplitudes observed in the ERPs. The CSD maps of the P3 indicate that especially in children, the P3 is generated by widely distributed brain areas. In adolescent subjects, the pattern is less complex. However, in both age groups, autistic subjects seem to show a more diverse pattern of activation than controls. The complexity of the foci of activation indicates that it would be very difficult to characterize the P3 with a discrete number of dipoles, and that more dipoles would be necessary to characterize the P3 in autism than in controls. The more complex patterns of activation in autism observed here may be in accord with more widespread and variable activation as seen in fMRI studies [Pierce et al. 2001;Muller et al. 2001].

We wanted to investigate whether profound amplitude abnormalities of P1 and P3 in autism could be related to different neural generators. To this end, we applied high-resolution source localization techniques to ERP data from a visual task. This resulted in different locations for the generators of P1 in the young autistic group, which could be a consequence of a different organization of the brain. CSD maps of P1 reflected a similar effect. CSD maps of P3 were suggestive of more a more complex pattern of cortical generators in autism compared to controls. The high-resolution source localization techniques used in this study seem to add to the interpretation and understanding of psychophysiological abnormalities in autism. Future quantifications of CSD maps to allow for statistical group comparisons will further enhance the usability of this approach.

Acknowledgements

The authors wish to express their gratitude to Rene Mandl and Saskia Palmen for their invaluable assistance with the MR data.

Chapter Six

Summary and conclusions

The investigations in this thesis were motivated by the often reported abnormalities in P3 amplitude in autism. Smaller P3 amplitudes in autistic groups have been fairly consistently reported in the autism literature. Not only have such abnormalities been demonstrated in different modalities, but also in different age groups. No study however has addressed the question of any age related differences directly. Therefore, the studies described in this thesis have included autistic children and adolescents. Aiming at an in-depth analysis of the abnormal P3 responses in autism, an attempt was made to answer the following questions:

Is the abnormal P3 preceded by, or related to, deficiencies in selective attention?

The studies of visual and auditory selective attention (chapters 2 and 3) investigated whether abnormal P3 responses were caused by preceding abnormalities in selective attention. A number of oddball studies have reported smaller P3 amplitudes in autism. However, none of these have found abnormalities in earlier aspects of attention. To date, only one ERP study of selective attention [Ciesielski et al. 1990] has been reported in the literature on autism that has shown that there may indeed be abnormalities in these earlier attentional components.

Is the abnormal P3 amplitude a reflection of abnormal processing capacity?

Chapter 4 dealt with a study of processing capacity. The inclusion of this study was motivated by a finding of Kemner et al [Kemner et al. 1995]. In an auditory oddball study, these authors found larger P3 amplitudes over the occipital scalp in autistic children. In an earlier study, these authors reported smaller P3 amplitudes in autistic children over occipital areas [Kemner et al. 1994]. Based on the findings from these studies, the authors then suggested that the larger P3 in the auditory task over visual areas could be the result of the auditory system 'borrowing' processing capacity from the visual system. In dual-tasks and tasks using irrelevant probes, the P3 to secondary task stimuli or -probes is sensitive to the amount of processing capacity allocated to the primary task. By using a combination of a primary auditory task and visual probes, the study described in chapter 4 investigated how auditory and visual processing capacity are intertwined in autism.

Which areas of the brain are implicated in abnormal ERP responses?

Chapter 5 described the localization of the visual ERPs from chapter 2, in particular the abnormally small P1 and P3 waves in autistic individuals. Up to now, only limited electrode configurations have been used in ERP studies in autism. Therefore, source localization studies have never before been reported. Using advanced localization techniques based on MRI, chapter 5 attempted to clarify where the neural sources of these ERP abnormalities in autism originate in the brain.

Visual and auditory selective attention

In a visual selective attention task (chapter 2), the autistic groups showed smaller P3 amplitudes than controls on electrode Oz and Pz. Although the reduction of P3 was significant for both autistic age groups, the autistic children especially showed a dramatically small P3 amplitude. Also, in autistic children the smaller occipital P3 was preceded by a smaller P1. Both effects, the smaller P3 in autistic groups and the smaller P1 in autistic children, were not related to selective attention. That is, amplitudes were smaller to all stimuli, regardless whether they were attended or not. No defects in selective attention preceding P3 were observed in the young autistic subjects, who showed the most marked P3 reduction. In the adolescent autistic group however, N2b was found to be larger than in the young autistic group. A similar enhancement of N2b amplitude with age was not seen in the control groups. Thus, the adolescent autistic group showed larger N2b than autistic children and improved P3 amplitude compared to the latter group. These results suggest that there may be a relationship between P3 abnormalities and selective attention nonetheless. The enhanced N2b in autistic adolescents could be a reflection of a compensatory process, normalizing the P3.

Similar results were found in the auditory selective attention study (chapter 3). The autistic groups showed an abnormal P3 response, in that they showed no significant differentiation in P3 amplitude between attended and unattended deviants. In the control groups, the attended deviants evoked a larger P3 than unattended deviants. No abnormalities in selective attention were present in the youngest group. Strikingly, the adolescent autistic subjects showed an earlier and broader frontal PN than their healthy controls, a finding that seems in line with the finding of a larger N2b in the visual task. Moreover, normalization of P3 amplitudes in the adolescent autistic group was paired with an abnormally large PN. Therefore, also in the auditory task abnormal P3 seems to be preceded by normal selective attention and vice versa.

The results from the selective attention studies prove three things. First, the expected relationship of abnormal P3 being preceded by abnormal selective attention does not hold. The largest P3 abnormalities were seen in the young autistic group, but there was no effect of selective attention on P3 amplitudes (that is, P3 amplitudes were overall smaller), nor were they preceded by abnormalities in selective attention. On the other hand, adolescent autistic individuals did show abnormalities in selective attention, as evident in enhanced PN and N2b, but largely normal P3 amplitudes.

Second, the findings from these studies seem to contrast with previous reports of abnormal P3 in autism. The reduction of occipital and parietal P3 in the young autistic group is in line with previous findings [Verbaten et al. 1991;Kemner et al. 1994], but the largely normal P3 amplitudes in adolescent autistic persons in this study do not parallel previous reports (e.g. Ciesielski et al. 1990). Also, the finding of enhanced N2b and PN in this thesis contrasts with the findings of Ciesielski and others, who found an absence of attention related negativity in autistic individuals in visual and auditory selec-

tive attention [Ciesielski et al. 1990]. The differences in experimental setup between the current study and the study by Ciesielski et al may explain this discrepancy in results. Especially the simultaneous presentation of auditory and visual stimuli in the study by Ciesielski et al is a major difference with the current studies. Also Ciesielski and colleagues compared stimuli from different experimental blocks for the effect of selective attention. In the present studies, attended and unattended stimuli were presented in the same stimulus block and in only the visual or auditory modality. Therefore, any unwanted cross-modal effects that might be present in the data from Ciesielski et al. are removed from the present data. Also, measuring attended and unattended stimuli in the same experimental block controls for any possible differences in attentional state between sessions.

Third, extending these previous remarks, there seem to be differences in aspects of selective attention and P3 between autistic individuals of different age groups. Abnormal P3 amplitudes in young autistic individuals seem to normalize with age, but this seems to go together with the emergence of abnormal selective attention, in the form of enhanced attention-related negativities. It could be that abnormal attention in the adolescent autistic group acts as a compensation for the abnormal P3. It should be noted that both in the young and adolescent groups, the behavioural performance on the selective attention task is comparable to controls. However, the adolescent groups were significantly more accurate and faster in their responses than the young groups. It might be that autistic adolescents can only attain such a level of performance by means of enhanced selective attention mechanisms. Such a compensatory mechanism might be a correlate of the more general behavioural improvement which may be observed over time in autism [Piven et al. 1996b]. In a recent MRI study of brain growth in autism, Courchesne and colleagues found that brain development in autism is characterized by an early overgrowth (i.e., autistic children have larger brains than their healthy controls) followed by a developmental arrest in later childhood and adolescence (i.e., autistic adolescents show brain volumes comparable to those of controls) [Courchesne et al. 2001]. It might be that the ERP differences between the autistic age groups in this thesis are a functional reflection of these stages of brain growth.

Processing capacity

Attention and processing capacity (as indexed by P3) are closely related. Attention serves as a filter to prevent the information processing system from overload. It can not be directed simultaneously to all stimuli in the environment or, to be more specific, processing capacity can not be allocated in an unlimited fashion. Attention can be seen as a 'gatekeeper', giving some streams of information a higher priority than others. There is a dependency between streams of information that need simultaneous processing. Extra capacity that is needed for the processing of one stream of information (or task) is borrowed or taken at the expense of the second stream. Such a dependency can be visualized in the P3 by means of a probetask. When the difficulty, and thus the processing

priority of the primary task is increased, leading to increased P3, it will go together with a decrease in available processing capacity for the secondary (probe) task, and a subsequent reduction of P3 amplitude for this task. The probe stimuli thus literally probe the amount of spare processing capacity that is left by the primary task.

The probe task described in chapter 4 was set up as an auditory primary task with visual probes, in order to test the suggestion by Kemner et al [Kemner et al. 1995], who argued that in autism processing capacity from the visual system might be relocated to the auditory system. An important argument for this reasoning was a finding of larger occipital P3 amplitudes in an auditory task in autistic children [Kemner et al. 1995]. However, the autistic groups showed normal auditory P3 amplitudes and normal increase of auditory P3 with increased load of the primary task. Based on the suggestion by Kemner and colleagues, one would expect a disproportionate decrease of visual P3 amplitude with increasing auditory P3, but no evidence was found for such an effect either. Taken together, no evidence was found for a relocation of visual processing capacity to the auditory system.

On the other hand, in the normal groups increased auditory P3 amplitude was paired with a concurrent decrease of P3 to visual probes, but this effect was not found in the autistic groups. The absence of a load-dependent decrease in probe P3 in autism does point to a deficiency in capacity allocation. It suggests that autistic individuals process all incoming information with the same high priority without prioritizing one stream of information above the other.

Another interesting point is that in the probetask, young autistic individuals showed smaller occipital P3 amplitudes in the easy condition only. When task difficulty was increased, P3 amplitudes of these subjects were comparable to the control group. This suggests that the available capacity pool in the young autistic group is not fully utilized in the easy task, and is fully taxed only with higher load. This phenomenon stands in stark contrast with the normal decrease of P3 amplitude for visual probes with increased load in the control group. This finding also sheds a different light on the smaller P3 amplitudes in the visual selective attention task. Apparently, the visual system in young autistic individuals has the capacity to produce a P3 that is comparable to controls, but does so only under special circumstances. The choice of experimental tasks therefore seems to be a critical point in ERP studies in autism.

Source localization

The results from the selective attention tasks and the processing capacity task indicated that autistic individuals show specific abnormalities in ERP amplitudes. These amplitude differences were largest in children and mostly independent of manipulations of attention. The most dramatic abnormalities were seen in the visual selective attention task (chapter 2), where autistic children showed markedly smaller P1 and P3 amplitudes. Autistic adolescents did not show these abnormalities. Chapter 5 tried to answer the question of whether these abnormalities are related to different locations or configura-

tions of underlying electrical sources of P1 and P3. In order to answer these questions, sophisticated source localization techniques were used. With the aid of structural MRI, detailed individual head models were constructed which were used in the localization of the generators of P1 and P3.

Dipole localizations of P1 showed that autistic children had a more dorsal locus of its underlying sources than controls. No such difference was found in the adolescent groups. The results in the young autistic group resemble the findings of fMRI studies showing abnormal ventral occipital activations related to face processing in autistic individuals [Pierce et al. 2001;Schultz et al. 2000]. Surely, the visual selective attention task used in this thesis is very different from face processing. However, it can be argued that both tasks depend on the analysis of visual features and conjunctions. Given this mechanism that the tasks may share, it still remains puzzling why only the young autistic groups show a different location for the sources of P1, since the mentioned fMRI studies were conducted in adults. As noted earlier, the choice of tasks may be very critical in ERP studies in autism.

Although P3 amplitude was largely abnormal in autistic children, no differences with normal controls were found in the locations of P3 for the young autistic group, nor in the adolescent autistic groups. The young autistic group did show a weaker dipole strength than their controls, however. In the probe task, it was found that the young autistic group showed smaller P3 amplitudes in the easy condition only. When task load was increased, P3 amplitudes were comparable to the control group. The finding of a P3 source in autistic children that is comparable to controls with respect to location, but shows a lower level of activation seems in line with this observation. Possibly, the P3 generator in itself is largely normal in autistic children, but apparently it is not functioning at full capacity. Since the P3 is a very broad potential with a widespread topography, the results of the dipole analyses which were conducted with a single source must be interpreted with caution, however. P3 is likely to be produced by multiple generators [Picton 1992], and therefore the reconstruction of P3 with one dipole is very likely to be an oversimplification. An indication of the complexity of the generators of P3 is given by the CSD reconstructions presented in chapter 5. CSD reconstructions have the advantage over dipole estimation that they do not require a-priori estimations of the number of sources producing the ERP that is under analysis. Indeed, the CSD reconstructions were suggestive of multiple P3 generators. Furthermore, the picture proved to be more complex in the autistic groups, with multiple scattered regions of activity. CSD reconstructions are dependent on a good signal-to-noise ratio (SNR), and the observed differences between autistic and control groups might be related to differences in SNR. However, the true value of CSD comparisons as presented here can only be appreciated when appropriate methods for statistical group comparisons become available. Recently, such methods are beginning to emerge.

Although quite a number of ERP studies have been conducted in autistic individuals, none have ever localized the sources of resulting abnormal ERPs. The study presented in chapter 5 is the first to make such an attempt and should be interpreted in

this manner. Electrophysiological localization studies in autism open interesting new perspectives on the neurophysiological origins of the disorder. Future localization studies could benefit from a combination of functional MRI and electrophysiological localizations, thus pairing the high spatial accuracy of fMRI with the temporal resolution of ERPs. Such combinations should be able to pinpoint functional abnormalities with greater accuracy and confidence. Furthermore, information on abnormal brain anatomy from structural MRI could help in the interpretation of ERP results and could guide ERP researchers in devising experiments that are especially geared at tapping the functional correlates of such structural abnormalities.

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Samenvatting

Autisme is een ernstige ontwikkelingsstoornis die zich in de eerste drie levensjaren manifesteert. De belangrijkste afwijkingen in het gedrag zijn te vinden in de taalontwikkeling, communicatie en sociale interacties. Verder vertonen patienten met autisme vaak stereotype gedragingen en vernauwde interesses. Autisme komt voor bij ongeveer 5.5 op de 10000 mensen, en ongeveer vier keer zo vaak bij jongens als bij meisjes.

Hoewel autisme vanwege het ontstaan op vroege leeftijd al vroeg werd gezien als een stoornis met een neuropathologische achtergrond, verschoof de aandacht in de jaren vijftig van de vorige eeuw naar de zogenaamde 'koele moeders' als belangrijkste oorzaak. Ouders van autistische kinderen zouden in enige mate dezelfde sociale beperkingen vertonen als hun kinderen. Tegenwoordig heeft onderzoek aangetoond dat autisme wel degelijk een neuropathologische achtergrond heeft, met een sterke genetische basis. Er wordt op dit moment belangrijke vooruitgang geboekt op het gebied van het typeren van de genetische afwijking. De overerfbaarheid van autisme is ongeveer 90%, hetgeen betekent dat er nog ruimte is voor omgevingsfactoren in de vorming van de stoornis.

In dit proefschrift zijn studies beschreven die gebruik maken van Event-related Potentials (ERPs) om de hersenactiviteit van personen met autisme te bestuderen. ERPs zijn weergaven van de elektrische activiteit die de hersenen genereren bij het verwerken van stimuli. Deze elektrische activiteit kan met behulp van electroden op de schedel worden gemeten. Hoewel er andere methoden zijn om hersenactiviteit te meten, zoals functionele Magnetische Resonantie Imaging (fMRI) of Single Photon Emitting Computerized Tomography (SPECT), heeft geen van de andere methoden een oplossend vermogen in de tijd dat vergelijkbaar is met dat van ERPs. Met ERPs is het mogelijk om de verwerking van een stimulus in de hersenen met een resolutie van enkele milliseconden (of minder) te meten. Zulke studies leveren daarmee niet alleen informatie over welke aspecten van informatieverwerking afwijken in autisme, maar ook over wanneer zulke afwijkingen optreden. Wanneer ERP studies worden uitgebreid met geavanceerde localisatiemethoden (zoals in hoofdstuk 5), kunnen ook conclusies worden getrokken over waar zulke afwijkingen in de hersenen kunnen worden aangetroffen. De studies die in dit proefschrift beschreven staan zijn uitgevoerd in twee verschillende leeftijdsgroepen: kinderen op basisschoolleeftijd en adolescenten. Door deze twee groepen in de studies te betrekken is het mogelijk uitspraken te doen over de stabiliteit van de afwijkingen over de leeftijdsgroepen. Tot nu toe zijn er geen studies verschenen die een dergelijke directe vergelijking van leeftijdsgroepen heeft gemaakt.

Theory of Mind, Central Coherence en Executive functions zijn op dit moment de drie belangrijkste cognitieve theorieën die de cognitieve afwijkingen, en indirect de klinische presentatie, van autisme proberen te verklaren. De theorieën zijn vrij uitgebreide raamwerken die vele aspecten van cognitie bestrijken. Een afwijking in aandacht kan een belangrijk aspect zijn dat deze theorieën gemeenschappelijk hebben. Door hun hoge tijdsresolutie zijn ERPs bij uitstek geschikt om aandachtsprocessen te bestuderen. Eer-

dere ERP studies hebben uitgewezen dat autistische patiënten kleinere P3 amplitudes vertonen dan controles. De P3 is een grote positieve golf die ongeveer 300 milliseconden nadat een stimulus is aangeboden optreedt. Zulke kleinere P3 amplitudes zijn in verschillende leeftijdsgroepen aangetoond; autistische volwassenen, adolescenten en kinderen vertonen dit effect. Kleinere P3s zijn bovendien gevonden in de auditieve en visuele modaliteit. De P3 is gerelateerd aan target-verwerking, en representeert daarmee het eindpunt van attentionele verwerking. Vroege aandachtprocessen, zoals het filteren en selecteren van informatie, gaan aan de P3 vooraf.

De studies in hoofdstuk 2 en 3 gaan in op de vraag of de afwijkingen in visuele en auditieve P3 voorafgegaan worden door afwijkingen in vroegere aandachtsprocessen. In de visuele selectieve aandachtstaak (hoofdstuk 2) vertoonde de autistische groep kleinere P3 amplitudes dan controles op parietale en occipitale electroden, achter op het hoofd. Vooral bij de autistische kinderen was dit effect duidelijk. Bovendien werd de afwijking in P3 amplitude bij autistische kinderen voorafgegaan door een verkleinde P1. Beide effecten, de kleinere P3 en P1, waren evenwel onafhankelijk van de manipulatie van aandacht. Hoewel in de groep autistische kinderen de afwijking in P3 amplitude niet voorafgegaan werd door een afwijking ERP pieken die gerelateerd zijn aan selectieve aandacht, was dat wel het geval in de adolescenten groep. Autistische adolescenten vertoonden een grotere N2b dan autistische kinderen, maar een normale P3 amplitude. Een vergelijkbare vergroting van N2b met leeftijd werd niet waargenomen in de controlegroepen. Mogelijk is de vergrote N2b een teken van een compensatoir proces dat de P3 amplitude normaliseert.

Vergelijkbare effecten werden waargenomen in de auditieve selectieve aandachtsstudie in hoofdstuk 3. De autistische groepen vertoonden een abnormale P3 respons; er was geen significante differentiatie in P3 amplitude tussen geattendeerde en niet geattendeerde devianten. In de controlegroepen was de P3 op geattendeerde devianten groter dan op niet geattendeerde devianten. Net als in de visuele aandachtstaak in hoofdstuk 2 vertoonde de groep autistische kinderen geen afwijkingen in selectieve aandacht. De groep autistische adolescenten vertoonde echter een vroegere en bredere Processing Negativity (PN) dan hun controles. Dit effect lijkt vergelijkbaar met de grotere N2b in de visuele taak. Ook in deze taak vertoonde de groep autistische adolescenten normale P3 amplitudes.

Op grond van de gegevens uit de selectieve aandachtstaken kunnen drie dingen worden geconcludeerd. In de eerste plaats lijkt het niet zo te zijn dat abnormale P3 amplitudes voorafgegaan worden door afwijkingen in selectieve aandacht. Ten tweede lijken de gevonden P3 abnormaliteiten slechts gedeeltelijk overeen te komen met eerdere bevindingen. De kleinere occipitale en parietale P3 in de jonge autisten komen overeen met eerdere resultaten, de grotendeels normale P3 amplitudes in adolescente autisten niet. Tenslotte lijkt het normaliseren van P3 amplitude met toenemende leeftijd gepaard te gaan met het ontstaan van afwijkingen in selectieve aandacht, hier gerepresenteerd door vergrote N2b en PN amplitudes.

Aandacht en verwerkingscapaciteit, zoals door de P3 gereflecteerd, zijn nauw met elkaar verbonden. Aandacht werkt als een filter, dat het informatieverwerkingssysteem beschermt tegen overbelasting. Aandacht kan niet simultaan op alle stimuli in de omgeving worden gericht, en meer specifiek kan verwerkingscapaciteit niet ongelimiteerd worden toegewezen. Aandacht kan worden gezien als een soort poortwachter die sommige informatiestromen een hogere prioriteit geeft dan anderen. Er is een afhankelijkheid tussen informatiestromen die simultane verwerking behoeven. Extra capaciteit die nodig is voor het verwerken van de ene informatiestroom (of taak) wordt geleend of afgenomen van de tweede stroom. Zo'n afhankelijkheid kan worden gevisualiseerd met behulp van een probetaak zoals in hoofdstuk 4 staat beschreven. In deze taak werd een auditieve taak aangeboden tegen een achtergrond van visuele stimuli waar de proefpersoon niet op hoefde te reageren, de zogenaamde probes. Wanneer de moeilijkheid van de auditieve taak werd opgevoerd nam de P3 amplitude voor de auditieve stimuli toe in de controle groep, met een gelijktijdige daling van de P3 amplitude op de probes. Een vergelijkbaar effect werd niet gezien in de groepen autistische patiënten. Deze groepen vertoonden geen afname van probe P3 amplitude. Dit resultaat suggereert dat autistische personen alle inkomende informatie met een gelijke prioriteit verweken, zonder dat een informatiestroom een hogere prioriteit krijgt dan de andere.

De probetaak wees verder uit dat de groep autistische kinderen uitsluitend kleinere occipitale P3 amplitudes op probes vertoonde in de makkelijke conditie. Wanneer de taakmoeilijkheid werd verhoogd waren de P3 amplitudes vergelijkbaar met die van de controlegroep. Daarmee lijkt het erop dat de beschikbare hoeveelheid verwerkingscapaciteit in autistische kinderen niet ten volle wordt gebruikt in de makkelijke conditie en dat deze pas volledig wordt aangesproken in de moeilijke conditie. Een dergelijk fenomeen wijkt sterk af van de normale daling van P3 amplitude op probes met stijgende taakmoelijkheid in de controlegroep. Blijkbaar is het visuele systeem in autistische kinderen in staat om P3 amplitudes te produceren die vergelijkbaar zijn met die in controles, maar doet het dat alleen onder speciale omstandigheden. Deze bevinding werpt een interessant licht op de resultaten van de visuele selectieve aandachtstaak in hoofdstuk 2.

De studies in hoofdstuk 2 tot en met 4 probeerden antwoorden te vinden op de vragen welke aspecten van informatieverwerking verstoord zijn in autisme en wanneer deze verstoringen optreden. In hoofdstuk 5 werd een antwoord gezocht op de vraag waar de afwijkingen in visuele informatieverwerking, de kleinere P1 en P3, kunnen worden gelocaliseerd in de hersenen. Om tot een antwoord op die vraag te komen is gebruik gemaakt van geavanceerde localisatietechnieken. Het is mogelijk om met behulp van een fysisch model van het hoofd de bronnen te localiseren van de elektrische signalen zoals die op de schedel zijn gemeten. Voorheen werd voor een dergelijk hoofdmodel vaak een bol gebruikt. In hoofdstuk 5 is gebruikt gemaakt van een techniek waarbij voor elke proefpersoon een individueel realistisch model van het hoofd is gemaakt op basis van MRI beelden. Wanneer de op de schedel gemeten P1 werd gelocaliseerd met behulp van twee puntbronnen (dipolen), bleek dat die bronnen in de autistische kinderen hoger in

de occipitaalkwab lagen dan in controle kinderen. In de oudere groepen werden geen verschillen voor de P1 gevonden.

De localisatie van de P3 met een enkele dipool leverde geen significante verschillen in locatie op. De autistische kinderen vertoonden echter wel een belangrijk zwakkere bronsterkte dan de jonge controles. Hoewel het localiseren an de P3 met een enkele bron waarschijnlijk een oversimplificatie inhoudt, lijkt de zwakkere P3 bron wel overeen te komen met de afwijkingen in P3 amplitude zoals die gevonden zijn in hoofdstuk 2.

Om tot een indicatie te komen van de werkelijke complexiteit van de generatoren die ten grondslag liggen aan de P1 en P3 is in hoofdstuk 5 ook gebruik gemaakt van Cortical Current Source Density (CSD) mapping. Voor het gebruik van deze techniek zijn minder a priori aannames nodig (zoals over de hoeveelheid bronnen) om tot een oplossing te komen. De aldus verkregen oplossingen voor de P1 kwamen grotendeels overeen met de dipool localisaties. De CSD voor de P3 leek inderdaad te wijzen op meerdere bronnen voor de P3, waarbij de complexiteit van de bronnen groter leek in de autistische groepen dan in de controlegroepen. De CSD gegevens kunnen evenwel pas op waarde worden geschat als in de toekomst geschikte methodes beschikbaar komen om passende statistische analyses op deze data te kunnen uitvoeren.

De localisatiestudie in hoofdstuk 5 is de eerste studie van deze soort in autisme. Dergelijke studies kunnen een belangrijke bijdrage leveren aan de zoektocht naar de neurobiologische grondslagen van autisme. Wanneer in de toekomst localisaties van ERPs worden gekoppeld aan fMRI gegevens is het mogelijk om de spatiele accuratesse van fMRI en het superieure temporele oplossend vermogen van ERPs optimaal te gebruiken. Ook het koppelen van ERPs aan structurele informatie uit MRI, zoals over locale afwijkingen in de dikte van de hersenschors, kan in de toekomst bijdragen aan een beter inzicht in autisme.

Curriculum Vitae

Marco Hoeksma werd op 27 juni 1971 geboren te Groningen. Vanaf 1983 bezocht hij het Praedinius Gymnasium aldaar, waar hij in 1989 het diploma Gymnasium Ongedeeld behaalde. In hetzelfde jaar werd begonnen met de studie Toegepaste Onderwijskunde aan de Technische Universiteit Twente in Enschede. Van deze studie behaalde hij het propedeutisch examen. In 1992 werd deze studie echter verwisseld voor de studie Psychologie aan de Universiteit Utrecht. Van deze studie werd in 1995 het doctoraal examen in de richting Biopsychologie behaald. Aansluitend volgde een aanstelling als AIO bij de afdeling Kinder- en Jeugd Psychiatrie van het huidige UMC Utrecht, alwaar het onderzoek dat in dit proefschrift is beschreven werd uitgevoerd. Door een samenwerkingsverband tussen deze afdeling en de disciplinegroep Psychofarmacologie van de faculteit Farmacie van de Universiteit Utrecht verbleef de auteur gedurende het grootste deel van zijn aanstelling bij de laatstgenoemde disciplinegroep. Daar is hij ook sinds 1 maart 2002 in dienst als medewerker fysische aspecten van de psychofysiologie en zal hij in dienst treden als post-doc onderzoeker.

Marco woont in Amersfoort, is getrouwd met Mariken en is vader van Marleen.

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