Searching for new genes in bipolar disorder sub-types through gene-based analysis approach

Aleix Ramon Brugada Iglesias

Scientific director: María Cristina Rodriguez Fontenla[1]

[1]Grupo de Medicina Xenómica, CIBERER, CIMUS (Centre for Research in Molecular Medicine and Chronic Diseases), Universidade de Santiago de Compostela, Santiago de Compostela, Spain

Abstract

Bipolar disorder, formerly called manic depression, is a mental health condition characterized by extreme mood swings that can range from extreme highs (mania) to extreme lows (depression). Episodes of mania and depression often last for several weeks or months. As for the genetic basis, little is known for certain. While occasional families may present a single gene which determines the susceptibility, the majority of cases involve the interaction of numerous genes. Various studies have been performed to identify these genetic variations in an effort to unravel the genetic basis of bipolar disorder.

The purpose of this study is to carry out a genetic and functional in silico characterization of bipolar disorder (BD) and 2 of its sub-types (BD1 and BD2). Thus, the pathogenesis of BD and its sub-types will be explored at a genetic level, in which we will use summary statistics from the GWAS meta-analysis done by the PGC (Psychiatric Genomics Consortium) and a gene-based analysis method called PASCAL to search for the association of new genes, at a biological pathway level, in which we will correlate the previously associated genes with GO processes, and at the cellular level in which we will use cell-type enrichment analyses to identify those BD genes expressed in each neuronal cell type.

1 Introduction

Bipolar Disorder (BD) is a mental disorder that causes unusual shifts in mood, energy, concentration and social behaviour. Many other mental health issues are associated with BD such as anxiety disorders and substance use disorders. It occurs in about 1% of the general population and presents itself most commonly at young childhood, around 20 years of age [1]. Since it impacts daily life substantially, a quarter to a third of those affected have social and financial problems due to BD. This has lead to it being one of the most substantial costs for society [2]. Bipolar Disorder also increases the risk of suicide by 10-30 times. Up to 20% of individuals with untreated BD lose their life to suicide while 20-60% attempt it at least once [3].

BD is characterized as a spectrum of disorders. The DSM-5 lists 3 sub-types [4]: bipolar type I disorder (BD1), bipolar type II disorder (BD2) and cyclothymatic disorder. BD1 is characterized by the occurrence of one or more severe maniac episodes while BD2 is characterized by no severe maniac episodes but one or more hypomaniac episodes and one or more major depressive episodes. Cyclothymatic disorder however has no presence of severe maniac episodes nor major depressive episodes.

The causes of BD are not deeply understood as both genetic and environmental factors are a known cause. BD does not follow a Mendelian inheritance pattern but it is still found within multiple individuals in the same family. However, most patients are isolated cases, further confirming that the BD is most likely influenced by multiple genetic risk factors with small effect. While some genes like *CACNA1C*, *ODZ4* and *NCAN* have appeared promising candidate genes for BD in genome-wide association studies[5], none can be considered a risk factor as many individuals that carry these variations are considered healthy [6].

The most recent study performed by the Bipolar Disorder Workgroup from the Psychiatric Genomics Consortium explored common genetic variations (SNPs) in 20,352 cases and 31,358 controls [7]. Using a gene-based analysis tool called MAGMA [8], they found 30 new loci associated with BD.

In this study we are going to use the same GWAS dataset which is provided by the PGC, as well as 2 more datasets for BD1 and BD2 in order to observe the genetic and functional differences between sub-types. However, we are going to implement a different gene-based analysis tool called PASCAL in order to search for novel genes. Furthermore, we are going to explore which biological pathways are affected by these genes and how they interact with each other since BD is most likely the result of epistasis or multiple gene interactions. Finally we will perform an expression weighted cell-type enrichment analysis in order to identify the most enriched neuronal cell-type.

2 Methods

2.1 GWAS Datasets

The datasets used in this analysis consist of GWAS summary statistics for Bipolar Disorder published by the Psychiatric Genomics Consortium[7][9]. The main dataset is comprised of 20,352 cases and 31,358 controls of European descent. The BD1 dataset contains 14,879 cases and 30,992 controls and the BD2 dataset contains 3,421 cases and 22,155 controls.

2.2 Gene-based analysis

To perform the gene based analysis we used PASCAL (Pathway Scoring Algorithm). As input, we used the summary statistics obtained from the BD, BD1 and BD2 meta-analysis. First we mapped the individual SNPs from the GWAS results to genes using the default $\pm 50kb$ window around the start and end of each gene. The maximum number of SNPs per gene was left at the default 3000. Furthermore, we considered LD information from 1000 Genomes to account for linkage between markers for each gene. The significance threshold for BD was set to 2.26×10^{-6} as per the Bonferroni correction (0.05 / 22094 genes). The threshold for BD1 was set to 2.28×10^{-8} (0,05 / 21888 genes) and the threshold for BD2 was set to 2.29×10^{-8} (0,05 / 21875 genes).

2.3 Enrichment analysis at a biological pathway level

To perform the functional characterization for gene ontologies we used *Enrichr*. The gene lists of the relevant genes obtained previously were used as input to find the related GO terms. First we selected the databases we want to search through from the Enrichr database list. In this case we selected the most recent available versions of the 3 GO databases (Molecular Function 2018, Cellular Component 2018 and Biological Process 2018). We then queried the gene lists to obtain their related GO terms.

To visualize the GO terms we used REViGO [10]. REViGO takes as input a long list of Gene Ontology terms and associated p-values, and summarizes them by removing redundant GO terms. We visualised the 40 most significant GO terms for both BD and BD1. We set an allowed similarity of 0.5 and SimRel semantic similarity measure. The plot generation was done by modifying the R script provided by the REViGO web server.

The gene lists were further used to construct gene networks using GeneMANIA [11]. The association between genes includes protein and genetic interactions, pathways, co-expression, co-localization and protein domain similarity. Furthermore, GeneMANIA finds other genes related to the set of input genes, thus giving further insight into their function.

2.4 Enrichment analysis at a cellular level

The R package EWCE (Expression Weighted Celltype Enrichment) [12] was used to analyse the differential genetic expression in different neural celltypes. The gene lists of relevant genes for BD and BD1 were again used as input. EWCE first tests whether these genes are enriched in any particular celltype. This is done by first setting a background set which in this case was all human genes with mouse orthologs. The reason behind this is that the single cell transcriptome data used in this analysis was from the brain of mice provided by the Karolinska Institute.

The probability distribution for the gene list was calculated by creating 100000 lists of random genes from the background set, adjusting for length and GC content, and determining the expression

level for each. This bootstrapping method was used for both annotation levels, with level 1 being cell type and level 2 being sub-cell type.

The scripts used can be found at: `https://github.com/aleixyuki/FGP/tree/main/Rscripts`

3 Results

3.1 Gene-based analysis

We performed a gene-based analysis on the GWAS meta-analysis for Bipolar Disorder (BD), Bipolar Disorder type 1 (BD1) and Bipolar Disorder type 2 (BD2) from the PGC using PASCAL. PASCAL computes gene scores by aggregating SNP p-values from our GWAS meta-analysis, without the need for individual phenotypes, while correcting for linkage disequilibrium (LD) structure. It computes gene p-values based on the most significant SNP and the average association signal across the region.

We obtained a total of 22094 genes for BD to which we applied a Bonferroni correction (0.05 / 22094) in order to obtain the threshold for the significant genes (Figure 1). In this case the threshold was a p-value of 2.26×10^{-6}. For BD1 we obtained 21888 genes which resulted in a threshold of 2.28×10^{-6} and for BD2 we obtained 21875 genes and a threshold of 2.29×10^{-6}.

This filtering resulted in 86 significant genes for BD, of which *ITIH4* (p-value = 1.05×10^{-8}) and *KCNS1* (p-value = 4.36×10^{-9}) were the most significant, 52 for BD1, of which *WFDC5* (p-value = 2.31×10^{-8}) and *KCNS1* (p-value = 2.98×10^{-9}) were the most significant and none for BD2.

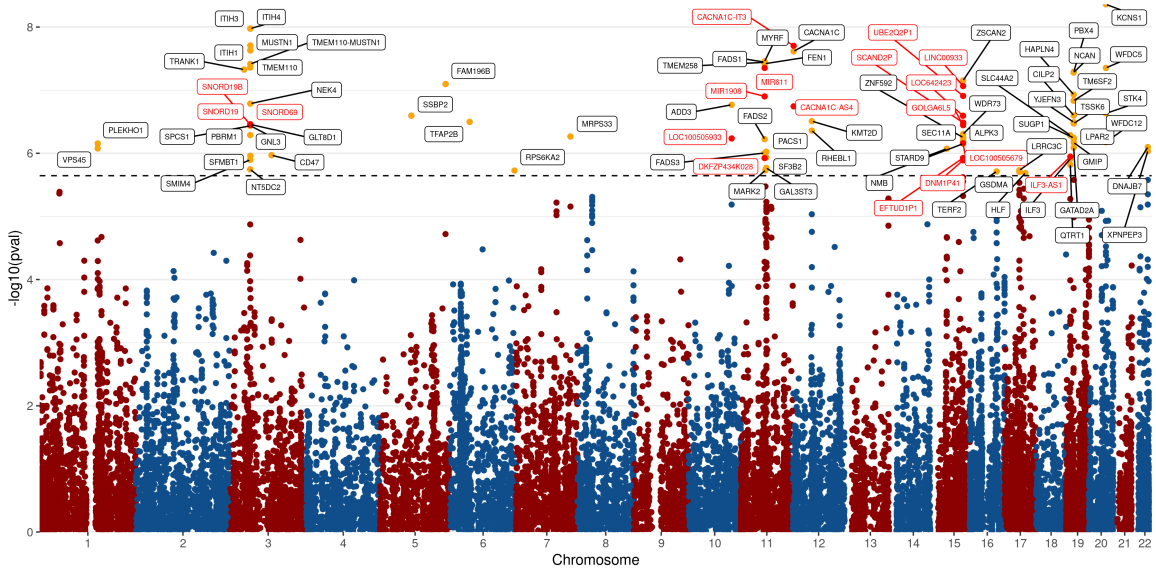

Figure 1: **PASCAL results Manhattan plot.** GWAS Manhattan plot with the significant genes in orange and the novel genes in red. The dotted grey line is the threshold set by the Bonferroni correction.

3.2 Enrichment analysis of gene ontologies

To perform the functional characterization at a biological pathway level, we used an R package called Enrichr to associate the significant genes to their corresponding GO terms.

For BD, we obtained 125 GO terms within the Molecular Function database of which hyaluronic acid binding (GO:0005540) is the most significant with a p-value of 2.67×10^{-3}. As for the Cellular Component database, we obtained 60 results with platelet dense granule lumen (GO:0031089) being the most significant with a p-value of 1.6×10^{-3}. Lastly for the Biological Process database we obtained

408 results with the most significant being forelimb morphogenesis (GO:0035136) with a p-value of 8.04×10^{-4} (Figure 2).

For BD1, we obtained 79 GO terms related to Molecular Function, of which hyaluronic acid binding (GO:0005540) is also the most significant with a p-value of 9.87×10^{-4}. In the Cellular Component database we obtained 53 related GO terms with endoplasmic reticulum-Golgi intermediate compartment (GO:0005793) being the most significant with a p-value of 1.08×10^{-4}. Finally, we obtained 336 GO terms related to the Biological Process database. The most significant GO term was pyroptosis (GO:0070269) with a p-value of 2.94×10^{-4}.

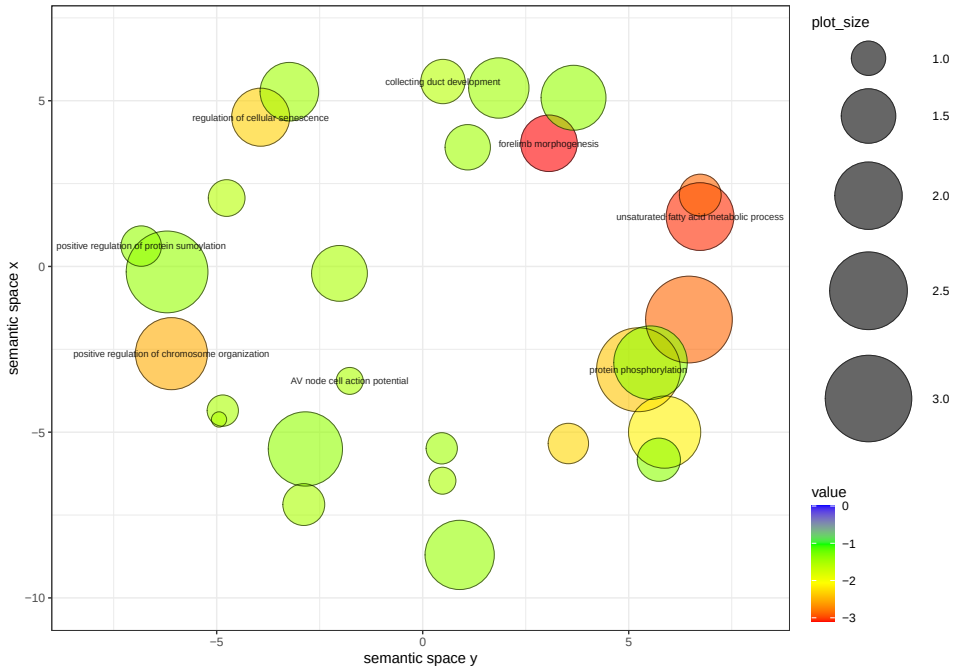

Figure 2: **Biological Processes bubble plot.** Bubble plot showing the top 30 enriched Biological Processes for our BD gene list.

3.3 Network analysis

GeneMANIA identified interactors for the loci associated with BD and BD1 in the GBA done with PASCAL.

The gene network for BD was constructed with a total of 89 genes (69 associated genes and 30 interactors) and 371 links which include shared protein domain, co-expression, co-localization , physical interactions, genetic interactions, pathway and prediction. 4 of the 69 associated genes present no type of interaction between themselves nor any other gene (*DNAJB7*, *LRRC3C*, *STARD9*, *ZSCAN2*).

The gene network for BD1 contains 61 total genes (41 associated genes and 20 interactors) and 163 total links which only includes shared protein domain, co-expression, co-localization and prediction. 6 of the 42 associated present no type of interaction (*KMT2D*, *LRRC3C*, *PBX4*, *ZSCAN2*, *CDAN1*, *STARD9*)

3.4 Cell-type enrichment analysis

We examined if the genes were differentially expressed in the transcriptional datasets corresponding to both cell-type annotation levels. For the cell-type annotation level 1 (Figure 3) we observed significant enrichment in the astrocytes and ependymal cell-type (p-value = 0.0069) for BD as shown by the asterisk above the corresponding column. For BD1 there was no significant enrichment but the astrocytes and ependymal (p-value = 0.0903), pyramidal CA1 (p-value = 0.0397) and pyramidal SS (p-value = 0.0925) cell-types were differentially expressed.

We then proceeded to see if any of the microglial subtypes (annotation level 2) showed significant enrichment (Figure 4). We observed no significant enrichment of any subtype although *Int15* (Interneuron 15) was differentially expressed both in BD and BD1 with a p-value of 5.9×10^{-3} and 1.4×10^{-3} respectively. Furthermore, in BD, *Int6* (Interneuron 6) also showed differential expression above the rest of the microglial subtypes (p-value = 0.0158) but not enough to be considered significant.

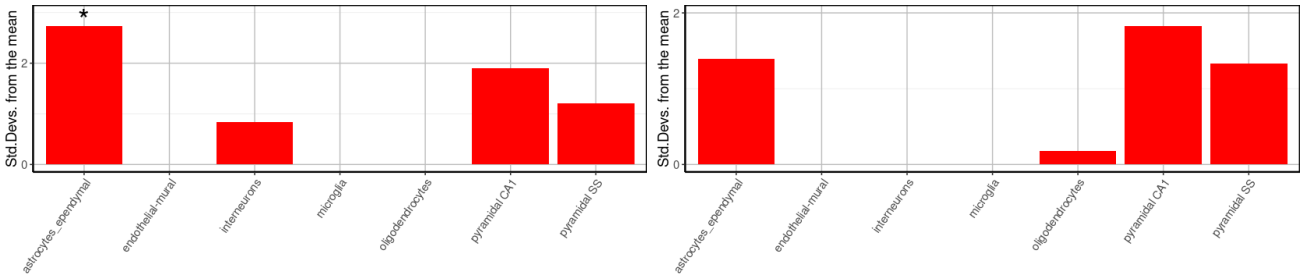

Figure 3: **EWCE plot for annotation level 1.** The left graph corresponds to BD where we observe the enrichment of the astrocytes_ependymal cell type and the right graph corresponds to BD1.

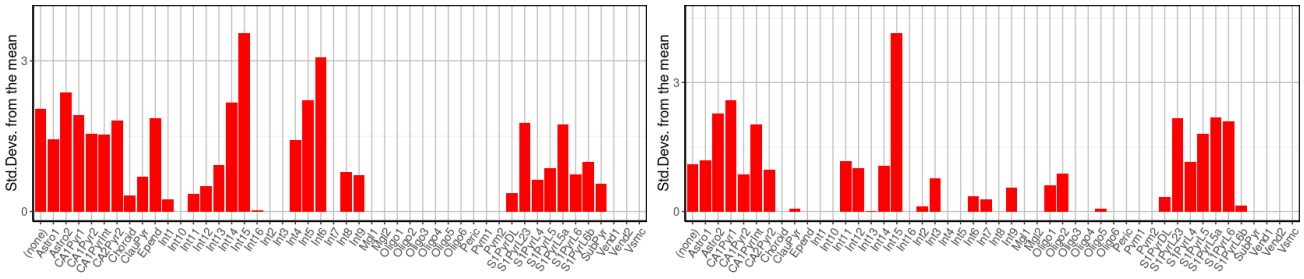

Figure 4: **EWCE plot for annotation level 2.** The left graph corresponds to BD and the right graph corresponds to BD1.

4 Discussion

We identified 18 genes not previously mentioned in the 2019 paper by the PGC [7]. We should mention that most of the novel genes are related to previously known significant genes and may be relevant because of co-localization or co-expression with respect to these genes.

Of the novel BD associated genes found, *SNORD19*, *SNORD19B* and *SNORD69* are small nucleolar RNAs (snoRNAs) that belong to the C/D box family. They are also non-coding RNAs (ncRNAs) whose function is directing site-specific 2'-O-methylation of substrate RNAs [13]. Copy number variations of snoRNAs have also been associated with neuropsychiatric disorders [14]. For example, the gain of additional copies of *SNORD115* has been linked to autism [15]. *DKFZP434K028* or *MYRF-AS1* is an antisense transcript (asRNA). Similar to other asRNAs like *CACNA1C-AS4* and *ILF3-AS1*, its function is to regulate its target gene's expression [16] which is the *MYRF* gene. Their significance with respect to BD and its sub-types stems from the significance of their target genes. For example, *MYRF* (myelin regulatory factor) encodes a transcription factor that is required for central nervous system myelination and may regulate oligodendrocyte differentiation. It has been observed that myelin and oligodendrocyte dysfunction contributed to the risk of schizophrenia and bipolar disorder [17]. As can be seen in the Manhattan plot (Figure 1), these genes are co-localized with their target gene. *MIR611* and *MIR1908* can be considered relevant since microRNAs have been found to be associated with schizophrenia although no direct link has been established for BD [18]. Their relation to mental

disorders is still being studied. Furthermore, animal studies have shown that manipulating the levels of particular microRNAs in the brain can alter behaviour [19]. The last relevant genes are pseudogenes, like *SCAND2P* and *UBE2Q2P1* which may be relevant not only for mental disorders like BD but for any genetic disorder [20].

Although these genes may be associated with BD, further functional studies are required to determine their functional role.

For BD1, we obtained 11 novel genes compared to the recent PGC study [7] but all were present in the novel genes found for BD. As for BD2, no novel genes were found. Furthermore, no significant genes were found, most likely due to the heterogeneity of the data compared to the 20,352 cases and 31,358 controls of European descent present in the BD data.

As for the enrichment analysis of gene ontologies (GO), the most significant Molecular Function GO term for BD and BD1 was hyaluronic acid binding (GO:0005540). Hyaluronic acid plays the main structural role in the formation of brain extracellular matrix (ECM)[21]. Since the ECM has an important role in brain development, maturation of neural circuits and adult neuroplasticity, processes that affect the composition of ECM could lead to altered brain function [22]. There have been studies that link altered ECM function and neuropsychiatric disorders like addiction and schizophrenia [23].

The Biological Processes were different for BD and BD1. The most significant for BD was forelimb morphogenesis (GO:0035136) most likely due to it being a process in which *CACNA1C* is involved. Many association studies have shown that genetic variations in *CACNA1C* increase risk for psychiatric disorders such as BD in this case [24]. The most significant for BD1 was pyroptosis(GO:0070269) which is an inflammatory cell death which is triggered by pathological stimuli [25]. If pyroptosis occurs in the central nervous system it can cause neuroinflammation, whose evidence of pathogenesis in BD has been reviewed but no conclusive evidence has been found [26].

All the novel genes found are not functionally characterized and thus GeneMANIA didn't recognize them so their interactions with the other significant genes are unknown at the moment.

Finally, regarding the cell-type enrichment analysis, despite the enrichment of *Int15* (Interneuron 15) in both BD and BD1, the level 1 annotation shows no enrichment for the interneuron cell-type due to not all interneuron subtypes being enriched. However the enrichment of interneurons was unexpected since it is normally associated with intellectual disabilities and epilepsy rather than mood disorders [27]. For BD1, the fact that the most enriched cell-type are pyramidal neurons confirms previous findings that BD1 is more genetically correlated with schizophrenia since they share significant genes [28]. BD also presents this slight enrichment of pyramidal neurons due again to the heterogeneity of the data which presents no distinction between BD subtypes. The most relevant finding was the significant enrichment of the astrocytes and ependymal cell-type. While this cell-type has also been attributed to intellectual disabilities and epilepsy [12], more recent studies indicate that astrocyte dysfunction is involved in BD, schizophrenia and Alzheimer's disease [29].

5 Conclusion

This study has shown that PASCAL could be a viable method in gene-based analysis in order to uncover new associated genes and provide more insight into other conditions similar to bipolar disorder such as schizophrenia and other mood disorders. PASCAL has also defined associations between BD and ncRNAs and microRNAs, in which future genetic and functional studies could determine their role in the biological etiology of BD.

Supplementary Material: `https://github.com/aleixyuki/FGP`

References

[1] Iria Grande, Michael Berk, Boris Birmaher, and Eduard Vieta. "Bipolar disorder". In: *The Lancet* 387.10027 (2016), pp. 1561–1572. ISSN: 0140-6736. DOI: `https://doi.org/10.1016/S0140-6736(15)00241-X`. URL: `https://www.sciencedirect.com/science/article/pii/S014067361500241X`.

[2] Alize J Ferrari, Emily Stockings, Jon-Paul Khoo, Holly E Erskine, Louisa Degenhardt, Theo Vos, and Harvey A Whiteford. "The prevalence and burden of bipolar disorder: findings from the Global Burden of Disease Study 2013". In: *Bipolar Disorders* 18.5 (2016), pp. 440–450. DOI: https://doi.org/10.1111/bdi.12423. eprint: https://onlinelibrary.wiley.com/doi/pdf/10.1111/bdi.12423. URL: https://onlinelibrary.wiley.com/doi/abs/10.1111/bdi.12423.

[3] P. Dome, Z. Rihmer, and X. Gonda. "Suicide Risk in Bipolar Disorder: A Brief Review". In: *Medicina* 55 (July 2019), p. 403. DOI: 10.3390/medicina55080403.

[4] American Psychiatric Association. *Diagnostic And Statistical Manual Of Mental Disorders*. 2013. DOI: 10.5555/appi.books.9780890425596. URL: https://dsm.psychiatryonline.org/doi/abs/10.5555/appi.books.9780890425596.

[5] Janice M Fullerton, Phil H Lee, and Fan Meng. "Large-scale genome-wide association analysis of bipolar disorder identifies a new susceptibility locus near ODZ4". In: *Nature Genetics* 43.10 (Sept. 2011), pp. 977–983. DOI: 10.1038/ng.943.

[6] Kerner B. "Genetics of bipolar disorder". In: *Appl Clin Genet* 7 (2014), pp. 33–42. DOI: https://doi.org/10.2147/TACG.S39297.

[7] Eli A Stahl et al. "Genome-wide association study identifies 30 Loci Associated with Bipolar Disorder". In: *Nature Genetics* (2019). DOI: https://doi.org/10.1038/s41588-019-0397-8.

[8] Christiaan A. de Leeuw, Joris M. Mooij, Tom Heskes, and Danielle Posthuma. "MAGMA: Generalized Gene-Set Analysis of GWAS Data". In: *PLOS Computational Biology* 11.4 (Apr. 2015), pp. 1–19. DOI: 10.1371/journal.pcbi.1004219. URL: https://doi.org/10.1371/journal.pcbi.1004219.

[9] Jonathan R.I. Coleman et al. "The Genetics of the Mood Disorder Spectrum: Genome-wide Association Analyses of More Than 185,000 Cases and 439,000 Controls". In: *Biological Psychiatry* 88.2 (July 2020), pp. 169–184. ISSN: 0006-3223. DOI: 10.1016/j.biopsych.2019.10.015. URL: https://doi.org/10.1016/j.biopsych.2019.10.015.

[10] Fran Supek, Matko Bošnjak, Nives Škunca, and Tomislav Šmuc. "REVIGO Summarizes and Visualizes Long Lists of Gene Ontology Terms". In: *PLOS ONE* 6.7 (July 2011), pp. 1–9. DOI: 10.1371/journal.pone.0021800. URL: https://doi.org/10.1371/journal.pone.0021800.

[11] David Warde-Farley et al. "The GeneMANIA prediction server: biological network integration for gene prioritization and predicting gene function". In: *Nucleic Acids Research* 38.suppl_2 (June 2010), W214–W220. ISSN: 0305-1048. DOI: 10.1093/nar/gkq537. URL: https://doi.org/10.1093/nar/gkq537.

[12] Nathan G. Skene and Seth G. N. Grant. "Identification of Vulnerable Cell Types in Major Brain Disorders Using Single Cell Transcriptomes and Expression Weighted Cell Type Enrichment". In: *Frontiers in Neuroscience* 10 (2016), p. 16. ISSN: 1662-453X. DOI: 10.3389/fnins.2016.00016. URL: https://www.frontiersin.org/article/10.3389/fnins.2016.00016.

[13] Laurent Lestrade and Michel J. Weber. "snoRNA-LBME-db, a comprehensive database of human H/ACA and C/D box snoRNAs". In: *Nucleic Acids Research* 34.suppl_1 (Jan. 2006), pp. D158–D162. ISSN: 0305-1048. DOI: 10.1093/nar/gkj002. eprint: https://academic.oup.com/nar/article-pdf/34/suppl_1/D158/3923176/gkj002.pdf. URL: https://doi.org/10.1093/nar/gkj002.

[14] Edwin H. Cook Jr and Stephen W. Scherer. "Copy-number variations associated with neuropsychiatric conditions". In: *Nature* 455 (Jan. 2008), pp. 919–923. ISSN: 1476-4687. DOI: 10.1038/nature07458. URL: https://doi.org/10.1038/nature07458.

[15] J. Nakatani et al. "Abnormal behavior in a chromosome-engineered mouse model for human 15q11-13 duplication seen in autism". In: *Cell* 137.7 (Apr. 2009), pp. 1235–1246. ISSN: 0092-8674. DOI: 10.1016/j.cell.2009.04.024. URL: https://doi.org/10.1016/j.cell.2009.04.024.

[16] Vicent Pelechano and Lars M. Steinmetz. "Gene regulation by antisense transcription". In: *Nature Reviews Genetics* 14.12 (Dec. 2013), pp. 880–893. DOI: 10.1038/nrg3594. URL: https://doi.org/10.1038/nrg3594.

[17] Hao Yu, Wenjian Bi, Chenxing Liu, Yanlong Zhao, Dai Zhang, and Weihua Yue. "A hypothesis-driven pathway analysis reveals myelin-related pathways that contribute to the risk of schizophrenia and bipolar disorder". In: *Progress in Neuro-Psychopharmacology and Biological Psychiatry* 51 (2014), pp. 140–145. ISSN: 0278-5846. DOI: https://doi.org/10.1016/j.pnpbp.2014.01.006. URL: https://www.sciencedirect.com/science/article/pii/S0278584614000074.

[18] Leif G. Hommers, Katharina Domschke, and Jürgen Deckert. "Heterogeneity and Individuality: microRNAs in Mental Disorders". In: *Journal of Neural Transmission* 122.1 (Jan. 2015), pp. 79–97. ISSN: 1435-1463. DOI: 10.1007/s00702-014-1338-4. URL: https://doi.org/10.1007/s00702-014-1338-4.

[19] Orna Issler and Alon Chen. "Determining the role of microRNAs in psychiatric disorders". In: *Nature Reviews Neuroscience* 16.4 (Apr. 2015), pp. 201–212. ISSN: 1471-0048. DOI: 10.1038/nrn3879. URL: https://doi.org/10.1038/nrn3879.

[20] Kamalika Sen. "Relevance of Pseudogenes to Human Genetic Disease". In: *eLS*. American Cancer Society, 2013. ISBN: 9780470015902. DOI: https://doi.org/10.1002/9780470015902.a0025002. eprint: https://onlinelibrary.wiley.com/doi/pdf/10.1002/9780470015902.a0025002. URL: https://onlinelibrary.wiley.com/doi/abs/10.1002/9780470015902.a0025002.

[21] Amico Bignami, Mark Hosley, and Doris Dahl. "Hyaluronic acid and hyaluronic acid-binding proteins in brain extracellular matrix". In: *Anatomy and Embryology* 188.5 (Nov. 1993), pp. 419–433. ISSN: 1432-0568. DOI: 10.1007/BF00190136. URL: https://doi.org/10.1007/BF00190136.

[22] B. R. Lubbers, A. B. Smit, S. Spijker, and M. C. van den Oever. "Neural ECM in addiction, schizophrenia, and mood disorder". In: *Progress in brain research* 214 (2014), 263–284. DOI: https://doi.org/10.1016/B978-0-444-63486-3.00012-8.

[23] Sabina Berretta. "Extracellular matrix abnormalities in schizophrenia". In: *Neuropharmacology* 62.3 (2012). Schizophrenia, pp. 1584–1597. ISSN: 0028-3908. DOI: https://doi.org/10.1016/j.neuropharm.2011.08.010. URL: https://www.sciencedirect.com/science/article/pii/S0028390811003431.

[24] Anna L Moon, Niels Haan, Lawrence S Wilkinson, Kerrie L Thomas, and Jeremy Hall. "CACNA1C: Association With Psychiatric Disorders, Behavior, and Neurogenesis". In: *Schizophrenia Bulletin* 44.5 (July 2018), pp. 958–965. ISSN: 0586-7614. DOI: 10.1093/schbul/sby096. eprint: https://academic.oup.com/schizophreniabulletin/article-pdf/44/5/958/25516143/sby096.pdf. URL: https://doi.org/10.1093/schbul/sby096.

[25] Tessa Bergsbaken, Susan L. Fink, and Brad T. Cookson. "Pyroptosis: host cell death and inflammation". In: *Nature Reviews Microbiology* 7.2 (Feb. 2009), pp. 99–109. ISSN: 1740-1534. DOI: 10.1038/nrmicro2070. URL: https://doi.org/10.1038/nrmicro2070.

[26] Kotzalidis GD, Ambrosi E, Simonetti A, Cuomo I, Casale AD, Caloro M, Savoja V, and Rapinesi C. "Neuroinflammation in bipolar disorders". In: *Neuroimmunol Neuroinflammation* 2 (2015), pp. 252–262. DOI: 0.4103/2347-8659.167309. URL: http://dx.doi.org/10.4103/2347-8659.167309.

[27] Ikuo Ogiwara et al. "Nav1.1 Localizes to Axons of Parvalbumin-Positive Inhibitory Interneurons: A Circuit Basis for Epileptic Seizures in Mice Carrying an Scn1a Gene Mutation". In: *Journal of Neuroscience* 27.22 (2007), pp. 5903–5914. ISSN: 0270-6474. DOI: 10.1523/JNEUROSCI.5270-06.2007. eprint: https://www.jneurosci.org/content/27/22/5903.full.pdf. URL: https://www.jneurosci.org/content/27/22/5903.

[28] Niamh Mullins et al. "Genome-wide association study of more than 40,000 bipolar disorder cases provides new insights into the underlying biology". In: *Nature Genetics* 53.6 (June 2021), pp. 817–829. ISSN: 1546-1718. DOI: 10.1038/s41588-021-00857-4. URL: https://doi.org/10.1038/s41588-021-00857-4.

[29] Andrea D. Rivera and Arthur M. Butt. "Astrocytes are direct cellular targets of lithium treatment: novel roles for lysyl oxidase and peroxisome-proliferator activated receptor-γ as astroglial targets of lithium". In: *Translational Psychiatry* 9.1 (Sept. 2019), p. 211. ISSN: 2158-3188. DOI: 10.1038/s41398-019-0542-2. URL: https://doi.org/10.1038/s41398-019-0542-2.