Global-To-Local Segmentation and Genotypic Analysis Of Brain Shape Asymmetry

Vasileios Lemonidis

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Thesis supervisors:
Prof. Peter Claes
Prof. Isabelle Cleynen

Mentors:
MSc. Meng Yuan
MSc. Seppe Goovaerts

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Preface

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The Author
1 January 2010

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Abbreviations

3D three-dimensional

ANOVA analysis of variance

CCA canonical correlation analysis

CNS central neural network

D-V dorsal-ventral

DA directional asymmetry

DK Desikan-Killiany

DNA deoxyribonucleic acid

DOF degrees of freedom

FA fluctuating asymmetry

GWAS genome wide association studies

HSC hierarchical spectral clustering

L-R left-right

LD linkage disequilibrium

MRI magnetic resonance imaging

mvGWAS multivariate genome-wide association study

NPC neuroepithelial cell

R-C rostral-caudal

RGC radial glial cell

RNA ribonucleic acid

RSS residual sum of squares

SNP single nucleotide polymorphism

Abstract

Overall purpose of this thesis is to complement the existing bibliography on the detection and examination of the genetic associations of brain shape asymmetry. Asymmetry components are computed based on the brain magnetic resonance imaging (MRI) dataset provided by UK Biobank database. A data-driven approach is followed, where the brain surface is partitioned in an unsupervised manner, through hierarchical spectral clustering (HSC), a technique that allows for a coarse-to-fine segmentation. Aggregated asymmetry measurements are retrieved from the segments, whose genetic correlation is examined through a multivariate genome-wide association study (mvGWAS) statistical analysis. Recognized significant single nucleotide polymorphisms (SNPs) are then analyzed individually or in groups, through comparison with existing results and databases. The genetic overlap with neurodevelopmental disorders and traits, that have been reported to exhibit phenotypic associations with brain structure asymmetry, such as Autism, Alzheimer's Disease or intelligence, are examined. Functional annotations of variants associated with the genes where significant SNPs were detected are constructed, offering an insight into the functional reasoning behind the brain shape asymmetry existence. Further comparisons with other past human phenotypic characteristics studies are lastly applied.

Chapter 1

Introduction

1.1 Bilateria lineage

Cerebral bilateral symmetry is a universal quality of organisms belonging to the Bilateria lineage [9, 10], the phylum incorporating all species with a single plane of symmetry, in contrast with their sister group, Cnidaria (Figure 1.1). Bilateral symmetry is a byproduct of the activity of two separate developmental processes, that produce two axes of polarity [18], and therefore a symmetry plane; the formation of a primary body axis, that corresponds to the long anatomical dimension of the animal, called rostral-caudal (R-C) (i.e. head-to-tail), primarily dictated by highly conserved controlled activation of HOX genes during cell differentiation; the shaping of a secondary body axis, orthogonal to R-C, named dorsal-ventral (D-V) (i.e. back-to-front), attributed to a variety of genes, such as the chromatin organizer CTCF, the left-right determination factor Nodal and central HOX genes [22]. The remaining axis, left-right (L-R), is the one along which the symmetry pattern is manifested. On account of the high biodiversity that bilateria group includes, only the subgroup of vertebrates is examined in the following literature study. In addition, any reference to symmetry or asymmetry from now on corresponds to the L-R direction, unless explicitly mentioned.

This study makes an effort to statistically identify the genetic origins of a complex structural phenotype. Hence, examining, based on existing research, the main brain developmental stages is essential to discern the processes that induce bilateral symmetry. An important vertebrates (and bilateria) common characteristic is the germ line **triploblasticity**: the embryo begins as a flat disk, through a process called **gastrulation**, with three distinct cell layers; **endoderm**, **mesoderm**, and **ectoderm** [15]. Of significance in the neural system formation is the ectoderm, which is initially equivalent to one of the flat disk sides. Under the context of this study, although a fact not directly connected to the brain cortex, it is necessary to mention that the perfect bilateral symmetry pattern appears to break even before gastrulation. In Xenopus (frog species) embryos, during fertilization and the initial 4-cell cleavage of the fertilized egg , the **cytoskeleton microtubules** appear to asymmetrically localize the ion channels proteins, whose RNA has been passed on by

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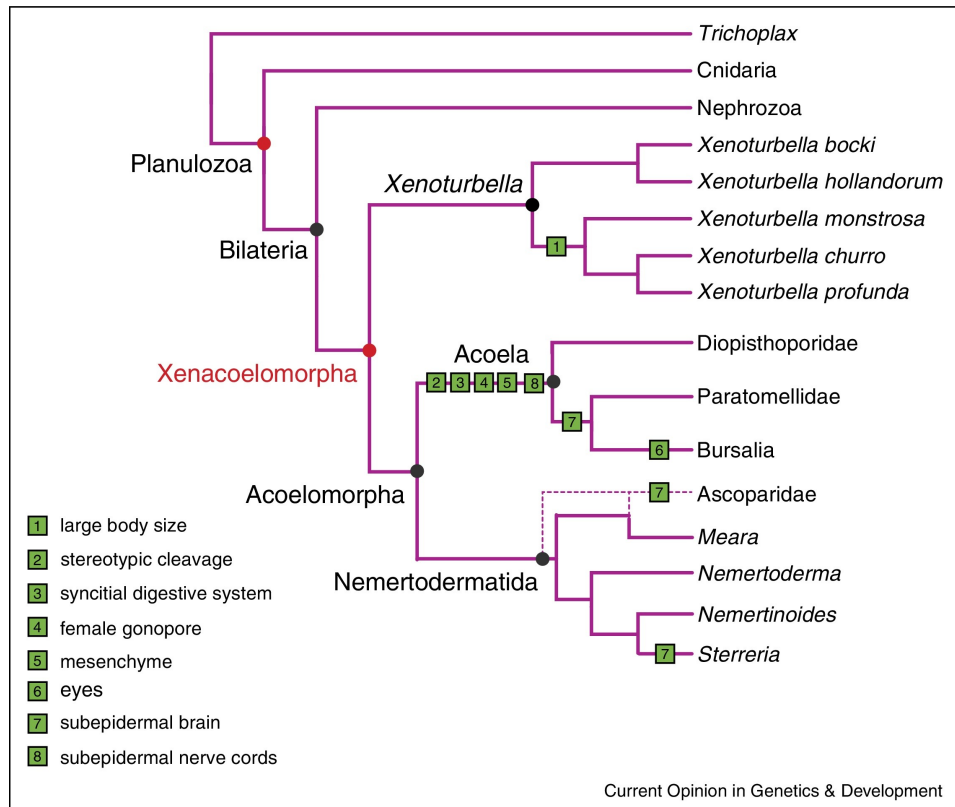
FIGURE 1.1: Species phylogenetic tree subset, displaying bilateria clade, its sister clade, Cnidaria, and the direct children[23]. Of great importance on the evolutionary studies of bilateral symmetry is the Xenacoelomorpha clade.

the mother, with a preference for the right side of the complex [3]. Chicks embryos also exhibit a similar pattern. The occurrence of asymmetry at this extremely early time point underlines the significant role it has on the embryo development, species fitness, and, concomitantly, the conservation potential of this trait drivers [4]. Another cellular component that is considered to enhance asymmetry, during gastrulation, is the motile cilia, hair-like organelles on the cell surface with the ability to beat [21]. Their movement is by construction asymmetric, causing a leftward flow of extraembryonic fluid and, subsequently, asymmetric distribution of exogenously introduced proteins [38]. Both studied phenomena point to early initiation of asymmetric genes expression.

1.2   Symmetry during CNS formation

Shortly after gastrulation, the disk folds, in a way that the central region of the ectoderm, called neural plate, forms a tube-like shape, the **neural tube**, which acts as the neural system precursor, under a process called **neurulation**. All bilateria have a central neural network (CNS), which entirely develops from the neural tube walls [15].

The next pivotal step in the brain development, **differentiation**, leads to the creation of three distinct compartments along rostral-caudal (R-C) axis, at the neural tube rostral end, the **prosencephalon** (forebrain), which develops into the brain cerebrum, the **mesencephalon** (midbrain), and the **rhombencephalon** (hindbrain), that is later attached to the spinal cord in vertebrates. For the subsequent mechanisms and terminology to be compatible with human cerebrum related literature, the focus is shifted on mammals phylum and, spatially, on the prosencephalon. The differentiation proceeds, with two pairs of lumps extruding symmetrically from the prosencephalon, the **telencephalic** vesicles, the predecessors of cerebral region, and the optic vesicles, the precursors of optic nerves and retinas, while the central remaining structure is called **diencephalon** [14]. The telencephalic vesicles continue to grow, expanding also caudally and in parallel with the diencephalon, gradually assuming the form of the two hemispheres, while a new pair of vesicles appears on the rostral part of the diencephalon, giving rise to the **olfactory bulbs**. The neural tube shape also reacts to the changes, forming four distinct **ventricles** along the neural tube, with two of them, named lateral ventricles, being mirrored inside each of the telencephalic vesicles. That is the earliest stage where asymmetry is noted in an anatomic level inside the human brain, during the end of the first trimester of gestation [2]. Specifically, the choroid plexus, a specialized cell network that lies inside the ventricles, attached to the diencephalon, and produces most of the **cerebrospinal fluid** in the CNS, develops asymmetrically in each lateral ventricle. The cerebrospinal fluid is of great value for the developing brain, as the main source of nourishment, waste removal and protection [44]. Such an asymmetry manifestation in a macroscopic level, therefore, may be the progenitor of other forms of asymmetry at a later developmental stage [42], even at the brain surface. Cerebral bilateral symmetry therefore begins breaking down during fetal development, producing an asymmetric brain (Figure 1.2), and giving rise to partial functional disassociation, called **brain lateralization**. Lateralization becomes visible when examining organisms' behavior, with the most studied trait in humans being handedness and language [42, 10]. To better understand why and how the inner functions are related with the external brain cortex development, the underlying cellular processes of **neurogenesis** and **neuron migration**, active throughout differentiation, need to be identified, before introducing the reader to the anatomy of the fully grown brain. For this purpose, a further focus on primates phylum is needed, given the differences exhibited when comparing different mammals, such as rodents[33].

1.3 Neurogenesis and Neuronal Migration

The cells initially comprising the neural tube walls are named neuroepithelial cells (NPCs), and exhibit similar properties with stem cells, that is limited multipotency (i.e. they can differentiate into multiple cell types) and limited self-renewing (i.e. they can divide symmetrically into new NPCs a finite number of times), while also properties of epithelial cells, that is polarity (i.e. asymmetric cellular organization, with distinct basal and apical surfaces) and attachment (i.e. junctions tightly connect
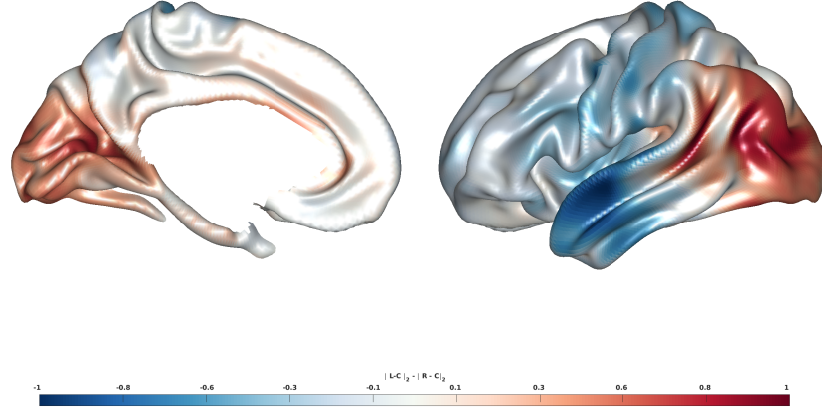
$$| \text{L-C} |_2 - | \text{R - C} |_2$$

Figure 1.2: Illustration of human cerebrum brain asymmetry. Normalized differences of the distances of landmarks from the center of mass of each averaged hemisphere surface. See section 1.6 for more details on the preprocessing.

adjacent cells) [20]. This cells array is contained between the basal and apical laminae, lipid membranes lateral to each other, with the apical lamina facing the neural tube lumen [1], and the cells being radially distributed. During anatomical differentiation, around the 7th gestational week in humans [37], self-renewing is activated, leading to cells proliferation and CNS bilateral expansion, while attachment is hindered, gradually exchanging the NPCs with RGCs, the fate-restricted progenitors of neurons, marking the initiation of **neurogenesis**[20]. A RGC acts as the main building block of the brain, from which a single neuron or a neural progenitor, that later divides symmetrically in neurons, is generated. RGC's pivotal role does not end here. As it can be seen in Figure 1.3, RGCs are stretched during development, with processes connected to the surface of neural tube successor ventricles and to the outer cortical region surface, forming thread-like scaffolds. Newly formed neurons, generated from the RGCs main, oval body, which remains close to the ventricles, use this structure as a guide to move towards the outer region of the cortex, under a process named **neuronal migration**[40]. This type of movement actually implies that the newly formed neurons head towards the brain surface, building the brain in an inside first, outside last fashion [33]. At later stages of gestation, around week 19 for humans, new studies have shown that a morphological transition is observed, where the majority of RGCs stops being attached to the **pial surface**, the outer surface of the brain, limiting the migration ability of neurons [37] and affecting the way new layers are formed. Human neurogenesis extends to the third gestation trimester, being suppressed in case of premature birth [32]. Postnatal neurogenesis is therefore presumed to be quite limited for primates [13], despite the fact that the postnatal brain dramatically increases in size , with that attributed to a rapid increase in neurons connections and glial cells (i.e. cells that provide physical and metabolic
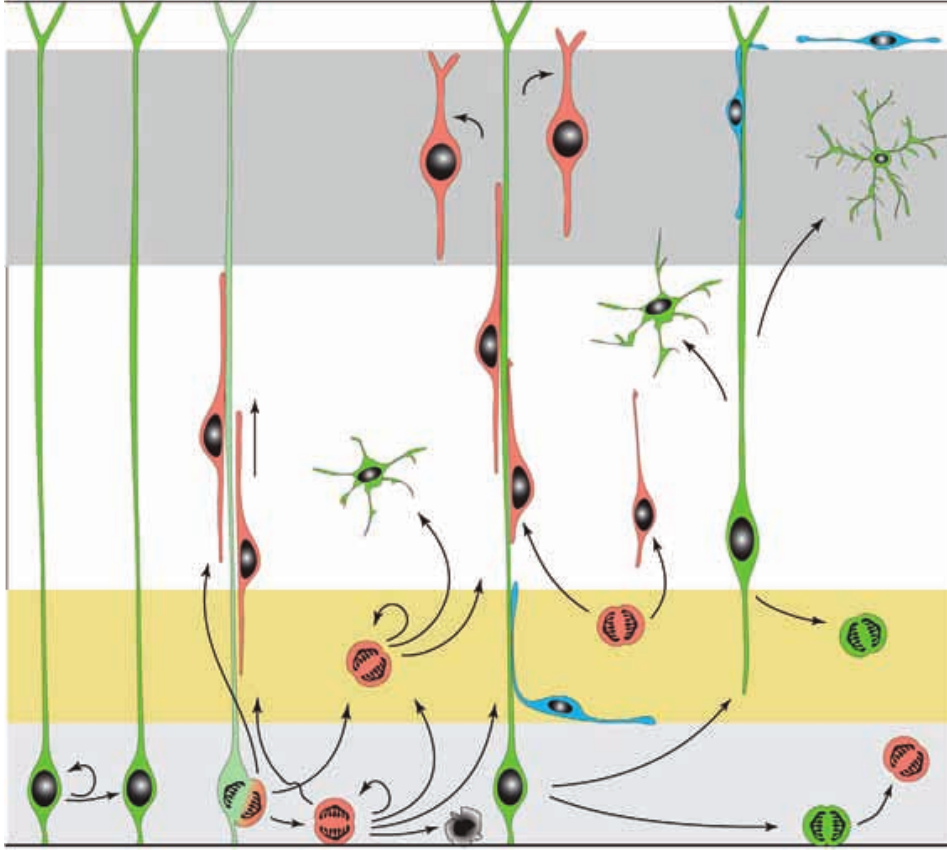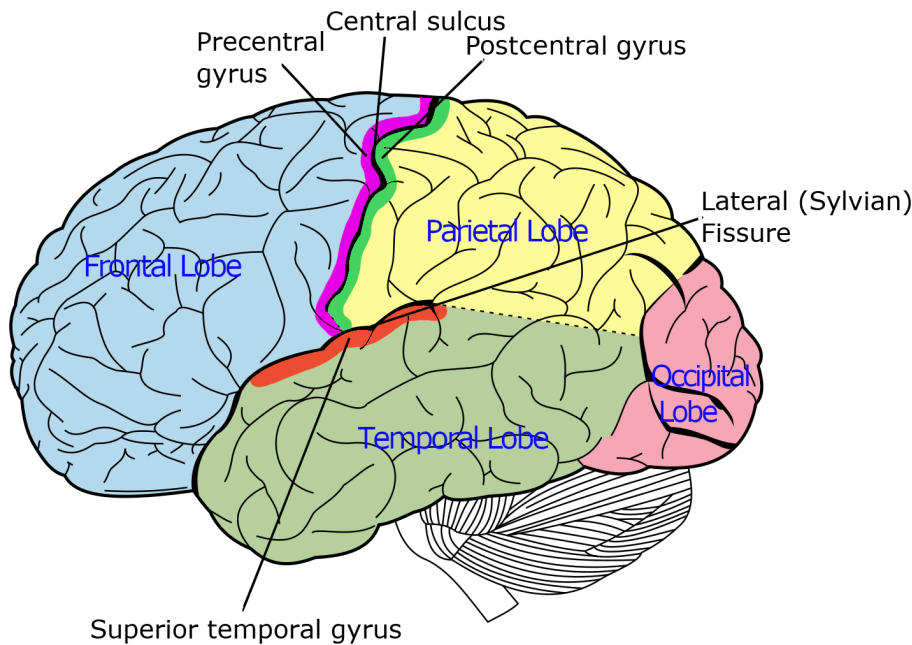
FIGURE 1.3: Illustration of a classical model of radial glial cells complex nonlinear division processes and neuronal migration [40]. From left to right: NPCs (green) originally divide symmetrically; During differentiation, NPCs become RGCs, which divide asymmetrically, generating neurons or neural progenitor cells (orange). Neural progenitor cells eventually divide symmetrically into neurons. The majority of neurons in humans is produced by neuronal progenitors. A part of the generated neurons migrate radially towards the cortical plate, by attaching on the RGCs projections; Eventually, after brain maturation, most RGCs in humans undergo apoptosis (i.e. cell death) or generate neurons-supporting cells, such as astrocytes.

support to neurons) number [12]. The environmental factors that may affect brain symmetry are mainly detected before or during birth, with epigenetics and birth complications appearing to be mostly correlated with handedness [42, 6]. In general, though, the more complex the phenomenon and the closer it is to humans, the higher the uncertainty and the greater the ethical restrictions. Only recently, non-invasive imaging and transcriptomic techniques have given further details regarding the brain development sequence, with genetic studies indirectly identifying the landscape of the underlying genes that affect different brain regions formation and symmetry [6]. Moving on the literature study path and getting closer to the studied phenotype, the fully grown human brain cerebrum is subsequently anatomically described.

1.4   The adult human cerebrum anatomic and functional atlas

Human cerebrum is the center of sensations and thinking. As aforementioned, it is produced entirely from the telencephalon during fetal development, with the telencephalic vesicles ending up becoming the two hemispheres, that remain connected through what is known as the **Corpus callosum**. The human cerebrum outer covering surface is called **cerebral cortex**, the region on which the current study focuses. The human cerebrum appears distinctly different from other organisms, mainly due to the **sulci** (i.e. grooves) and **gyri** (i.e. bumps), being the result of the tremendous expansion of the cerebral cortex surface area during fetal development, folding and wrinkling in order to fit the skull [16]. The precise pattern of gyri and sulci varies significantly across populations, rendering the brain surface unique per individual. Under a biopsy dissection or a magnetic resonance imaging (MRI) scan, the cerebrum appears to consist of two distinctly colored types of matter, implying changes in composition and consistency; the gray matter, at the outer part of the cerebrum, which contains the cell bodies, dendrites and the axon terminals, where all synapses are, and the white matter, at the inner part, made up of myelinated (i.e. biologically insulated) axons, which connect different parts of gray matter to each other (**??**). Protective layers on top of the gray matter, called **meninges**, ensure that the brain does not come in contact with the outer bone, with the one attached on the gray matter named **pial surface**. In this study, the midthickness surface is examined, a term referring to the surface halfway between the pial and white matter surface.

Efforts of partitioning the brain have been numerous throughout the years of medicine, with diverse resolution and purpose. The reader is directed to the figures captions for a detailed description of each such segmentation. Crudely, the cerebrum hemisphere is divided into lobes, that are named, by convention, after the bones of the skull that lie over them (**??**). There is also a rich nomenclature regarding the gyri and sulci, with deep grooves being identified as **fissures** (**??**). A more detailed approach is based on the identification of the functional processes that take place in each part of the cortex (**??**). In later years, with the advance of imaging methods, new maps have been manufactured, to automatically partition the extracted three-dimensional (3D) cortical projection, based on morphological characteristics. One such gyral-based atlas, Desikan-Killiany (DK), is derived from the changes in curvature under an expert-driven model of gyri locations [11] and provides automatic **cortical parcellation**.

Central sulcus
Precentral gyrus
Postcentral gyrus
Frontal Lobe
Parietal Lobe
Lateral (Sylvian) Fissure
Occipital Lobe
Temporal Lobe
Superior temporal gyrus

1.5 Genetic studies on cerebral symmetry

1.6 Phenotypic trait analysis

1.6.1 Summary on cortex anatomy

1.6.2 Dataset Description

1.6.3 Shapes Normalization

MRI output is affected by the subject positioning and technical error. Volumetric differences also increase the level of discrepancies among MRI samples. To remedy positioning and volume deviations, a normalization is required, achieved by projecting each subject onto **Kendall shape space** [26]. TO BE CONTINUED

1.6.4 Asymmetry Components

TO BE CONTINUED Of primary interest in this work is directional asymmetry (DA), the asymmetric component that arises by comparing a single individual's hemispheric surfaces landmarks differences, computed through a process of alignment, reflection and subtraction of the landmarks pairs. DA captures information about anatomic characteristics, such as the overall counterclockwise torque, named 'Yakovlevian torque' [30], that is observed in humans between the right and left hemisphere (Figure 1.4). Past studies have shown that abnormal DA may be an indication of certain diseases. The lack of it may imply schizophrenia predisposition [41]. Any

significant abnormalities may be indicative of other psychiatric disorders, such as autism or developmental language disorder [24][29].

Statistical analysis

Bilateral asymmetry is mainly described using three components in literature [27][47]; directional asymmetry (DA), the focus of this study, corresponds to the hemispheric side effect; antisymmetry, which is related to the effect where sidedness is random in a population (i.e. left-right randomly switches to right-left), is not observed in the human cerebral cortex, in contrast to other internal organs positions, or organisms [35]; fluctuating asymmetry (FA), encompasses any random developmental and environmental effects, that cannot be explained with the existing knowledge. The observed deviations can be statistically linearly modeled as products of two effects, the hemisphere side studied and the individual specimen analyzed, as well as their interaction [27]. Formally, based on [45] assuming the presence of replications of the observation per individual, to account for technical error, a mixed linear model representing the aforementioned dependencies is defined as:

$$Y_{ijk} = \mu + \beta + I_i + S_{ij} + E_{ijk} \tag{1.1}$$

where Y_{ijk} is the phenotype of the i-th individual, from the j-th side, under the k-th replication, μ and β are the fixed intercept and fixed side effect respectively, $I_i \sim \mathcal{N}(0, \sigma_{ind}^2)$ is the random individual effect, $S_{ij} \sim \mathcal{N}(0, \sigma_{FA}^2)$ is the random side and individual specific effect, matched to FA, and $E_{ijk} \sim \mathcal{N}(0, \sigma_{ME}^2)$ is the measurement error. Replications are necessary in such a study, in order to differentiate FA effect from the measurement error. Given this definition, a way to measure the statistical significance is through an F-test applied on 2-way analysis of variance (ANOVA), to relate the residual sum of squares (RSS) ratios of effects to observable error terms, and of fluctuating effect to the measurement error. Extra care needs to be given on the determination of the degrees of freedom (DOF) of each term, given the preprocessing applied to bring the hemispheres surfaces into Kendall shape space. Those are extracted from the rigorous work in [27]. Given that the analysis is performed on a pair of symmetric objects, and not on a single symmetric object, this configuration is named matching asymmetry analysis. In order to avoid further explicit assumptions, regarding the distribution of TO BE CONTINUED

1.6.5 Phenotypic partitioning

Hierarchical spectral clustering (HSC) is an unsupervised method of iterative partitioning, that makes use of the distance matrix eigenvectors [36]. It results into a binary tree structure (i.e. each parent shape is partitioned into two children). In the current study, a level-4 partitioning is performed, resulting into 31 partitions. Subsequently, they are transformed to the corresponding principal components that explain 80% of the variance, for reasons of further dimensionality reduction. TO BE CONTINUED
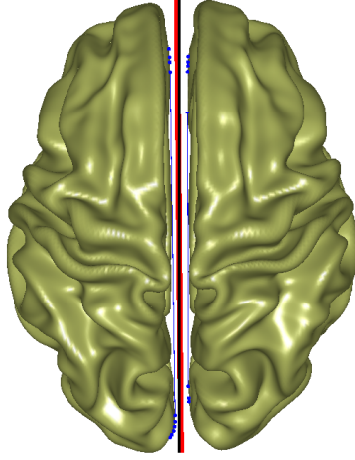
FIGURE 1.4: Illustration of the Yakovlevian torque. Displayed by a red line rotated counterclockwise 0.51 degrees in relation to the perfectly vertical black line, as calculated by using the average angle of the longest edges (in blue) of the convex hulls of the horizontal plane projection of each hemisphere midthickness surface, after a scaling and alignment process, across the studied population.

1.7 Breaking the complexity into parts

TO BE EXPANDED

The present work evaluates the brain asymmetry genetic landscape in a coarse-to-fine segmentation, through the application of HSC[36], discussed in subsection 1.6.5. The technique has been used in a number of different related studies [8][34], yielding results that are in accordance with the underlying anatomic features. The main reason behind this partitioning is the intrinsic complexity of the studied phenotype, eliciting expected differences in the genomic profiles of each cerebral cortex region. The basic assumption made is that topologically close landmarks share similar genetic background. In general though, this type of distance-based clustering is governed by the least quantity of assumptions, regarding the shape or form of the cluster [48]. The partitions' genetic juxtaposition is valuable for identifying which regions share similar significant genetic loci, highlighting the corresponding genes contribution, or showcasing the specialization of certain regions that share little to no similarities with their neighbors. Identifying the latter provides a way of mapping the developmental activation of each locus, bringing forth the opportunity to augment the results of related developmental studies [46].

1.8 Searching for the origin

TO BE EXPANDED

The genomic studies are performed under the framework of single nucleotide polymorphism (SNP)-by-SNP canonical correlation analysis (CCA). The goal is to incorporate multi-allelic SNPs and, more importantly, multivariate phenotype, in a single hypothesis test per SNP, that is whether the phenotype is significantly correlated with each analyzed SNP. In general, there is an abundance of strategies on how to perform multivariate genome wide association studies (GWAS), ranging from direct methods, that approximate the inputs relation either in an unbiased manner or making certain educated guesses, to more complex techniques, that increase statistical power by transforming the inputs, at the expense of explanatory ability [19]. There are also methods that are based on the meta-analysis of outcomes from univariate studies, commonly used to juxtapose experiments from separate sources, for which the original data is absent or the exact replication of the study is arduous [7]. Which approach performs best mainly lies on the dataset properties and the nature of the scientific question. Factors such as low sample size [43], genes pleiotropic effects [17] or within-study variation [25] tend to handicap the statistical modeling and increase the type I and II errors of the corresponding hypothesis tests. In this study, CCA was primarily chosen due to the high capacity in efficiently reducing the inputs dimensionality while preserving most information regarding their correlation. Diverse experiments, analyzed in chapter 5, have been applied to identify the method that gives high fidelity results, consistent with relevant literature. The analysis outcome requires further processing, as explained in section 5.1, to account for the main weakness of this method, that it does not consider the SNP-to-SNP effect, tackled using as proxy the notion of linkage disequilibrium (LD), and subsequently to topologically and functionally enhance the filtered findings. Once this additional step has been performed, a cross-traits analysis is applied, described in section 5.2, where the DA genetic signature is compared with the signatures of phenotypic traits, analyzed in a similar study [34], the cerebral and facial shapes.

1.9 Data description

In this study, targeted on humans, a cross section between the dependent cerebral asymmetry and the independent genetic factors is performed, in an effort to discover affiliated genetic regions and provide a novel understanding of the related genes cooperation. With the advent of technology capable to collect and process genomes from different individuals in relatively high speed, vast databases have been constructed. One of the main players in the data collection has been UK Biobank; a large-scale database from a randomized consortium of 500,000 individuals, whose genome has been collected, from whom 48,000 subjects had also participated in brain MRI collection process, as of December 2020 [31]. In this thesis, we exploit this newly acquired dataset to identify the key loci that are related to the human brain surface symmetry. Only healthy self-proclaimed white European individuals were considered.

1.10 Novelties based on related literature

Due to the biological importance of cerebral bilateral asymmetry, it is a subject that has been rigorously studied from multiple viewpoints.

1.10.1 Evolution

From an evolutionary stand, it is extremely rare for the right conditions to occur, in order for any soft tissue specimen to be preserved, across a considerable amount of time. The only known way is through mineralization [39]. This fact renders a mammal's ancestor brain almost impossible to retrieve. Nevertheless, endocranial imprints have been used as a proxy to describe the relationships between hominids and their ancestors [5][35]. The reason behind this phenotypic delegation is purely practical. The brain size and shape follow the container volume restrictions. Although such studies support the theory of propagating asymmetry among studied individuals, with the most evident signs of DA in human skulls, little information about the surface shape can be retrieved, as only the convex hull shape of the brain can be delineated from such process. Through the association of brain asymmetry with DNA, a universal code among organisms, it becomes possible to deploy tools used by evolutionary geneticists, to identify the phylogenetic tree of this complex trait, locating conserved regions among organisms and their predicted divergence in time, under a pleiotropic model [28].

1.10.2 Clinical studies

Chapter 2

Dataset Description

2.1 General

A large dataset of 19,654 individuals was used as the main, discovery dataset, while a smaller one, coming from a different batch, of 16,342 individuals was used as a replication dataset during GWAS.

2.2 Phenotype

2.3 Genome

Chapter 3

Data Preprocessing

Chapter 4

Asymmetry Phenotypic Analysis

Chapter 5

Asymmetry Genetic Analysis

Chapter 6

Discussion

Bibliography

[1] E. Aaku-Saraste, B. Oback, A. Hellwig, and W. B. Huttner. Neuroepithelial cells downregulate their plasma membrane polarity prior to neural tube closure and neurogenesis. *Mechanisms of Development*, 69(1-2):71–81, dec 1997.

[2] R. S. Abu-Rustum, M. F. Ziade, and S. E. Abu-Rustum. Reference Values for the Right and Left Fetal Choroid Plexus at 11 to 13 Weeks. *Journal of Ultrasound in Medicine*, 32(9):1623–1629, sep 2013.

[3] S. Aw, D. S. Adams, D. Qiu, and M. Levin. H,K-ATPase protein localization and Kir4.1 function reveal concordance of three axes during early determination of left–right asymmetry. *Mechanisms of Development*, 125(3-4):353–372, mar 2008.

[4] S. Aw and M. Levin. Is left-right asymmetry a form of planar cell polarity? *Development*, 136(3):355–366, feb 2009.

[5] A. Balzeau, E. Gilissen, and D. Grimaud-Hervé. Shared Pattern of Endocranial Shape Asymmetries among Great Apes, Anatomically Modern Humans, and Fossil Hominins. *PLoS ONE*, 7(1):e29581, jan 2012.

[6] M. L. Cara, I. Streata, A. M. Buga, and D. G. Iliescu. Developmental Brain Asymmetry. The Good and the Bad Sides. *Symmetry 2022, Vol. 14, Page 128*, 14(1):128, jan 2022.

[7] A. Cichonska, J. Rousu, P. Marttinen, A. J. Kangas, P. Soininen, T. Lehtimäki, O. T. Raitakari, M. R. Järvelin, V. Salomaa, M. Ala-Korpela, S. Ripatti, and M. Pirinen. metaCCA: summary statistics-based multivariate meta-analysis of genome-wide association studies using canonical correlation analysis. *Bioinformatics*, 32(13):1981–1989, jul 2016.

[8] P. Claes, J. Roosenboom, J. D. White, T. Swigut, D. Sero, J. Li, M. K. Lee, A. Zaidi, B. C. Mattern, C. Liebowitz, L. Pearson, T. González, E. J. Leslie, J. C. Carlson, E. Orlova, P. Suetens, D. Vandermeulen, E. Feingold, M. L. Marazita, J. R. Shaffer, J. Wysocka, M. D. Shriver, and S. M. Weinberg. Genome-wide mapping of global-to-local genetic effects on human facial shape. *Nature Genetics*, 50(3):414–423, 2018.

[9] M. L. Concha, I. H. Bianco, and S. W. Wilson. Encoding asymmetry within neural circuits. *Nature Reviews Neuroscience 2012 13:12*, 13(12):832–843, nov 2012.

[10] M. C. Corballis. The evolution and genetics of cerebral asymmetry. *Philosophical Transactions of the Royal Society B: Biological Sciences*, 364(1519):867, 2009.

[11] R. S. Desikan, F. Ségonne, B. Fischl, B. T. Quinn, B. C. Dickerson, D. Blacker, R. L. Buckner, A. M. Dale, R. P. Maguire, B. T. Hyman, M. S. Albert, and R. J. Killiany. An automated labeling system for subdividing the human cerebral cortex on MRI scans into gyral based regions of interest. *NeuroImage*, 31(3):968–980, jul 2006.

[12] L. I. Dyck and E. M. Morrow. Genetic control of postnatal human brain growth. *Current opinion in neurology*, 30(1):114, 2017.

[13] A. Ernst and J. Frisén. Adult Neurogenesis in Humans- Common and Unique Traits in Mammals. *PLoS Biology*, 13(1), 2015.

[14] M. F. Bear, B. W. Connors, and M. A. Paradiso. Differentiation of the Forebrain. In *Exploring the brain*, chapter 7, pages 196–199. Wolters Kluwer, 4th edition, 2016.

[15] M. F. Bear, B. W. Connors, and M. A. Paradiso. Formation of the Neural Tube-Three Primary Brain Vesicles. In *Exploring the brain*, chapter 7, pages 193–196. Wolters Kluwer, 4th edition, 2016.

[16] M. F. Bear, B. W. Connors, and M. A. Paradiso. Special Features of the Human CNS. In *Exploring the brain*, chapter 7, pages 205–207. Wolters Kluwer, 4th edition, 2016.

[17] S. B. Fernandes, K. S. Zhang, T. M. Jamann, and A. E. Lipka. How Well Can Multivariate and Univariate GWAS Distinguish Between True and Spurious Pleiotropy? *Frontiers in Genetics*, 11:1747, jan 2021.

[18] J. R. Finnerty. The origins of axial patterning in the metazoa: how old is bilateral symmetry? *International Journal of Developmental Biology*, 47(7-8):523–529, dec 2003.

[19] T. E. Galesloot, K. Van Steen, L. A. Kiemeney, L. L. Janss, and S. H. Vermeulen. A comparison of multivariate genome-wide association methods. *PLoS ONE*, 9(4), apr 2014.

[20] M. Götz and W. B. Huttner. The cell biology of Neurogenesis. *Nat Rev Mol Cell Biol*, 6:777–788, 2005.

[21] D. T. Grimes and R. D. Burdine. Left-right patterning: breaking symmetry to asymmetric morphogenesis. *Trends in genetics : TIG*, 33(9):616, sep 2017.

[22] P. Heger, W. Zheng, A. Rottmann, K. A. Pan Lio, and T. Wiehe. The genetic factors of bilaterian evolution. *eLife*, 9:1–45, jul 2020.

[23] A. Hejnol and K. Pang. Xenacoelomorpha's significance for understanding bilaterian evolution. *Current Opinion in Genetics And Development*, 39:48–54, aug 2016.

[24] M. R. Herbert, D. A. Ziegler, C. K. Deutsch, L. M. O'Brien, D. N. Kennedy, P. A. Filipek, A. I. Bakardjiev, J. Hodgson, M. Takeoka, N. Makris, and V. S. Caviness Jr. Brain asymmetries in autism and developmental language disorder: a nested whole-brain analysis. *Brain*, 128(1):213–226, jan 2005.

[25] D. Jackson, R. Riley, and I. R. White. Multivariate meta-analysis: Potential and promise. *Statistics in Medicine*, 30(20):2481, sep 2011.

[26] C. P. Klingenberg. Walking on Kendall's Shape Space: Understanding Shape Spaces and Their Coordinate Systems. *Evolutionary Biology*, 47(4):334–352, dec 2020.

[27] C. P. Klingenberg, M. Barluenga, and A. Meyer. Shape analysis of symmetric structures: Quantifying variation among individuals and asymmetry. *Evolution*, 56(10):1909–1920, 2002.

[28] E. M. Koch and S. R. Sunyaev. Maintenance of Complex Trait Variation: Classic Theory and Modern Data. *Frontiers in Genetics*, 12:2198, nov 2021.

[29] X.-Z. Kong, M. C. Postema, T. Guadalupe, C. de Kovel, P. S. W. Boedhoe, M. Hoogman, S. R. Mathias, D. van Rooij, D. Schijven, D. C. Glahn, S. E. Medland, N. Jahanshad, S. I. Thomopoulos, J. A. Turner, J. Buitelaar, T. G. M. van Erp, B. Franke, S. E. Fisher, O. A. van den Heuvel, L. Schmaal, P. M. Thompson, and C. Francks. Mapping brain asymmetry in health and disease through the ENIGMA consortium. *Human Brain Mapping*, 43(1):167–181, jan 2022.

[30] M. LeMay. Morphological cerebral asymmetries of modern man, fossil man, and nonhuman primate. *Annals of the New York Academy of Sciences*, 280:349–366, 1976.

[31] T. J. Littlejohns, J. Holliday, L. M. Gibson, S. Garratt, N. Oesingmann, F. Alfaro-Almagro, J. D. Bell, C. Boultwood, R. Collins, M. C. Conroy, N. Crabtree, N. Doherty, A. F. Frangi, N. C. Harvey, P. Leeson, K. L. Miller, S. Neubauer, S. E. Petersen, J. Sellors, S. Sheard, S. M. Smith, C. L. Sudlow, P. M. Matthews, and N. E. Allen. The UK Biobank imaging enhancement of 100,000 participants: rationale, data collection, management and future directions. *Nature Communications 2020 11:1*, 11(1):1–12, may 2020.

[32] S. Malik, G. Vinukonda, L. R. Vose, D. Diamond, B. B. Bhimavarapu, F. Hu, M. T. Zia, R. Hevner, N. Zecevic, and P. Ballabh. Neurogenesis continues in

the third trimester of pregnancy and is suppressed by premature birth. *The Journal of neuroscience : the official journal of the Society for Neuroscience*, 33(2):411–423, jan 2013.

[33] Z. Molnár, G. J. Clowry, N. Šestan, A. Alzu'bi, T. Bakken, R. F. Hevner, P. S. Hüppi, I. Kostović, P. Rakic, E. S. Anton, D. Edwards, P. Garcez, A. Hoerder-Suabedissen, and A. Kriegstein. New insights into the development of the human cerebral cortex. *Journal of Anatomy*, 235(3):432, 2019.

[34] S. Naqvi, Y. Sleyp, H. Hoskens, K. Indencleef, J. P. Spence, R. Bruffaerts, A. Radwan, R. J. Eller, S. Richmond, M. D. Shriver, J. R. Shaffer, S. M. Weinberg, S. Walsh, J. Thompson, J. K. Pritchard, S. Sunaert, H. Peeters, J. Wysocka, and P. Claes. Shared heritability of human face and brain shape. *Nature Genetics*, 53(6):830–839, 2021.

[35] S. Neubauer, P. Gunz, N. A. Scott, J. J. Hublin, and P. Mitteroecker. Evolution of brain lateralization: A shared hominid pattern of endocranial asymmetry is much more variable in humans than in great apes. *Science Advances*, 6(7):9935–9949, 2020.

[36] A. Y. Ng, M. I. Jordan, and Y. Weiss. On spectral clustering: Analysis and an algorithm. In *Advances in Neural Information Processing Systems*, 2002.

[37] T. J. Nowakowski, A. A. Pollen, C. Sandoval-Espinosa, and A. R. Kriegstein. Transformation of the Radial Glia Scaffold Demarcates Two Stages of Human Cerebral Cortex Development. *Neuron*, 91(6):1219–1227, sep 2016.

[38] Y. Okada, S. Takeda, Y. Tanaka, J. C. I. Belmonte, and N. Hirokawa. Mechanism of Nodal Flow: A Conserved Symmetry Breaking Event in Left-Right Axis Determination. *Cell*, 121(4):633–644, may 2005.

[39] M. A. Purnell, P. J. Donoghue, S. E. Gabbott, M. E. McNamara, D. J. Murdock, and R. S. Sansom. Experimental analysis of soft-tissue fossilization: opening the black box. *Palaeontology*, 61(3):317–323, may 2018.

[40] P. Rakic. Radial glial cells: Brain functions. In *Encyclopedia of Neuroscience*, volume R, pages 15–21. Elsevier Ltd, 2009.

[41] M. Ribolsi, Z. J. Daskalakis, A. Siracusano, and G. Koch. Abnormal Asymmetry of Brain Connectivity in Schizophrenia , 2014.

[42] J. Schmitz, O. Güntürkün, and S. Ocklenburg. Building an asymmetrical brain: The molecular perspective. *Frontiers in Psychology*, 10(APR):982, 2019.

[43] J. Sheng, L. Wang, H. Cheng, Q. Zhang, R. Zhou, and Y. Shi. Strategies for multivariate analyses of imaging genetics study in Alzheimer's disease. *Neuroscience Letters*, 762:136147, sep 2021.

[44] L. N. Telano and S. Baker. Physiology, Cerebral Spinal Fluid. *StatPearls*, jul 2021.

[45] S. Van Dongen, G. Molenberghs, and E. Matthysen. The statistical analysis of fluctuating asymmetry: REML estimation of a mixed regression model. *Journal of Evolutionary Biology*, 12(1):94–102, jan 1999.

[46] N. Vijayakumar, N. B. Allen, G. Youssef, M. Dennison, M. Yücel, J. G. Simmons, and S. Whittle. Brain development during adolescence: A mixed-longitudinal investigation of cortical thickness, surface area, and volume. *Human Brain Mapping*, 37(6):2027–2038, jun 2016.

[47] G. Vingerhoets, R. Gerrits, and H. Verhelst. Atypical Brain Asymmetry in Human Situs Inversus: Gut Feeling or Real Evidence? *Symmetry 2021, Vol. 13, Page 695*, 13(4):695, apr 2021.

[48] U. Von Luxburg. A tutorial on spectral clustering. *Statistics and Computing 2007 17:4*, 17(4):395–416, aug 2007.