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# GYRIFICATION BRAIN ABNORMALITIES AS PREDICTORS OF OUTCOME IN ANOREXIA NERVOSA

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# GYRIFICATION BRAIN ABNORMALITIES AS PREDICTORS OF OUTCOME IN

# ANOREXIA NERVOSA

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**Short title:** Brain giryfication in anorexia nervosa

**Key words:** anorexia nervosa, cortical folding, outcome, MRI, brain gyrification

## ABSTRACT

Gyrification brain abnormalities are considered a marker of early deviations from normal

developmental trajectories and a putative predictor of poor outcome in psychiatric disorders. The

aim of this study was to explore cortical folding morphology in patients with anorexia nervosa

(AN). A MRI brain study was conducted on 38 patients with AN, 20 fully recovered patients, and

38 healthy women. Local gyrification was measured with procedures implemented in FreeSurfer.

Vertex-wise comparisons were carried out to compare: 1) AN patients and healthy women; 2)

patients with a full remission at a 3-year longitudinal follow-up assessment and patients who did not

recover.

AN patients have significantly lower gyrification when compared with healthy controls. Patients

with a poor 3-year outcome had significantly lower baseline gyrification when compared to both

healthy women and patients with full recovery at follow-up, even after controlling for the effects of

duration of illness and grey matter volume. No significant correlation has been found between

gyrification, body mass index, amount of weight loss, onset age, and duration of illness. Brain

gyrification significantly predicts outcome at follow-up even after controlling for the effects of

duration of illness and other clinical prognostic factors. Although the role of starvation in

determining our findings cannot be excluded, our study showed that brain gyrification might be a

predictor of outcome in AN. Further studies are needed to understand if brain gyrification

abnormalities are indices of early neurodevelopmental alterations, the consequence of starvation, or

the interaction between both factors.

**INTRODUCTION**

Anorexia nervosa (AN) is a severe psychiatric disorder characterized by a high female/male ratio, onset during adolescence (Favaro et al. 2009), and high levels of chronicity and mortality. Individuals suffering from AN reduce their food intake or increase their energy expenditure, have a distorted body image and often display low levels of insight about their condition. Although several studies have been conducted to explore the effects of having a diagnosis of anorexia nervosa (AN) on brain volumes, no study to date has been carried out to investigate the presence of cortical morphological abnormalities in this group of patients. With few exceptions (Brooks et al. 2011; Frank et al. 2013), AN has been associated with a reduction of both gray and white matter (Fonville et al. 2014; King et al. 2015; Seitz et al. 2014; Titova et al. 2014; Van den Eynde et al. 2012) which is usually considered a side effect of weight loss and malnutrition, given that an improvement of brain volumes has been observed in longitudinal studies (Seitz et al. 2014; Van den Eynde et al. 2012). Whether any scarring effect might persist after full recovery is still a matter of debate, since some studies found a persistent reduction of gray matter volume in the anterior cingulate cortex of fully recovered AN patients (Muhlau et al. 2007), while others found no differences between recovered patients and healthy women (Wagner et al. 2006).

Since the effects of weight loss and malnutrition on brain volumes are known, although not completely characterized, and since malnutrition often begins a considerable amount of time before the onset of the disorder, only long-term longitudinal studies of large cohorts of healthy adolescents would allow to infer the role of brain morphological aberrations in the risk of developing AN using volumetric studies. This question is of some importance since evidence about AN having a possible neurodevelopmental origin is increasing: the disorder has its onset during adolescence (Favaro et al. 2009), is associated with an excess of left-handedness (Tenconi et al. 2010), specific

neuropsychological impairments (Lopez et al. 2008; Roberts et al. 2007; Tenconi et al. 2010), and subtle neurological abnormalities (Gillberg et al. 1994). In addition, prenatal and perinatal risk factors that have been implicated in other neurodevelopmental psychiatric disorders, such as autism and schizophrenia, seem to increase the risk of developing AN (Favaro et al. 2006; 2014; Tenconi et al. 2014). For this reasons, it is important to look for measures of brain morphology and function that are potentially not affected by malnutrition and other illness-related factors.

In this study, we explored whether the investigation of brain gyrification can provide useful information about neurodevelopment outcome in AN. Cortical folding is known to reflect a person's prenatal development (Dubois et al. 2008) and change only minimally after birth (White et al. 2010): it is the result of a substantial expansion of cortical surface area despite a relatively minor gain in cortical thickness during evolutionary development (Sun and Hevner, 2014). The processes leading to cortical gyrification are not fully understood, but recent theories consider cortical folding to be the result of underlying patterns of connectivity (Van Essen, 1997), thus emphasizing the possible functional significance of gyrification alterations (Sun and Hevner, 2014). The gyrification index, which is the ratio between the inner folded contour and the length of the coronal outline, excluding the sulcal regions, has been found to increase dramatically during the third trimester of pregnancy, and then to remain relatively constant throughout development (Armstrong et al. 1995): this means that the process of gyrification continues through the postnatal developmental period, maintaining this constant ratio throughout the process (White et al. 2010). For this reason, the degree of folding is considered to be a crucial valid marker to our understanding of the timing and the nature of brain alterations during neurodevelopment (Dubois et al. 2008; White et al. 2010; Schaer et al. 2013). Gyrification alterations have been found to be associated with prematurity (Gimenez et al. 2006; Kesler et al. 2006) and exposure to obstetric complications (Haukvik et al. 2012), and to be frequent in neurodevelopmental psychiatric disorders such as autism and schizophrenia (Nanda et al. 2014; Nesvag et al. 2014; White and Hilgetag, 2011).

Furthermore in first episode psychosis, cortical folding in specific brain areas seems to significantly predict response to antipsychotic treatment (Palaniyappan et al. 2013). Despite the importance of having reliable predictors of poor response in AN, to our knowledge, no attempt to date has been made to explore the relationship between brain characteristics and outcome. The aims of this study were twofold: 1) to investigate the pattern of brain gyrification in a sample of patients with AN and study the relationship between gyrification and relevant clinical variables, such as body mass index, the amount of weight loss, duration of illness, age of onset; 2) to explore the relationship between brain gyrification and the outcome at a 3-year follow-up.

## METHODS

A total of 38 patients with acute AN and 38 healthy controls were included in this study. A further group of 20 patients in full remission from AN were included to test the state/trait nature of any MRI finding. Women who had recovered from AN were subjects who had had full AN in their lifetime, but were asymptomatic from at least 6 months at the time of scanning (mean remission time: 38.5 months (standard deviation=33.2); range 6-96). All recovered subjects were asked to participate during follow-up visits carried out in our ED Unit after recovery. None of the subjects of this group relapsed in the year following the study. Table 1 describes the main characteristics of the sample.

Exclusion criteria for all subjects were male gender, history of head trauma or injury with loss of consciousness, history of any serious neurological or medical illness, active use of systemic steroids, pregnancy, active suicidality or major depression, history of substance/alcohol abuse or dependence, bipolar disorder or schizophrenia spectrum disorder, moderate mental impairment (IQ<60) or learning disabilities, use of medications other than antidepressants, and known contraindications to conventional MRI. History of any psychiatric disorder and any first-degree

relatives with an eating disorder were additional exclusion criteria for healthy women. Amenorrhea,

food restriction, bingeing, excessive exercise, fasting and purging in the last 6-months were

additional exclusion criteria for the recovered AN group.

Ethical permission was obtained from the ethics committee of the Padova Hospital. After

completely describing the study to the subjects, informed written informed consent was obtained.

Clinical Assessment and Follow-up

In all subjects, diagnostic interviews were performed according to the eating disorders section of the

Structured Clinical Interview for DSM-IV (First et al. 1995) and a semi-structured interview to

gather socio-demographic and clinical variables, as previously described (Favaro et al. 2012; 2013).

Subjects were also asked to complete the Hopkins Symptoms Checklist (Derogatis et al. 1974) to

assess depressive and obsessive-compulsive symptoms and the Eating Disorders Inventory (Garner

et al. 1983) to assess eating psychopathology. Handedness was assessed by the Edinburgh

Handedness Inventory (Oldfield, 1971).

All subjects were medically stable at the time of scanning and all were recruited at the Padova

Hospital Eating Disorders Unit and all fulfilled all the diagnostic criteria for AN according to DSM-

IV at the time of scanning. The diagnostic subtype at the time of scanning was restrictive in 32 AN

patients (84%) and binge eating/purging type in 6 patients, but 7 patients who were restrictive at the

time of the present study reported previous recurrent binge eating and/or purging. Concerning the

use of medications, 14 AN patients and 4 recovered women were under treatment with

antidepressants at the time the study was conducted.

Follow-up was performed 3 years later (on average 3.4 years, range 1.7-3.9) in acute AN patients.

Both face-to-face interviews and reports from informants were used to gather diagnostic

information. Patients were assessed using the eating disorders section of the Structured Clinical

Interview for DSM-IV (First et al. 1995). Full recovery was defined as weight being in the normal

range, regular menses, absence of binge eating, purging, food avoidance or restraint, excessive

exercise, undue body dissatisfaction, or drive for thinness in the past 3 months.

MRI Data Acquisition

Data were collected on a Philips Achieva 1.5 Tesla scanner equipped for echo-planar imaging. A

high-resolution 3D T1-weighted anatomical image was also acquired, in a gradient-echo sequence

(repetition-time=20 sec, echo time=3.78 msec, flip angle= 20°, 160 sagittal slices, acquisition voxel

size=1×0.66×0.66 mm, field of view 21-22 cm).

Data Processing and Statistics

Surface extraction was completed using the FreeSurfer package (Martinos Center for Biomedical

Imaging, Massachusetts General Hospital, Boston) version 5.3.0. Preprocessing, cortical

reconstruction and segmentation, and cortical thickness estimation were performed according to

standard protocols (Desikan et al. 2006; Fischl et al. 1999; Fischl and Dale, 2000). The local

Gyrification Index (lGI) was measured at thousands of points of the reconstructed cortical surface

using previously validated algorithms (Schaer et al. 2008). LGI is a measurement of the degree of

cortical folding that quantifies the amount of cortex buried with in the sulcal folds in the

surrounding circular region. A general linear model controlling for the effects of age and

intracranial volume to estimate differences among AN patients and healthy women at each vertex of

the right and left hemispheric surfaces. We also compared patients with full recovery to those not

making a full recovery over the subsequent three years (and both of these groups were compared to

healthy women) controlling for the effects of age, duration of illness, and intracranial volume. In

order to reduce the possibility of biased findings due to the reduced cortical volume in AN patients,

all vertex-wise analyses were repeated including cortical thickness as a vertex-wise covariate.

The Monte-Carlo permutation approach (10,000 simulations) implemented in the FreeSurfer

software was used for statistical correction of multiple comparisons with an additional Bonferroni

correction to account for the two hemispheres. Clusterwise probability values of p<0.01 were

considered statistically significant in the present study.

Statistics

General linear models were used to test differences in the overall gyrification index between groups

using age and total intracranial volume as covariates. Nonparametric statistical tests were conducted

in correlations and group comparisons for the other variables. These procedures were implemented

with Statistical Product and Service Solutions software (SPSS, Inc, Chicago, Ill).

**RESULTS**

Diagnostic Group Comparisons

The overall lGI of both hemispheres was significantly lower in AN patients than in healthy women

(left: $2.85 \pm 0.09$ vs. $2.90 \pm 0.11$; $F(3,72) = 4.93$; $p = 0.03$; right: $2.85 \pm 0.10$ vs. $2.90 \pm 0.12$;

$F(3,72) = 4.37$; $p = 0.04$). AN patients who had recovered from the disorder showed no differences

in comparison to healthy women in overall hemispheric lGI (left: $2.90 \pm 0.09$; $F(3,54) = 0.11$; $p =$

$0.745$; right: $2.90 \pm 0.09$; $F(3,54) = 0.12$; $p = 0.730$). Figure 1 shows the results of the vertex-wise

comparison between AN patients and healthy women: both in the left and in the right hemisphere

an area in the parietal cortex showed significantly lower gyrification in AN patients. These areas

mainly encompassed: in the left hemisphere: precentral, postcentral, superior parietal,

supramarginal, and rostral-middle-frontal gyri; in the right hemisphere: precentral and superior-

frontal gyri. Performing the analysis including cortical thickness as a vertex-wise covariate changed

only marginally the brain areas of significant differences (Supplementary Figure 1). The

comparison between recovered AN patients and healthy women revealed no cluster of significant

difference after the multiple comparison correction.

Neither overall lGIs, nor vertex-wise analyses, led to a significant correlation between gyrification

and body mass index, amount of weight loss, age of onset, or duration of illness (Table 2). No

significant differences emerged between patients who were taking antidepressants and those who

were not (left overall lGI: $2.84 \pm 0.06$ vs. $2.85 \pm 0.11$; $F(3,34) = 0.28$; $p = 0.60$; right overall lGI:

$2.83 \pm 0.06$ vs. $2.86 \pm 0.12$; $F(3,34) = 0.03$; $p = 0.87$). In the 20 AN patients where bioimpedence

analysis was performed, no significant correlation between gyrification and hydration index was

found (left cluster: rho=0.03; p=0.91; right cluster: rho=0.11; p=0.65). Significant negative age

correlations emerged in both the AN and healthy control groups, but no correlation × group

interaction was found (Supplementary Figure 2). AN patients also revealed several areas of

significantly reduced cortical thickness (Supplementary Table 1), that only partially overlapped

with cortical areas displaying a reduced gyrification index (Supplementary Figure 3).


Outcome Analysis

Out of the patients who performed the 3 year follow-up assessment (n=37), 13 (35%) were

completely recovered. Figure 2 and Table 2 shows the baseline differences between patients with

full recovery and those not making a full recover over the subsequent 3 years. Performing the

analysis including cortical thickness as a vertex-wise covariate changed only marginally the brain

areas of significant differences (Supplementary Figure 4). Areas of between-group significant

differences were located in bilateral superior parietal cortex and, in the left hemisphere, overlap

with brain areas that showed significant difference in the comparison between the whole group of

AN patients and healthy women. The group with poor outcome displayed significantly lower

baseline overall gyrification both in comparison to healthy women (left: $2.82 \pm 0.10$ vs. $2.90 \pm 0.11$;

$F(3,58) = 6.98$; $p = 0.011$; right: $2.82 \pm 0.10$ vs. $2.90 \pm 0.12$; $F(3,58) = 6.60$; $p = 0.013$) and in

comparison to those AN patients who recovered (left: $2.82 \pm 0.10$ vs. $2.90 \pm 0.06$; $F(3,33) = 4.75$; $p = 0.04$; right $2.82 \pm 0.10$ vs. $2.90 \pm 0.07$; $F(3,33) = 5.15$; $p = 0.03$). No differences in gyrification emerged between good outcome AN patients and healthy women. At vertex-wise analysis, the AN group with poor outcome showed brain areas of significantly decreased gyrification in comparison to healthy women (Supplementary Figure 5).

Finally, we tested outcome predictive ability for both the overall hemispheric gyrification indices and the average gyrification index of clusters significantly differentiating patients with full recovery and those not making a full recover. Both overall gyrification indices and those specific to significant clusters showed a significant predictive ability in identifying good outcome patients (Table 3). No relevant changes in the models were observed after including other possible prognostic variables, such as SSRI therapy, depressive/anxiety symptoms, diagnostic subtype, and severity of eating psychopathology.

## DISCUSSION

This study was the first to explore the presence of cortical folding aberrations in AN and the relationship between cortical morphology and outcome. In both hemispheres, AN patients showed decreased gyrification in comparison to healthy women in brain areas (superior parietal and frontal cortex) which are considered crucial for integration and elaboration of somatosensory perceptions, visuo-spatial abilities, and executive functions. These cognitive functions may be related to body image disturbance, which is considered one of the 'core' characteristics of this disorder (Favaro et al. 2012). Functional neuroimaging studies relying on different measurement techniques have consistently implicated aberrations in the bilateral parietal cortex (Kaye et al. 2013; Van Kuyck et al. 2009) as possible markers of AN, given its role in body image elaboration. However, AN patients often also display problems in cognitive style, such as poor set-shifting (Roberts et al.

2007; Tenconi et al. 2010) and decision making abilities (Bodell et al. 2014), these being

considered to mainly rely on prefrontal cortex functioning.

Although the mechanisms behind developmental aspects of cortical folds are still unclear, it is

hypothesized that the adult pattern of gyrification somewhat reflects the integrity of the process of

connectivity development during prenatal life (Dubois et al. 2008; White et al. 2010). In this

context, our findings would confirm a neurodevelopmental hypothesis for AN (Favaro, 2013),

supported by evidence of similar patterns of hypogyria in other neurodevelopmental psychiatric

disorders, such as schizophrenia (Palaniyappan and Liddle, 2012) and autism (Libero et al. 2014).

However, starvation in AN is associated with anatomical brain alterations that potentially could

account for our findings. The local gyrification index is not able to distinguish between a decrease

in number or in development of sulci, from one hand, and reduced sulcal depth or increased sulcal

width on the other (Palaniyappan et al. 2013). In other words, a reduced gyrification index could

imply either hypogiria or cortical flattening due to a decrease in cortical volume and thickness. To

date, histological cortical changes associated with weight loss and malnutrition in AN have received

little attention in literature (Neumarker et al. 1997). The decrease in both gray and white matter

volumes and their potential (at least partial) reversibility lead to the hypothesis that rather than

neuronal death or other types of neurodegeneration, it is the reduced dendritic ramification and

spine density, as well as the reduced number/volume of glia cells, that must be held accountable for

these changes (Neumarker et al. 1997; Seitz et al. 2014). Moreover, it is worth noting that the

adolescent brain is characterized by developmental changes that physiologically imply reduced gray

matter volume (due to cortical thinning) and increased white matter volume associated with the

continuing process of myelination in the neuropil and deep intracortical layers (Aleman-Gomez et

al. 2013; Klein et al. 2014). The impact of AN on brain developmental trajectories is not known and

longitudinal studies would be of great importance for understanding the specific brain changes in

this type of patients. In our cross-sectional MRI study, we found both evidence in favor of as well

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as evidence against the hypothesis of an effect of weight loss in the determination of gyrification

alterations. Our finding of an absence of gyrification alterations in the sample of recovered AN

patients would support a weight loss effect hypothesis lacking long-term consequences. This would

be in line with previous studies about cortical thinning in AN (King et al. 2015) and many

preceding voxel-based morphometry studies (Van den Eynde et al. 2012; Seitz et al. 2014).

According to this hypothesis, decreased gyrification in our sample of acute AN patients could

simply be the result of reduced cortical thickness. In contrast with this hypothesis, however, we

found that maps of reduced cortical thickness and of reduced gyrification only show a marginal

overlapping in our sample and including maps of cortical thickness as a vertex-wise covariate did

not affect our findings. In addition, we observed a decrease of brain gyrification along with age in

large brain areas of both samples (as reported in previous studies on healthy individuals (Aleman-

Gomez et al. 2013; Klein et al. 2014)), with no evidence of differences between groups in the

pattern of gyrification changes.

While analyzing our second objective, namely the impact of brain gyrification in predicting

outcome at follow-up, an alternative hypothesis emerged: in our analyses, we found that low levels

of gyrification significantly predicted poor outcome at a 3-year follow-up, whereas the 'good

outcome' group showed no differences in gyrification index when compared to healthy women

even though they were, on average, severely underweight.

The pattern of our findings could thus fit the hypothesis that abnormal brain gyrification is more

evident in a subgroup of 'poor outcome' AN patients, probably as the consequence of a more severe

disruption of neurodevelopmental processes. Since the recovered AN group is 'per definition' a

group with a good prognosis, this hypothesis would explain why gyrification was not impaired in

this group. Moreover, such result is in line with previous papers dealing with other psychiatric

disorders which found that gyrification is not only a marker of integrity of normal cortical

development (Palaniyappan and Liddle, 2012; Schaer et al. 2008), but also characterizes a subgroup

of patients with low response to treatments (Palaniyappan et al. 2013). Furthermore, in our sample, gyrification indices were able to predict outcome even after controlling for the effects of well known prognostic factors such as weight loss, body mass index, severity of psychopathology, and duration of illness. If confirmed by future studies, this finding would be of great importance for the identification of patients with poor response to standard treatments as well as for the provision, in these cases, of more intensive or alternative biological (Lipsman et al. 2013) or non-biological (Tchanturia et al. 2014) treatments in order to obtain symptomatic relief. Since AN is an illness with relatively low levels of recovery and high levels of chronicization (Treasure et al. 2010), our findings might be taken as a warning against the tendency to generalize brain studies based on recovered patients. Indeed, in these studies although the effects of the presence of malnutrition on the brain are not present, it is very difficult to keep track of the effects of those favorable prognostic factors that made recovery possible.

This study has several strengths, as well as important limitations, which should be taken into consideration. It is the first to explore brain gyrification in terms of the possible relationships between brain characteristics and subsequent outcomes in a sample of AN patients. However, a particular word of caution must apply in interpreting brain findings in AN samples for which it is often difficult to disentangle the effects of early developmental factors on the brain and the consequences of starvation. In particular, although in our study no linear correlation has emerged between gyrification and duration of illness, we cannot exclude prolonged starvation as a possible cause of our findings as the poor outcome group had on average a duration of more than 8 years of illness. This is the reason why duration of illness was included as a covariate in the comparison between AN groups. Furthermore, according to a sort of 'two-hit' model similar to that hypothesized for schizophrenia, our findings could be the result of an interaction between early developmental factors and later weight loss (Favaro, 2013; Schmitt et al. 2014). Although in this study AN patients were assessed after their medical conditions had stabilized, hydration is another

factor that could account for variations in the shape and volume of the brain cortex. However, we tested this possible correlation in a subsample of severely underweight patients and found no significant effects.

In conclusion, the present study provides evidence of cortical gyrification alterations in the parietal and frontal cortex of patients with AN. These alterations did not show a linear relationship with body mass index, cortical thickness or dehydration and these changes were not present in patients with a good outcome, regardless of their body weight and recovery status. Longitudinal studies carried out on large cohorts of patients suffering from AN are needed to explore the effects of malnutrition and weight loss on cortical morphology and to understand how these interact with the duration of the illness, the response to treatment and aging. Moreover, to explore the possible involvement of these cortical aberrations in the psychopathology of AN and their clinical and scientific implications, it would be interesting to study at least two further aspects. One is the replicability of our findings in patients suffering from the illness for a short period of time or in high-risk individuals who do not have AN. The other is the exploration of possible structural/functional connectivity alterations that, according to the Van Essen's hypothesis, underly aberrations of cortical folding (Van Essen, 1997). Any possible hypothesis about a possible neurodevelopmental origin of these cortical aberrations requires a careful examination and exclusion of alternative hypotheses. However, if our findings will be replicated and supported by future studies, cortical folding alterations in AN patients, regardless of their origin, could represent a useful method for identifying patients who are resistant to conventional treatments at an early stage.

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# References

Aleman-Gomez Y, Janssen J, Schnack H, Balaban E, Pina-Camacho L, Alfaro-Almagro F, Castro-Fornieles J, Otero S, Baeza I, Moreno D, Bargallo N, Parellada M, Arango C, Desco M (2013): The human cerebral cortex flattens during adolescence. J Neurosci 33:15004-15010.

Armstrong E, Schleicher A, Omran H, Curtis M, Zilles K (1995): The ontogeny of human gyrification. Cereb Cortex 5:56-63.

Bodell LP, Keel PK, Brumm MC, Akubuiro A, Caballero J, Tranel D, Hodis B, McCormick LM (2014): Longitudinal examination of decision-making performance in anorexia nervosa: Before and after weight restoration. J Psychiatr Res 56:150-157.

Brooks SJ, Barker GJ, O'Daly OG, Brammer M, Williams SC, Benedict C, Schiöth HB, Treasure J, Campbell IC (2011): Restraint of appetite and reduced regional brain volumes in anorexia nervosa: A voxel-based morphometric study. BMC Psychiatry 11:179. doi 10.1186/1471-244X-11-179.

Derogatis LR, Lipman RS, Rickels K, Uhlenhuth EH, Covi L (1974): The Hopkins Symptom Checklist (HSCL): a self-report symptom inventory. Behav Sci 19:1-15.

Desikan RS, Segonne F, Fischl B, Quinn BT, Dickerson BC, Blacker D, Buckner RL, Dale AM, Maguire RP, Hyman BT, Albert MS, Killiany RJ (2006): An automated labeling system for subdividing the human cerebral cortex on MRI scans into gyral based regions of interest. Neuroimage 31:968-980.

Dubois J, Benders M, Borradori-Tolsa C, Cachia A, Lazeyras F, Ha-Vinh Leuchter R, Sizonenko SV, Warfield SK, Mangin JF, Hüppi PS (2008): Primary cortical folding in the human newborn: An early marker of later functional development. Brain 131:2028-2041.

Favaro A (2013): Brain development and neurocircuit modeling are the interface between genetic/environmental risk factors and eating disorders. Int J Eat Disord 46:443-446.

Favaro A, Caregaro L, Tenconi E, Bosello R, Santonastaso P (2009): Time trends in age at onset of anorexia nervosa and bulimia nervosa. J Clin Psychiatry 70:1715-1721.

Favaro A, Clementi M, Manara R, Bosello R, Forzan M, Bruson A, Tenconi E, Degortes D, Titton F, Di Salle F, Santonastaso P (2013): Catechol-O-methyltransferase genotype modifies executive functioning and prefrontal functional connectivity in women with anorexia nervosa. J Psychiatry Neurosci 38:241-248.

Favaro A, Santonastaso P, Manara R, Bosello R, Bommarito G, Tenconi E, Di Salle F (2012): Disruption of visuospatial and somatosensory functional connectivity in anorexia nervosa. Biol Psychiatry 72:864-870.

Favaro A, Tenconi E, Degortes D, Manara R, Santonastaso P (2014): Effects of obstetric complications on volume and functional connectivity of striatum in anorexia nervosa patients. Int J Eat Disord 47:686-695.

Favaro A, Tenconi E, Santonastaso P (2006): Perinatal factors and the risk of developing anorexia nervosa and bulimia nervosa. Arch Gen Psychiatry 63:82-88.

First MB, Spitzer RL, Gibbon M, Williams JBW (1995): Structured Clinical Interview for DSM-IV Axis I Disorders. Biometrics Research Department, New York.

Fischl B, Dale AM (2000): Measuring the thickness of the human cerebral cortex from magnetic resonance images. Proc Natl Acad Sci USA 97:11050-11055.

Fischl B, Sereno MI, Dale AM (1999): Cortical surface-based analysis. II: Inflation, flattening, and a surface-based coordinate system. NeuroImage 9:195-207.

Fonville L, Giampietro V, Williams SC, Simmons A, Tchanturia K (2014): Alterations in brain structure in adults with anorexia nervosa and the impact of illness duration. Psychol Med 44:1965-1975.

Frank GK, Shott ME, Hagman JO, Mittal VA (2013): Alterations in brain structures related to taste reward circuitry in ill and recovered anorexia nervosa and in bulimia nervosa. Am J Psychiatry 170:1152-1160.

Garner DM, Olmsted MP, Polivy J (1983): Development and validation of a multidimensional Eating Disorder Inventory for anorexia nervosa and bulimia. Int J Eat Disord 2:15-34.

Gillberg C, Rastam M, Gillberg IC (1994): Anorexia nervosa: physical health and neurodevelopment at 16 and 21 years. Dev Med Child Neur 36:567-575.

Giménez M, Junqué C, Vendrell P, Narberhaus A, Bargalló N, Botet F, Mercader JM (2006): Abnormal orbitofrontal development due to prematurity. Neurology 67:1818-1822.

Haukvik UK, Schaer M, Nesvåg R, McNeil T, Hartberg CB, Jönsson EG, Eliez S, Agartz I (2012): Cortical folding in Broca's area relates to obstetric complications in schizophrenia patients and healthy controls. Psychol Med 42:1329-1337.

Kaye WH, Wierenga CE, Bailer UF, Simmons AN, Bischoff-Grethe A (2013): Nothing tastes as good as skinny feels: the neurobiology of anorexia nervosa. Trends Neurosci 36:110-120.

Kesler SR, Vohr B, Schneider KC, Katz KH, Makuch RW, Reiss AL, Ment LR (2006): Increased temporal lobe gyrification in preterm children. Neuropsychologia 44:445-453.

King JA, Geisler D, Ritschel F, Schober I, Seidel M, Roschinski B, Soltwedel L, Zwipp J, Pfuhl G, Marxen M, Roessner V, Ehrlich S (2015): Global cortical thinning in acute anorexia nervosa normalizes following long-term weight restoration. Biol Psychiatry 77:624-632.

Klein D, Rotarska-Jagiela A, Genc E, Sritharan S, Mohr H, Roux F, Han CE, Kaiser M, Singer W, Uhlhaas PJ (2014): Adolescent brain maturation and cortical folding: evidence for reductions in gyrification. PLos One 9: e84914. doi:10.1371/journal.pone.0084914.

Libero LE, DeRamus TP, Deshpande HD, Kana RK (2014): Surface-based morphometry of the cortical architecture of autism spectrum disorders: volume, thickness, area, and gyrification. Neuropsychologia 62:1-10.

Lipsman N, Woodside DB, Giacobbe P, Lozano AM (2013): Neurosurgical treatment of anorexia

nervosa: review of the literature from leucotomy to deep brain stimulation. Eur Eat Disord Rev

21:428-435.

Lopez C, Tchanturia K, Stahl D, Treasure J (2008): Central coherence in eating disorders: a

systematic review. Psychol Med 38:1393-1404.

Muhlau M, Gaser C, Ilg R, Conrad B, Leibl C, Cebulla MH, Backmund H, Gerlinghoff M, Lommer

P, Schnebel A, Wohlschläger AM, Zimmer C (2007): Gray matter decrease of the anterior

cingulate cortex in anorexia nervosa. Am J Psychiatry 164:1850-1857.

Nanda P, Tandon N, Mathew IT, Giakoumatos CI, Abhishekh HA, Clementz BA, Pearlson GD,

Sweeney J, Tamminga CA, Keshavan MS (2014): Local gyrification index in probands with

psychotic disorders and their first-degree relatives. Biol Psychiatry 76:447-455.

Nesvag R, Schaer M, Haukvik UK, Westlye LT, Rimol LM, Lange EH, Hartberg CB, Ottet M-C,

Melle I, Andreassen OA, Jönsson EG, Agartz I, Eliez S (2014): Reduced brain cortical folding

in schizophrenia revealed in two independent samples. Schizophr Res 152:333-338.

Neumarker KJ, Dudeck U, Meyer U, Neumarker U, Schulz E, Schonheit B (1997): Anorexia

nervosa and sudden death in childhood: clinical data and results obtained from quantitative

neurohistological investigations of cortical neurons. Eur Arch Psychiatry Clin Neurosci

247:16-22.

Oldfield RC (1971): The assessment and analysis of handedness: the Edinburgh inventory.

Neuropsychologia 9:97-113.

Palaniyappan L, Liddle PF (2012): Aberrant cortical gyrification in schizophrenia: a surface-based

morphometry study. J Psychiat Neurosci 37:399-406.

Palaniyappan L, Marques TR, Taylor H, Handley R, Mondelli V, Bonaccorso S, Giordano A,

DiForti M, Simmons A, David AS, Pariante CM, Murray RM, Dazzan P (2013): Cortical

folding defects as markers of poor treatment response in first-episode psychosis. JAMA

Psychiatry 70:1031-1040.

Roberts ME, Tchanturia K, Stahl D, Southgate L, Treasure J (2007): A systematic review and meta-

analysis of set-shifting ability in eating disorders. Psychol Med 37:1075-1084.

Schaer M, Cuadra MB, Tamarit L, Lazeyras F, Eliez S, Thiran JP (2008): A surface-based approach

to quantify local cortical gyrification. IEEE Trans Med Imaging 27:161-170.

Schaer M, Ottet M-C, Scariati E, Dukes D, Franchini M, Eliez S, Glaser B (2013): Decreased

frontal gyrification correlates with altered connectivity in children with autism. Front Hum

Neurosci 7, doi: 10.3389/fnhum.2013.00750.

Schmitt A, Malchow B, Hasan A, Falkai P (2014): The impact of environmental factors in severe

psychiatric disorders. Front Neurosci 8. doi: 10.3389/fnins.2014.00019.

Seitz J, Buhren K, von Polier GG, Heussen N, Herpertz-Dahlmann B, Konrad K (2014):

Morphological changes in the brain of acutely ill and weight-recovered patients with anorexia

nervosa. A meta-analysis and qualitative review. Z Kinder Jugendpsychiatr Psychother 42:7-

18.

Sun T, Hevner RF (2014): Growth and folding of the mammalian cerebral cortex: from molecules

to malformations. Nat Rev Neurosci 15:217-232.

Tchanturia K, Lounes N, Holttum S (2014): Cognitive remediation in anorexia nervosa and related

conditions: a systematic review. Eur Eat Disord Rev 22:454-462.

Tenconi E, Santonastaso P, Degortes D, Bosello R, Titton F, Mapelli D, Favaro A (2010): Set-

shifting abilities, central coherence, and handedness in anorexia nervosa patients, their

unaffected siblings and healthy controls: exploring putative endophenotypes. World J Biol

Psychiatry 11:813-823.

Tenconi E, Santonastaso P, Monaco F, Favaro A (2014): Obstetric complications and eating

disorders: A replication study. Int J Eat Disord epub ahead of print 26 May 2014; doi:

10.1002/eat.22304.

Titova OE, Hjorth OC, Schiöth HB, Brooks SJ (2014): Anorexia nervosa is linked to reduced brain

structure in reward and somatosensory regions: a meta-analysis of VBM studies. BMC

Psychiatry 13:110. doi: 10.1186/1471-244X-13-110.

Treasure J, Claudino AM, Zucker N (2010): Eating disorders. Lancet 375:583-593.

Van den Eynde F, Suda M, Broadbent H, Guillaume S, Van den Eynde M, Steiger H Israel M,

Berlim M, Giampietro V, Simmons A, Treasure J, Campbell I, Schmidt U (2012): Structural

magnetic resonance imaging in eating disorders: a systematic review of voxel-based

morphometry studies. Eur Eat Disord Rev 20:94-105.

Van Essen DC (1997): A tension-based theory of morphogenesis and compact wiring in the central

nervous system. Nature 385:313-318.

Wagner A, Greer P, Bailer UF, Frank GK, Henry SE, Putnam K, Meltzer CC, Ziolko SK, Hoge J,

McConaha C, Kaye WH (2006): Normal brain tissue volumes after long-term recovery in

anorexia and bulimia nervosa. Biol Psychiatry 59:291-293.

White T, Hilgetag CC (2011): Gyrification and neural connectivity in schizophrenia. Dev

Psychopathol 23:339-352.

White T, Su S, Schmidt M, Kao C-Y, Sapiro G (2010): The development of gyrification in

childhood and adolescence. Brain Cogn 72:36-45.

Van Kuyck K, Gerard N, Van Laere K, Casteels C, Pieters G, Gabriels L, Nuttin B (2009): Towards

a neurocircuitry in anorexia nervosa: Evidence from functional neuroimaging studies. J

Psychiatr Res 43:1133-1145.

Figure 1 - Vertex-wise comparisons between anorexia nervosa patients and healthy women. General

linear models with age and total intracranial volume were used, applying the Monte Carlo method

to control for multiple comparison and using a cluster-wise threshold for inclusion of p < 0.01. Left

hemisphere: a cluster of 6.761 mm2 with a vertex of maximum difference (p=0.0002) in the

postcentral gyrus (-36, -36, 54). Right hemisphere: a cluster of 1.419 mm2 with a vertex of

maximum difference (p=0.002) in the superior frontal gyrus (21, 9, 53). No cluster showed

significant results in the opposite direction.

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Figure 2 - Vertex-wise comparisons between anorexia nervosa patients who recovered at 3-year

follow-up and those who still had an eating disorder. General linear models with age, duration of

illness and total intracranial volume were used, applying the Monte Carlo method to control for

multiple comparison and using a cluster-wise threshold for inclusion of p < 0.05. Left hemisphere: a

cluster of 2.892 mm2 with a vertex of maximum difference (p=0.0018) in the superior parietal

cortex (-17, -49, 62). Right hemisphere: a cluster of 3.504 mm2 with a vertex of maximum

difference (p=0.0004) in the postcentral gyrus (27, -38, 51). No cluster showed significant results in

the opposite direction.

Table 1 - Baseline characteristics of the 3 groups

|  | AN patients (n=38) | Recovered AN patients (n=20) | Healthy women (n=38) | AN vs. HW | recAN vs. HW |
|---|---|---|---|---|---|
|  | mean (SD) | mean (SD) | mean (SD) | t (p) | t (p) |
| Age | 26.1 (7.2) | 26.3 (7.1) | 25.3 (6.3) | 0.54 (0.59) | 0.59 (0.56) |
| Age at onset | 18.3 (5.1) | 17.7 (3.2) | = | = | = |
| Duration of illness (months) | 78.6 (81.3) | 45.7 (65.0) | = | = | = |
| Duration of recovery (months) | = | 45.4 (46.8) | = | = | = |
| Baseline BMI | 15.8 (1.8) | 19.6 (1.6) | 21.7 (2.9) | 10.51 (<0.001) | 2.91 (0.005) |
| Weight loss* | 7.1 (2.8) | 5.2 (3.1) | 3.4 (1.7) | 7.01 (<0.001) | 2.95 (0.005) |
| Lowest BMI | 14.0 (1.8) | 15.7 (1.4) | 19.8 (2.5) | 11.56 (<0.001) | 6.71 (<0.001) |
| Education | 14.2 (2.2) | 14.2 (2.7) | 15.5 (2.3) | 2.44 (0.02) | 1.97 (0.05) |
| Edinburgh laterality index | 57.2 (37.6) | 60.6 (35.2) | 55.1 (42.0) | 0.23 (0.82) | 0.50 (0.62) |
| Drive for thinness | 9.9 (6.1) | = | 2.3 (4.2) | 6.22 (<0.001) | = |
| Depression | 1.4 (0.8) | = | 0.7 (0.6) | 4.06 (<0.001) | = |
| Trait anxiety | 56.6 (9.7) | = | 39.3 (9.6) | 7.82 (<0.001) | = |

* weight loss is defined as the difference between highest and lowest lifetime body mass index

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Table 2 - Correlations between hemispheric gyrification index and clinical variables, and differences in clinical characteristics in anorexia nervosa patients according to outcome at 3-year follow-up

| | Correlation with left lGI | Correlation with right lGI | AN patients with recovery at follow-up (n=13) | AN patients without recovery (n=24) | |
|---|---|---|---|---|---|
| | rho (p) | rho (p) | mean (SD) | mean (SD) | z (p) |
| Age | -0.11 (0.51) | -0.16 (0.33) | 25.5 (6.8) | 26.7 (7.5) | 0.33 (0.74) |
| Age at onset | 0.20 (0.23) | 0.01 (0.94) | 20.8 (6.5) | 17.1 (3.8) | 2.26 (0.02)** |
| Duration of illness (months) | -0.36 (0.02)** | -0.30 (0.07) | 40.0 (46.2) | 101.2 (89.8) | 2.10 (0.04) |
| Baseline BMI | 0.03 (0.87) | 0.01 (0.80) | 14.9 (1.75) | 16.2 (1.6) | 2.23 (0.03) |
| Weight loss* | -0.10 (0.54) | 0.01 (0.94) | 7.0 (2.4) | 7.2 (3.1) | 0.19 (0.85) |
| Lowest BMI | 0.31 (0.06) | 0.17 (0.31) | 14.4 (2.0) | 13.7 (1.7) | 1.13 (0.26) |
| Drive for thinness | -0.02 (0.91) | -0.07 (0.70) | 12.2 (6.1) | 8.3 (5.6) | 1.82 (0.07) |
| Depression | 0.02 (0.92) | -0.02 (0.89) | 1.38 (0.68) | 1.39 (0.84) | 0.03 (0.98) |
| Trait anxiety | -0.26 (0.12) | -0.14 (0.40) | 55.6 (10.9) | 57.0 (9.3) | 0.40 (0.69) |
| Duration follow-up (ys) | = | = | 3.2 (0.6) | 3.5 (0.5) | 1.48 (0.14) |

|  | Correlation with left lGI | Correlation with right lGI | AN patients with recovery at follow-up (n=13) | AN patients without recovery (n=24) |  |
|---|---|---|---|---|---|
| Final BMI | 0.20 (0.24) | 0.20 (0.23) | 19.6 (2.1) | 17.7 (4.3) | 3.66 (<0.001)** |

AN: anorexia nervosa; lGI: local gyrification index; BMI: body mass index

* weight loss is defined as the difference between highest and lowest lifetime body mass index

** according to false discovery rate method, differences are significant at p<0.027 and correlations at p<0.026

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Table 3 - Outcome predictive ability of gyrification indices (logistic regression analyses)

| Local gyrification indices* | OR (95% CI) | | OR (95% CI) adjusted for age, age at onset, body mass index and duration of illness | |
|---|---|---|---|---|
| | OR (95% CI) | % individuals correctly classified | OR (95% CI) | % individuals correctly classified |
| Overall lh LGI | 3.4 (1.1-10.3) p = 0.032 | 67.6% | 5.2 (1.1-23.9) p = 0.033 | 81.1% |
| Overall rh LGI | 2.9 (1.1-7.7) p = 0.033 | 73.0% | 3.8 (1.1-13.0) p = 0.031 | 83.8% |
| Left superior parietal cluster LGI | 2.4 (1.2-4.7) p = 0.014 | 67.6% | 5.4 (1.4-20.1) p = 0.012 | 81.1% |
| Right postcentral cluster LGI | 2.3 (1.2-4.5) p = 0.016 | 70.3% | 4.1 (1.4-12.5) p = 0.012 | 81.1% |

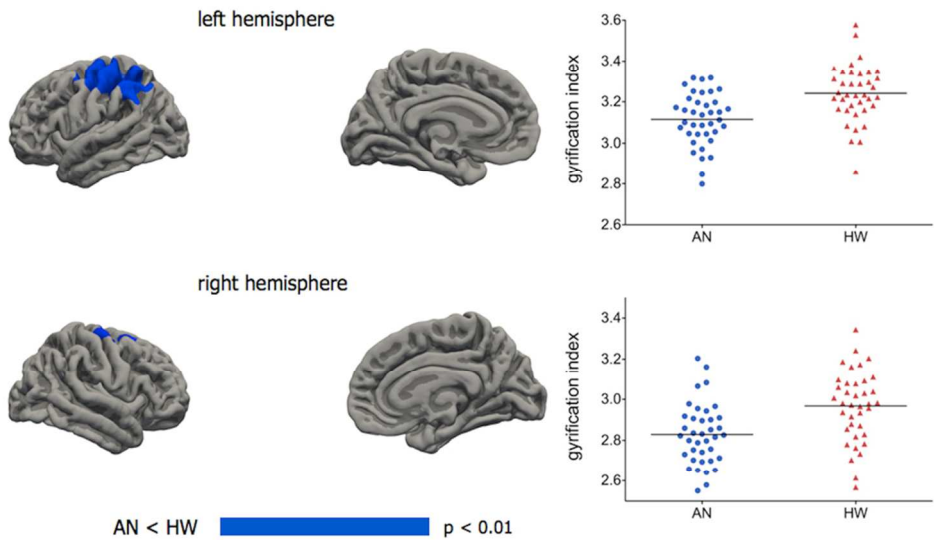* all LGI indices were multiplied by a factor of 10

Figure 1 - Vertex-wise comparisons between anorexia nervosa patients and healthy women. General linear models with age and total intracranial volume were used, applying the Monte Carlo method to control for multiple comparison and using a cluster-wise threshold for inclusion of $p < 0.01$. Left hemisphere: a cluster of 6.761 mm2 with a vertex of maximum difference (p=0.0002) in the postcentral gyrus (-36, -36, 54). Right hemisphere: a cluster of 1.419 mm2 with a vertex of maximum difference (p=0.002) in the superior frontal gyrus (21, 9, 53). No cluster showed significant results in the opposite direction.
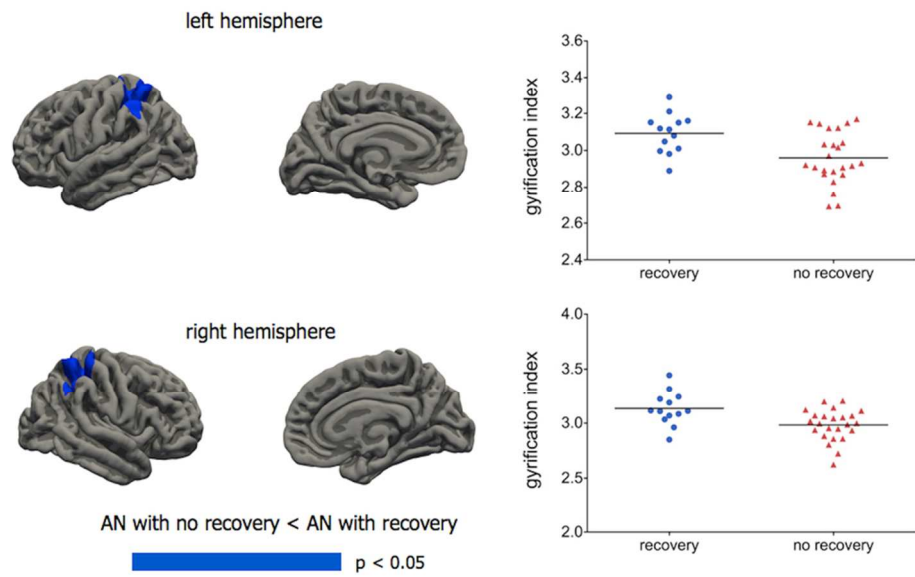
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Figure 2 - Vertex-wise comparisons between anorexia nervosa patients who recovered at 3-year follow-up and those who still had an eating disorder. General linear models with age, duration of illness and total intracranial volume were used, applying the Monte Carlo method to control for multiple comparison and using a cluster-wise threshold for inclusion of $p < 0.05$. Left hemisphere: a cluster of 2.892 mm2 with a vertex of maximum difference (p=0.0018) in the superior parietal cortex (-17, -49, 62). Right hemisphere: a cluster of 3.504 mm2 with a vertex of maximum difference (p=0.0004) in the postcentral gyrus (27, -38, 51). No cluster showed significant results in the opposite direction.

91x54mm (300 x 300 DPI)