Grey matter abnormality in autism spectrum disorder: An ALE - Meta-analysis study.

GRAY MATTER ABNORMALITY IN AUTISM SPECTRUM DISORDER: AN ALE META-ANALYSIS STUDY

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Abbreviations: FC, Functional Connectivity; BOLD, Blood Oxygenation Level Dependent; ROI, Region Of Interest; ALE, Activation Likelihood Estimation; ASD, Autism Spectrum Disorder; ASP, Asperger's Syndrome; HFA, High Functioning Autism; GM, Gray Matter; WM, White Matter; TAL, Talairach coordinates; VBM, voxel-based morphometry; FFX, Fixed Effects Analysis; RFX, Random Effects Analysis; MTG, Middle Temporal Gyrus; BA, Brodmann Area; ToM, Theory of Mind; Al, Anterior Insula

ABSTRACT

Autism Spectrum Disorder (ASD) is defined on a clinical basis by impairments in social interaction, verbal and non-verbal communication and repetitive or stereotyped behaviors. Voxel-based morphometry (VBM), a technique that gives a probabilistic measure of local gray matter (GM) and white matter (WM) concentration, has been used to study ASD patients: modifications in gray matter volume have been found in various brain regions, such as the corpus callosum, brainstem, amygdala, hippocampus and cerebellum. However, the findings are inconsistent with respect to the specific localization and direction of GM modifications, and no paper has attempted to statistically summarize the results available in the literature. The present study is a quantitative meta-analysis of the current VBM findings aimed at delineating the cortical regions with consistently increased or reduced gray matter concentrations. We employed the Activation likelihood estimation (ALE), a quantitative voxel-based meta-analysis method which can be used to estimate consistent activations across different imaging studies. We generated co-occurrence statistics of a PubMed query employing "Autism Spectrum Disorder" as neuroanatomic lexicon. Significant ALE values related to GM increases were observed bilaterally in the cerebellum, in the middle temporal gyrus, in the right anterior cingulate cortex, caudate head, insula, fusiform gyrus, precuneus and posterior cingulate cortex, and in the left lingual gyrus. GM decreases were observed bilaterally in the cerebellar tonsil and inferior parietal lobule, in the right amygdala, insula, middle temporal gyrus, caudate tail and precuneus, and in the left precentral gyrus.

INTRODUCTION

Autism Spectrum Disorder is a group of genetic neurodevelopmental pathologies with varying degrees of impairments in three social domains: social interaction, communication skills, and repetitive and stereotyped patterns of behavior, interests, and activities. ASD comprises a group of disorders including Autistic disorder, Asperger's syndrome (ASP), Pervasive Developmental Disorder Not Otherwise Specified (PDD-NOS), Rett's disorder and Disintegrative disorder. The autism phenotype exhibits a broad spectrum of symptoms at presentation, differences in course and outcome, adaptive and cognitive levels and response to therapy. As a consequence, the term Autistic Spectrum Disorder is commonly used by researchers and clinicians in the field, though still not recognized by the DSM-IV-TR.[2] There appears to have been an increase in ASD rates in the last decade: the data patterns for the prevalence of ASD in the general population reported in research carried out today are different to those reported in the eighties. However, this difference in epidemiological data may reflect changes in diagnostic criteria and increased categorization of autism spectrum disorders adopted by clinicians and diagnostic manuals.[3] ASD is highly genetic: heritability estimates suggest that about 90% of variance is attributable to genetic factors.[4] Twin studies have shown that 60–90% of monozygotic twins are concordant for ASD, compared with about 10% for dizygotic twins.[5] This

difference in concordance rates suggests that some risk factors interact (gene–gene or gene–environmental interactions). Indeed, gene functions can be altered by toxic environmental factors or epigenetic factors, such as specific aspects of the physical environment (e.g., biochemically active compounds) or specific psychological experiences (e.g., stress). Maternal and pregnancy conditions may influence autism severity, especially a lower birth weight or precarious maternal health conditions like hypertension, albuminuria, and generalized edema. These conditions seem to be associated with higher repetitive behavior scores in children with autism.[6] Prenatal distress is also associated with the increased prevalence of ASD, particularly if the complications occur either toward the middle of gestation or in the weeks just before birth. Prenatal stress may be caused by both psychological and environmental factors.[7]

Gene modifications affect brain chemistry and alter neural tissue. As a neurodevelopmental disorder of prenatal and postnatal brain development, researchers have attempted to elucidate the nature of ASD by examining brain growth. This accelerates at 12 months,[8] and macrocephaly is noted by the age of 2–3 years in 20% of children with ASD. The crucial matter in treating ASD is early detection and diagnosis, which are often difficult: more than a third of children with ASD have not been diagnosed at the age of 2 years.[9] Indeed, clinical signs are usually present in the first years of life, but typical language development might delay identification of symptoms. In Asperger's Syndrome, the diagnosis is often not made until school age, because children show no signs of early impairment in language or cognitive skills.[10] However this condition is often associated with several other disorders, such as genetic disorders (X-Fragile Syndrome), Epilepsy, Mental retardation, Anxiety, Mood disorders, Behavior disorders and Metabolic Disorders,[11] therefore it is necessary to clarify the severity of the disorder and possible co-morbidity with other diseases. Identifying a neuroanatomical fingerprint might enable early detection or monitoring of progress.

Results of neuroimaging studies have shown alterations in gray matter volume in ASD, but findings relating to regional structural brain differences are inconsistent, particularly with regard to the localization and direction (increases or decreases in GM density).[12] We employed a cognitive ontology method to investigate the brain structures with which ASD has been associated: PubBrain [13] is a tool that permits researchers to extract information about associations between terms from the PubMed database and map the results using heatmaps superimposed on a probabilistic atlas of the brain. Fig. 1 shows how the term ASD is related to a wide group of brain areas, with areas in the limbic, prefrontal and cerebellar networks being those most often associated with our term.

Fig. 1 around here

This means that in the recent scientific literature we often find articles that specifically consider ASD and areas in the cerebellum or in the limbic system together. In actual fact, structural neuroimaging studies have found few converging results: increased brain volume in ASD, modified GM density or volume in the brainstem,[14] amygdala, hippocampus

and cerebellum.[15] To the best of our knowledge, no papers have attempted to statistically summarize the neuroanatomical results available in the literature. The aim of the present study is to provide a quantitative meta-analysis of the current Voxel-based morphometry findings to delineate cortical regions with consistently increased or reduced gray matter concentration associated with ASD. Voxel-based morphometry is a technique that gives a probabilistic measure of local GM and WM concentration.[16] To summarize the results of the database searches we employed the Activation likelihood estimation. ALE analysis is a quantitative voxel-based meta-analysis method which can be used to estimate consistent activation across different imaging studies.[17]

MATERIALS AND METHODS

PHENOTYPE MAPS

We generated co-occurrence statistics of a PubMed query employing "Autism Spectrum Disorder" as neuroanatomic lexicon, and projected the weighted results on a 3D probabilistic atlas of the human brain,[18] using PubBrain (http://www.pubbrain.org).[19] PubBrain supports examination and visualization of cognitive concepts in the scientific literature as represented in PubMed. Indeed it maps the literature into phenotype space using a query of any arbitrary PubMed search with respect to an ontology representing neuroanatomic terms (adapted from the Foundational Model of Anatomy and its NeuroNames lexicon) by displaying PubMed hits in the 3D phenotype geometry of brain anatomical regions. The results of a PubBrain search based on a "full" PubMed query are thus representative of the whole PubMed database.

LITERATURE META-ANALYSIS

SELECTION OF STUDIES

To find the relevant studies, we used the keywords “autism" and "voxel-based morphometry”, or “voxel-based morphometry" and "autistic spectrum disorders” (including acronyms and synonyms like “VBM", “ASD", “HFA”, “ASP”). We performed searches on PubMed, Scopus and the University of Turin search engine and downloaded twenty-three papers. Of these, any that did not meet the inclusion criteria were excluded:

i) studies that did not include any gray matter locations, ii) group comparisons not between autism and controls, but with other kinds of disorders (such as pervasive developmental disorders), iii) studies with unspecified VBM analysis as a preliminary study.

Based on these criteria, 16 papers were included, with a total of 728 subjects (350 ASD and 378 controls). The ASD group included diagnosis of High Functioning Autistic (66 subjects), Asperger's Syndrome (86 subjects) and 198 subjects with unspecified diagnosis. This group comprised 290 males and 60 females. The control group comprised 301 males and 77 females. (The complete characteristics of the sample can be viewed in table 1).

ACTIVATION LIKELIHOOD ESTIMATION (ALE)

Activation likelihood estimation analysis is a quantitative voxel-based meta-analysis method which can be used to estimate consistent activation across different imaging studies.[20] ALE maps of co-activations are derived on the basis of foci of interest, where multiple studies have reported statistically significant peak activations.

In the original formulation of Turkeltaub et Al. activation likelihood estimates were calculated for each voxel by modeling each coordinate with an equal weighting using a 3-D Gaussian probability density function. Then a permutation test was carried out to determine the voxel-wise significance of the resulting ALE values. The permutation test was implemented using a non-parametric statistical approach previously described by Turkeltaub et Al, in which a high number of permutations (usually 5000 or more) were generated using the same number of foci and kernel used to generate the ALE map. As such, no assumptions were made with respect to the distribution or spatial separation of these random foci.[21],[17] The resulting statistical maps were corrected for multiple comparisons using false discovery rates (FDR), and then thresholded at $p < 0.05$, corrected.

In the revised algorithm as in Eickhoff et Al,[22] to limit the inter-subject and inter-laboratory variability typical of neuroimaging studies, an algorithm which estimates the spatial uncertainty of each focus and takes into account the possible differences among studies was implemented. This algorithm was preferred to a pre-specified Full Width at Half Maximum (FWHM) as in the original ALE approach.[21] The advantage of the chosen algorithm is that it permits calculation of above-chance clustering between experiments (i.e., random effects analysis, RFX), rather than between foci (fixed effects analysis, FFX).[22] Several modifications have been applied to the original ALE method validated by Turkeltaub PE 2002. Lancaster and colleagues 2000 performed a cluster analysis script to identify areas of high activation likelihood and return the cluster extent above a user-specified threshold, the coordinates of the weighted center of mass and peak location and an anatomical label as assigned by the Talairach Daemon or MNI.[23] Laird and colleagues also added a correction for multiple comparisons and a method for computing statistical contrasts of pairs of ALE images.[17] Indeed these modifications aimed to solve the several limitations known to exist in the original implementation: 1) the aforementioned fixed rather than random effect analysis, 2) the size of the modeled Gaussian that was specified by the user, 3) the permutation t-test was not anatomically constrained. Recent advances in the technique have modified the user-specified Gaussian model into an empirically determined quantitative estimate of the between-subject and between-template variability. This correction modeled the spatial uncertainty of each

coordinate by weighting each study by the number of included subjects. These modifications added statistical power to such a method [22],[24] and eliminated the possibility of ALE results being driven by the results of a single study.

RESULTS

PHENOTYPE MAPS

Fig. 1 shows the results of a PubBrain query employing "Autism Spectrum Disorder" as the search term. The brain structures most often associated with the search term were found in the limbic system, in the brain stem, cerebellum and temporal lobe. More specifically the cingulate gyrus, the hippocampus and the superior frontal gyrus were the gyri most frequently reported to be associated with the search term.

LITERATURE META-ANALYSIS

Fig. 2 and Tables 1 and 2 show the ALE results of the literature meta-analysis.

The systematic search returned 16 papers (the complete list of papers can be viewed in Table 1) yielding 16 experiments performed on 350 subjects. 16 of 21 experiments matched the criteria, yielding 214 locations.

Tables 1 and 2 around here

GM INCREASES

The ALE returned a map of the statistically relevant GM increases (maps were computed at an FDR-corrected threshold of $p < 0.05$, with a minimum cluster size of $K>100$ mm^3 and visualized using MRIcron and BrainVoyager QX 2.2, see Fig. 2 and Table 2).

Significant ALE values related to GM increases were observed in the cerebellum, more specifically in the right declive, pyramis and culmen. Cerebral GM increases were found bilaterally in the Middle Temporal Gyrus as well as in the right Anterior Cingulate Cortex, Caudate head, Insula, Fusiform Gyrus, precuneus and Posterior Cingulate Cortex. GM increases in the left hemisphere were found only in the previously mentioned MTG and lingual gyrus.

Fig. 2 and Table 3 around here

GM DECREASES

The ALE algorithm returned a map of the statistically relevant GM decreases (ALE maps were computed at an FDR-corrected threshold of $p < 0.05$, with a minimum cluster size of $K>100$ mm^3 and visualized using MRIcron and BrainVoyager QX 2.2, see Fig. 2 and Table 3).

Significant ALE values related to GM decreases were observed bilaterally in the cerebellar tonsil and inferior parietal lobule; in the right hemisphere GM decreases were found in the amygdala, insula, middle temporal gyrus, caudate tail and precuneus. In the left hemisphere except for the previously mentioned areas only the precentral gyrus was found to have a reduced GM probability.

Table 4 around here

LATERALIZATION

The areas with a significant GM increase show a clear right lateralization, less predominant in the lingual and medial temporal gyrus (BA 37, 32, 19).

The areas with a significant GM decrease show a less clear-cut lateralization: subcortical areas such as the putamen, amygdala and hippocampus are right lateralized while cortical areas such as BA 6 and 40 are left lateralized. BA 7 and 13 fall in between. See Fig. 3.

Fig. 3 around here

DISCUSSION

Autism, which is derived from the Greek word “autos”, meaning “self”, is defined on a clinical basis by impairments in social interaction, verbal and non-verbal communication and repetitive or stereotyped behaviors. The assignment to different subtypes is based on the number and distribution of endorsed behavioral descriptors in each of the domains, as well as on the age of onset of the symptoms. A broad clinical variability exhibits a continuum among the different disorders, but with a broad spectrum of variables within the phenotype, so that the term “Autism spectrum disorder” is commonly used by researchers and clinicians.[1]; [2]

The original paper by Kanner, in 1943, describing 11 children with autism, did not mention any substantial dysmorphology, except for the presence of “large heads” in five of those children. Additional studies by Piven (1996) indicated that the brain volume increase differs between the frontal, temporal, parietal and occipital lobes.[25] Macrocephaly appears to develop after birth in 80% of cases, but from middle childhood onwards the growth seems to fall below normal so that by later childhood and adolescence cerebral measures in autism are either similar or smaller than normal.[26]

Neuropathological findings have demonstrated an increased cell packaging density and reduced cell size in the hippocampus, subiculum and amygdalae. This suggests a reduced density of axons and dendrites that reflect features of an immature brain.[27] A decreased number of Purkinje cells in the cerebellar hemisphere and vermis and a decrease of mean Purkinje cell size in the cerebellum have been observed.[28],[29] Cortical dysgenesis has been reported, with thickened cortices, high neuronal density, presence of neurons in the molecular layer, irregular laminar patterns and poor gray-white matter boundaries; a reduction of reelin (controlling neuronal migration, correct lamination, synaptic plasticity) and of Bcl-2 (controlling apoptosis) have been found in the cerebellar cortex.[30], [31]

The quantitative ALE meta-analysis of the current VBM findings in our study aimed to delineate brain regions with consistently increased or reduced GM concentrations. The limits of the studies described in the meta-analysis were the potentially confounding factors in terms of differences in age, gender, IQ, heterogeneity of disorders (from autistic disorder to Asperger’s syndrome) of the samples so that, given the limited number of papers included in this meta-analysis, it is not possible to rule out the use of different analyses for each group. Furthermore VBM results can be expressed in terms of volume or density. Both methods are valid but reflect different approaches to the study of brain morphology. Of the 16 papers we included, 10 examined volume and 6 concentration. It was not possible to analyze the two groups differently because we had too few papers to make any such additional analysis reliable. Indeed characterization of the studies in terms of modulation is important, but merging them in a meta-analysis is also effective, since the areas, highlighted in studies comparing the two preprocessing steps, are related (cit). In fact the results were highly overlapping and always qualitatively similar (cit). Upon comparison, the main difference consists in a better specificity of concentration analyses and a better sensitivity of volume analyses (cit).

Another possible limitation in the estimation of a proper activation likelihood is the variability or, in some cases, the uncertainty of the value of smoothing applied during the VBM estimation. In our data this value varied from 4.4 to 12

mm (FWHM) and in one case it was not reported. The ALE method has been developed to approximate the best activation likelihood when the characteristics of the original activations are unknown. This method has been found to be one of the best algorithms for voxel-based meta-analysis currently available. However the variability or lack of important information such as smoothing level, activation shape and cluster volume may lead to misestimation of a proper voxel-based meta-analysis.

Significant ALE values related to GM differences have been observed in cortical regions that are important in social cognitive and/or motor processes in ASD.[32]

Increased total GM brain volume is predictive of greater ASD severity; on the other side specific local GM increases result in a reduction in symptoms: better communication skills are associated with greater GM volume in frontal regions (especially the left middle frontal gyrus) and reduced severity of ASD symptoms is associated with greater GM volume in the right inferior frontal gyrus.[33]

GM thinning in autism in regions associated with the mirror-neuron system (pre-post central gyri, inferior frontal gyrus, medial frontal gyrus, middle temporal gyrus) has been correlated with social and communication deficits.[34]

Regions found to be dismorphic in autism roughly correspond to the brain areas usually involved in theory of mind (ToM) tasks, i.e. tasks measuring the ability to make mental state inferences about others: the superior frontal gyrus, the precuneus region of the medial parietal lobe (important in self-awareness and connected to the medial prefrontal regions and dorsal striatum), and the posterior middle temporal gyrus/sulcus region (biologically relevant motor perception, intentionality of eye gaze direction and inferring intentionality from stories).[32]

Furthermore Caudate nucleus volume correlates with repetitive and stereotyped behavior and social-communication ADI-R total score. In a similar fashion reduced gray matter density in the right cerebellar hemisphere, left temporo-parietal cortex and thalamus correlates with intellectual disabilities in ASD. [32][35]

High functioning autism (HFA) and Asperger's syndrome have partially distinct patterns of gray matter abnormality: HFA patients have significantly smaller gray matter volumes in the subcortical, posterior cingulated and precuneus regions than ASP subjects. Compared to controls, patients with HFA have smaller gray matter volumes in the fronto-pallidal regions, while in ASP patients these are mainly in the bilateral caudate and left thalamus. A significant negative correlation has been found between the size of a gray matter cluster around BA44 language area and the age of acquisition of phrase speech in children with HFA.[36]

These findings suggest that ASD is unlikely to be associated with abnormality in one particular location alone. Instead ASD reflects abnormalities within a particular neural system or multiple systems, that could be partly different in distinct clinical phenotypes.

On the other hand, there may also be a dissociation between ASD patients and controls on the basis of neuroanatomical differences in the spatially distributed cortical network, with two discriminative patterns: an excess network and a deficit network, with brain areas displaying increased or decreased volume.

Anomalies are predominantly in the neuroanatomical network including the limbic system, the fronto-striatal system, the fronto-temporal and the frontoparietal network, and the cerebellar system.[37] The fronto-striatal regions in particular are different in structure, metabolism and functionality in ASD patients and healthy individuals. The fronto and striatal brain regions (such as the head of the caudate nucleus, superior frontal gyrus BA11, anterior cingulated, dorsolateral prefrontal cortex) are reciprocally connected to each other and the thalamus and are involved not only in ASD but interestingly in disorders clinically related to the differential diagnosis of ASD, such as obsessive compulsive disorder and schizophrenia. Abnormalities of these regions in ASD patients have been related to motor abnormalities (abnormal gait sequencing, delayed walking development, abnormal hand positioning), impaired sensory-motor gating, repetitive and stereotyped behavior.

The cerebellum is involved in ASD and is intrinsically connected with itself as well as with the cerebral cortex via the thalamus; it receives afferents from the prefrontal cortex (BA4-primary motor cortex, BA6-premotor cortex), involved in ASD. The deficit network includes the temporo-parietal regions (BA40) and the superior parietal lobe (BA7), which have been linked to mentalizing deficits in Asperger's syndrome.[37]

In the limbic system, the core components (amygdalae, hippocampus, cingulated gyrus fornix, hypothalamus and the thalamus) are embedded into two separate circuits: the orbitofrontal-amygdala circuit (ventral path) and the dorsolateral prefrontal-hippocampal circuit (dorsal path). The ventral circuit, centered around the amygdala and including the anterior cingulated, orbital frontal cortex and temporal lobe, has been implicated in the monitoring of emotional states and social cognition as well as the self-regulation of socially acceptable behavior. The dorsal subsystem, centered around the hippocampus and comprising the parahippocampal, posterior cingulated, parietal and dorsolateral prefrontal cortices, has been linked to processing of events and actions in the service of the visuo-spatial domain and memory.[37]

Considering the critical interactions between multiple distinct brain systems in ASD, an anterior insula-based systems-level model has been proposed. The anterior insula (AI) (considered to be a component of the limbic integration cortex) is involved in interoceptive, affective and empathic processes and it is part of a salience network integrating external sensory stimuli with internal states [38]. The AI serves an integral function with respect to representing and evaluating salient stimuli, and is uniquely positioned as a hub mediating interactions between large-scale brain networks involved in attentional and self-directed processes, and mediates interactions between externally oriented attention and internally oriented cognitive processing. Dysfunctional AI connectivity could play an important role in ASD, resulting in an impaired drive to identify the emotions and thoughts of others and to respond with an appropriate emotion.[38], [39]

In recent genetic studies, polymorphisms of CNTNAP2 (contactin-associated like protein-2), a member of the neurexin family, have already been implicated as a susceptibility gene for ASD. Homozygotes for the risk allele showed significant reductions (gray and white matter volume and fractional anisotropy) in several regions that have already been implicated in ASD, including the cerebellum, fusiform gyrus, occipital and frontal cortices. The finding suggests the possibility that the heterogeneous manifestations of ASD can be etiologically characterized into distinct subtypes through genetic-morphological analysis.[40]

We adopted a Hebbian approach in interpreting the results related to an increase or decrease of volume in different brain areas. Indeed we also observed coherence of volume trends in structures belonging to the same functional network. This however does not explain all cases: there is also evidence of opposite trends in volume changes in structures that are anatomically connected and form a functional network. For example, the amygdalae show a decrease in GM volume, whereas other monosynaptically connected structures such as the temporal pole, the caudate head or the orbitofrontal cortex show an increase in GM volume. *A compensatory function may be involved as, similarly, in asymptomatic Parkinson's disease mutation carriers, where a bilateral compensatory increase in basal ganglia GM has been demonstrated (cit) or in patients with schizophrenia treated with conventional antipsychotics (but not atypical drugs), where a compensatory caudate volume increase was observed (cit).* In ASD, GM volume increases in several structures may reflect an effort to balance a functional decrease in another related structure. A basal ganglia disorder may reflect a frontal cortex deficit and vice-versa, resulting from network damage. Discordances in results for increases or decreases in the same structure may also be related to different steps of disease history, symptom quality and severity, and drugs, with transient functional compensatory increases occurring. Locally increased GM volume, such as in the right inferior frontal gyrus, was associated with reduced severity of symptoms of autism, however increased total GM volume is generally predictive of greater autistic severity, so GM levels should either be considered as a local response or as a compensatory result, also originating from remote structures. Autism could be associated with increased local cortical activity but reduced long-distance connectivity and gray-white matter imbalance may be involved.

CONCLUSION

In this paper we provided a quantitative meta-analysis of the current voxel-based morphometry (VBM) findings on brain regions with consistently increased or reduced gray matter concentrations associated with ASD, compared to controls. Significant ALE values related to GM differences were observed in brain regions that are important in social cognitive or motor processes. Our findings suggest that ASD is unlikely to be associated with abnormalities in one specific location alone; instead, the syndrome reflects abnormalities within multiple, spatially distributed, neural systems. Anomalies are predominantly in the neuroanatomical networks including the limbic system, the fronto-striatal system, the fronto-temporal and the fronto-parietal network, as well as the cerebellar system. It is likely that

the pattern of increased/decreased volume is partially different in distinct clinical phenotypes. Future research should characterize the heterogeneous manifestations of ASD into distinct subtypes, in order to identify specific neuroanatomical fingerprints. At the same time, the neuroanatomical patterns should be correlated with genetic analyses, in order to clarify etiology and help diagnosis.

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Table 1

Studies			Sample					
Author	Title	Journal	Autism			Controls		
			Participants	Age	Range	Participants	Age	Range
Boddaert et Al.	Superior temporal sulcus anatomical abnormalities in childhood autism: a voxel-based morphometry MRI study	NeuroImage, 2004	21 5 Female Unspecified Diagnosis	9.3 ±2.2	7-15	12 5 Female	10.8 ±2.7	7-15
Bonilha et Al.	Gray and white matter imbalance – Typical structural abnormality underlying classic autism?	Brain and development, 2008	12 Unspecified Diagnosis	12.4±4	8-23	16	13.2±5	\
Brieber et Al.	Structural brain abnormalities in adolescents with autism spectrum disorder and patients with attention deficit/hyperactivity disorder	Journal of Child Psychology and Psychiatry, 2007	15 13 ASP 2 HFA	\	10-16	15	\	10-16
Craig	Women with autistic-spectrum disorder:	British journal	14	37.9	\	19	35	\

et Al.	magnetic resonance imaging study of brain anatomy	of psychiatry, 2007	14 Female Unspecified Diagnosis	±11.4		19 Female	±14	
Ecker et Al.	Neuroimage Investigating the predictive value of whole-brain structural MR scans in autism: A pattern classification approach	NeuroImage, 2010	22 Unspecified Diagnosis	27±7	18-42	22	28±7	18-42
Hyde et Al.	Neuroanatomical differences in brain areas implicated in perceptual and other core features of autism revealed by cortical thickness analysis and voxel-based morphometry	Human Brain Mapping, 2009	15 Unspecified Diagnosis	22.7 ±6.4	14-33	13	19.2±5	14-34
Kosaka et Al.	Smaller insula and inferior frontal volumes in young adults with pervasive developmental disorders	Neuroimage, 2010	32 18 HFA 14 ASP	23.8 ±4.2	\	40	22.5 ±4.3	\
McAlonan et Al.	Distinct patterns of grey matter abnormality in high-functioning autism and Asperger's	Journal of Child Psychology and Psychiatry, 2008	33 6 Female 17 HFA	11.6 ±2.59	7-16	55 8 Female	10.7 ±2.74	7-16

	syndrome		16 ASP					
McAlonan et Al.	Mapping the brain in autism. A voxel-based MRI study of volumetric differences and intercorrelations in autism	Brain, 2005	17 1 Female Unspecified Diagnosis	12±1.8	8-12	17 1 Female	11±1.2	8-12
Kwon et Al.	Voxel-based morphometry elucidates structural neuroanatomy of high-functioning autism and Asperger's syndrome	Developmental Medicine & Child Neurology, 2004	20 9 HFA 11 ASP	13.5 ±2.5	\	13	13.7 ±3.1	\
Rojas et Al.	Regional gray matter volumetric changes in autism associated with social and repetitive behavior symptoms	BMC Psychiatry, 2006	24 Unspecified Diagnosis	20.79 ±10.58	7.8-44	23	21.41 ±10.91	7.8-44
Salmond et Al.	The role of the medial temporal lobe in autistic spectrum disorders	European Journal of Neuroscience, 2005	14 1 Female 3 HFA 10 ASP	12.9 ±0.7	8-18	13	12.1 ±0.7	8-18
Salmond et Al.	Heterogeneity in the patterns of neural abnormality in ASD	Cortex, 2005	22 2 Female	12.5	8-18	22 3 Female	12	8-18

			17 HFA 5 ASP					
Spencer et Al.	Structural correlates of intellectual impairment and autistic features in adolescents	NeuroImage, 2006	63 29 Female Unspecified Diagnosis	16±1.8	13-22	72 38 Female	16.7 ±2.1	13-22
Waiter et Al.	A voxel-based investigation of brain structure in male adolescents with autistic spectrum disorder	NeuroImage, 2004	16 16 ASP	15.4 ±2.24	12-20	16	15.5 ±1.6	12-20
Wilson et Al.	A voxel-based morphometry comparison of regional gray matter between fragile X syndrome and autism	Psychiatry Research: Neuroimaging, 2009	10 2 Female Unspecified Diagnosis	30.1 ±9.18	22.2 -47.2	10 3 Female	29.4 ±7.91	21.8 -43.8

Table 2

Studies	**VBM Analysis**			
Author	Scanner	VBM Algorithm	Voxel Size (mm)	Smoothing (mm)
Boddaert et Al.	1.5 T	Optimized VBM Unmodulated (Concentration)	0.86 x 1.33 x 1.20	12
Bonilha et Al.	2 T	Unified Segmentation VBM Modulated (Volume)	1 x 1 x 1	8
Brieber et Al.	1.5 T	Study-specific VBM (Customized Template) Modulated (Volume)	1 x 1.42 x 1	12
Craig et Al.	1.5 T	Optimized VBM Unmodulated (Density)	Thickness 1.5	5
Ecker et Al.	3 T	Unified Segmentation VBM Modulated (Volume)	1.09 x 1.09 x 1	8
Hyde et Al.	3 T	Study-specific VBM (Customized Template) Unmodulated (Concentration)	1 x 1 x 1	12
Kosaka et Al.	3 T	Dartel VBM Modulated (Volume)	0.75 x 1.25 x 1.06	8
McAlonan et Al.	1.5 T	Standard VBM (Talairach Template) Modulated (Volume)	0.86 x 0.86 x 3	-
McAlonan et Al.	1.5 T	Standard VBM (Talairach Template) Modulated (Volume)	Thickness 3 mm	4.4
Kwon et Al.	3 T	Standard VBM (MNI Template) Unmodulated (Density)	Thickness 1.5 mm	8
Rojas et Al.	1.5 T	Optimized VBM	0.94 x 0.94 x 1.7	8

		Modulated (Volume)		
Salmond et Al.	1,5 T	Standard VBM (MNI Template) Unmodulated (Density)	0.8 x 0.8 x 1	12
Salmond et Al.	1.5 T	Standard VBM (MNI Template) Unmodulated (Density)	0.8 x 0.8 x 1	12
Spencer et Al.	1.5 T	Optimized VBM Modulated (Volume)	0.86 x 1.33 x 1.7	12
Waiter et Al.	1.5 T	Optimized VBM Modulated (Volume)	1 x 1 x 1	12
Wilson et Al.	1.5 T	Optimized VBM Modulated (Volume)	0.94 x 0.94 x 1.7	12

Table 3

Cluster #	Volume (mm^3)	Weighted Center (x,y,z)			x	y	z	Label
1	1592	41.75	-57.01	-2.65	42	-56	-2	Right Middle Temporal Gyrus Brodmann area 37
2	1144	32.71	-72.11	-15.3	32	-76	-18	Right Declive
					32	-70	-14	Right Declive
3	680	-40.94	-59.13	-3.28	-42	-60	-4	Left Middle Temporal Gyrus Brodmann area 37
4	680	9.35	29.96	-3.18	4	34	-6	Right Anterior Cingulate Brodmann area 32
					14	26	-2	Right Caudate Head
5	288	29.33	26.57	16.47	30	26	16	Right Insula Brodmann area 13
6	272	43.22	-37	-11.19	44	-38	-12	Right Fusiform Gyrus Brodmann area 37
7	264	27.03	-70.17	-29.54	26	-70	-30	Right Pyramis
8	256	14.62	-44.43	-2.31	14	-44	-2	Right Culmen
9	256	45.43	-8.24	0.57	46	-8	0	Right Insula
10	256	17.76	-51.43	38.55	18	-52	38	Right Precuneus Brodmann area 7
11	232	13.85	-49.3	18.41	14	-50	18	Right Posterior Cingulate Brodmann area 29
12	192	-20.84	-87.99	0.91	-20	-88	2	Left Lingual Gyrus Brodmann area 17
13	104	-26.45	-73.54	0.76	-26	-74	0	Left Lingual Gyrus Brodmann area 19

Cluster #	Volume (mm^3)	Weighted Center (x,y,z)			x	y	z	Label
1	2664	31.41	-5.34	-10.6	30	-8	-12	Right Amygdala
2	1528	36.82	-13.9	14.75	36	-14	14	Right Insula Brodmann area 13
3	1320	-4.21	-54.83	-37.93	-4	-54	-38	Left Cerebellar Tonsil
4	832	-50.08	-31.91	23.25	-50	-34	24	Left Inferior Parietal Lobule Brodmann area 40

5	632	46.54	-35.62	-7.03	52	-40	-8	Right Middle Temporal Gyrus Brodmann area 20
					40	-32	-6	Right Caudate Tail
6	560	48.95	-48.4	-31.45	50	-48	-32	Right Cerebellar Tonsil
7	376	-45.92	-3.15	44.31	-46	-4	44	Left Precentral Gyrus Brodmann area 6
8	216	1.48	-61.79	38.36	2	-62	38	Right Precuneus Brodmann area 7
9	168	-26	-38.74	-44.5	-26	-38	-44	Left Cerebellar Tonsil
10	136	30.7	-39.76	52.94	30	-40	52	Right Inferior Parietal Lobule Brodmann area 40

CAPTIONS TO FIGURES

Fig. 1 Cognitive Ontologies

PubBrain output for the query “Autism Spectrum Disorder”, illustrating the retrieval results for an arbitrary location (crosshairs positioned in the cingulate gyrus).

Upper panels show heatmaps reporting the co-occurrence of the search term and brain anatomical locations.

Middle panels show a graph reporting the co-occurrence of the search term and each brain anatomical location.

Lower panels show a graph reporting the brain macroareas most often associated with the search term.

Fig. 2 Gray Matter ALE Results

Colors from red to yellow show GM increases, colors from blue to green show GM decreases (2D ALE maps were computed at an FDR-corrected threshold of $p < 0.05$, with a minimum cluster size of $K>100$ mm^3 and visualized using MRIcron)

Fig. 3 Gray Matter ALE Lateralization

Upper panels show lateralization graphs relative to GM increases

Lower panels show lateralization graphs relative to GM decreases

(Lateralization Graphs were computed with a custom-developed matlab script, see supporting online materials for a more detailed explanation)

CAPTIONS TO TABLES

Table 1 Overview of the included studies and Demographics of the included studies

Abbreviations:

HFA: High Functioning Autism

ASP: Asperger's Syndrome

ASD: Autistic Spectrum Disorders

Table 2 VBM analysis of the included studies

Table 3 Gray Matter Increases ALE Results

(ALE maps were computed at an FDR-corrected threshold of $p < 0.05$, with a minimum cluster size of $K>100\ mm^3$)

Table 4 Gray Matter Decreases ALE Results

(ALE maps were computed at an FDR-corrected threshold of $p < 0.05$, with a minimum cluster size of $K>100\ mm^3$)