# Identifying Mood Disorder Susceptibility Loci in a Densely Affected Pedigree from the Andalucian Region of Spain

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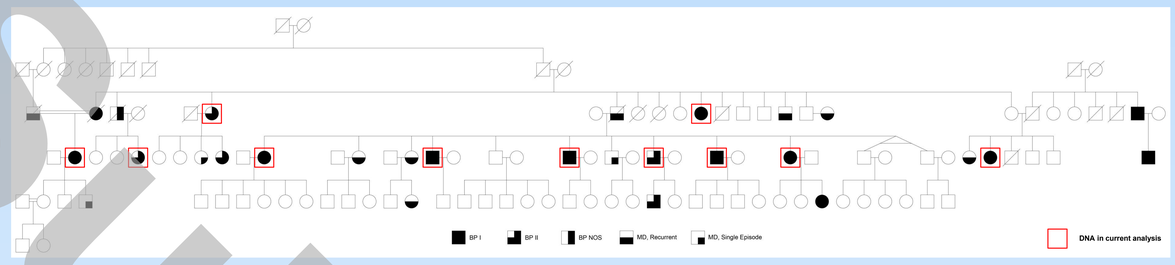



Figure 1: Pedigree of Andalucian family with high incidence of mood disorders

## Introduction

### *Bipolar disorder*

- Lifetime prevalence of ~3.9%.
- Complex genetics and high degrees of heterogeneity.
- Large, densely affected pedigrees may reduce heterogeneity and select for variants of large effect.

### *Pedigree*

- Identified and phenotyped large multigenerational pedigree (Figure 1).
- Densely affected with bipolar disorder (BP).
- Additional members are affected with major depressive disorder (MD).
- Family is from Andalucian region of Spain with limited in-migration.

We are using a multipronged approach to scan this family for variants that appear to be segregating with disease.

## Methods

We applied multiple complementary genomic technologies to this pedigree including SNP genotyping using the Illumina OMNI 1M chip, high density copy number variant mapping using Nimblegen 720K CGH tiling arrays, and exome sequencing using SureSelect v1 All Exome/Illumina PE X75. Whole genome sequencing is currently in progress using Illumina's HiSeq 2000 with version 3 chemistry.

| Technology | Purpose |
|---|---|
| Nimblegen 720K aCGH | Copy number variation |
| Illumina OMNI | Shared segments inherited IBD |
| Exome sequencing | Exonic mutations |
| *Genome sequencing* | *Any detectible mutation* |

## Results

### *CNV Analysis*

Obtained CNV calls from Nimblegen aCGH for 7 affected individuals (BPI or BPII)

- No large, novel CNVs.
- Some previously identified CNVs, but none in most individuals
- Confirmed some with data from Illumina OMNI chips (all 11 individuals).

### *Shared Segment Analysis*

Identify segments of DNA shared by affected individuals within the family. Under a simple, causal model, these regions could contain causal genetic variation.

- Multiple methods yielded partially overlapping results (Figure 2):
- **Germline**(1), **Beagle**(2)**, and PEDIBD**(3)**:** Uses pairwise analysis to predict number of pairs sharing a region IBD.
- **IBS regions**(4)**:** Detects longest regions with IBS in all individuals across all markers.
- Used multiple models due to varying limitations:
  - Dependent on phasing accuracy.
  - No heterogeneity allowed.
  - Not allowing for genotyping errors.
  - Detects centromeres or other regions with few markers as false positives.

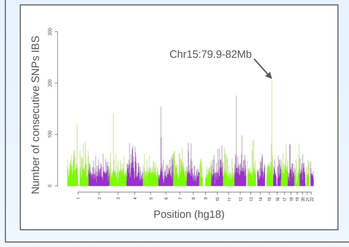
**Top Germline findings**

| Chr | start | end | # of pairs (Max=55) |
|---|---|---|---|
| 11 | 20455988 | 25098764 | 25 |
| 11 | 70111335 | 70872207 | 26 |
| 13 | 79343559 | 91059626 | 26-30 |
| 13 | 104041811 | 108162723 | 25-28 |
| 21 | 40305282 | 46899964 | 28-35 |

**Top Beagle findings**

| Chr | start | end | # of pairs (Max=55) |
|---|---|---|---|
| 6 | 25684356 | 27237736 | 15 -17 |
| 6 | 93731159 | 104759760 | 15-17 |
| 11 | 48651447 | 56368374 | 15 |
| 12 | 57427666 | 62233961 | 15 |
| 13 | 82110402 | 103876781 | 15-19 |

**IBS regions**




**PEDIBD**

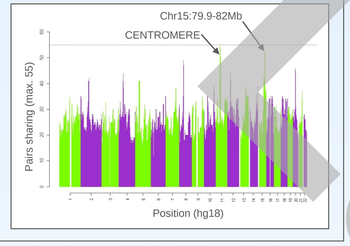



Figure 2: Four methods were used to identify shared segments from eleven individuals with bipolar disorder

**Shared Segments Identified by Multiple Methods**

| Region[1] (hg18) | Sharing | | Notes |
|---|---|---|---|
| | *Method* | *Max. sharing*[2] | |
| Chr6:~20-30MB | Germline | 20 pairs (~7/11) | MHC |
| | Beagle | 17 pairs (~6/11) | |
| | IBS | 0.3Mb/154 SNPs (11/11) | |
| Chr11:~48-56Mb | Beagle | 15 pairs (~6/11) | Centromere False positive |
| | PEDIBD | 11/11 | |
| Chr13:~82-103Mb | Germline | 30 pairs (~8/11) | Contains *SLTRK1*, implicated in trichotillomania |
| | Beagle | 19 pairs (~7/11) | |
| Chr15:~79.9-82Mb | IBS | 2.4Mb/204 SNPs (11/11) | |
| | PEDIBD | 52 pairs (~10-11/11) | |

1. Some methods identify only part of region listed
2. For IBS lists length of region for which all 11 share and number of markers in the region.
   For all others: Maximum number of pairs(approximation of number of individuals who could be sharing)

### *Exome Sequencing*

- Sequenced five female BPI individuals.
- Aligned with BWA(5) and followed by Genome Analysis Toolkit (GATK; 6-7) pipeline.

| Coverage | |
|---|---|
| % target bases ≥10X | 87-89% |
| Mean target coverage | 58-62X |

- Start with stringent filtering:
  An alternate allele in all five individuals
  Not present in an alternate study (autism family)
  Non-synonymous alteration
  Not present in dbSNP

| | | | |
|---|---|---|---|
| chr15:76845785 | *ADAMTS7* ADAM metallopeptidase with thrombospondin type 1 motif, 7 | S1175P | TOLERATED |

- This SNP is a few Mb from peak sharing on chr15 by IBS regions and PEDIBD.
- PEDIBD still shows 9-10 of 11 individuals sharing.
- SNP verification and genotyping in additional individuals in this family is in progress.

## Conclusions

- This family appears to carry a genetic factor increasing risk for mood disorders, including bipolar disorder.
- We have identified regions of potential interest,
- Verification of novel SNP on chromosome 15 and additional genotyping in the family are warranted.
- Whole genome sequencing on three individuals is in progress.
- Future analysis will focus on shared segments.

## References

1. Gusev A, Lowe JK, Stoffel M, Daly MJ, Altshuler D, Breslow JL, et al. Whole population, genome-wide mapping of hidden relatedness. Genome Res. 2009;19(2):318-26. PMCID: 2652213.
2. Browning SR, Browning BL. High-resolution detection of identity by descent in unrelated individuals. Am J Hum Genet. 2010;86(4):526-39. PMCID: 2850444.
3. Li X, Yin X, Li J. Efficient identification of identical-by-descent status in pedigrees with many untyped individuals. Bioinformatics. 2010;26(12):i191-8. PMCID: 2881406.
4. Thomas A, Camp NJ, Farnham JM, Allen-Brady K, Cannon-Albright LA. Shared genomic segment analysis. Mapping disease predisposition genes in extended pedigrees using SNP genotype assays. Ann Hum Genet. 2008;72(Pt 2):279-87. PMCID: 2964273.
5. Li H, Durbin R. Fast and accurate short read alignment with Burrows-Wheeler transform. Bioinformatics. 2009;25(14):1754-60. PMCID: 2705234.
6. McKenna A, Hanna M, Banks E, Sivachenko A, Cibulskis K, Kernytsky A, et al. The Genome Analysis Toolkit: a MapReduce framework for analyzing next-generation DNA sequencing data. Genome Res. 2010;20(9):1297-303. PMCID: 2928508.
7. DePristo MA, Banks E, Poplin R, Garimella KV, Maguire JR, Hartl C, et al. A framework for variation discovery and genotyping using next-generation DNA sequencing data. Nat Genet. 2011;43(5):491-8. PMCID: 3083463.

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