The Role of the Oxytocin Receptor Gene (*OXTR*) in Autism Spectrum Disorders (ASD)

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Autism Spectrum Disorders

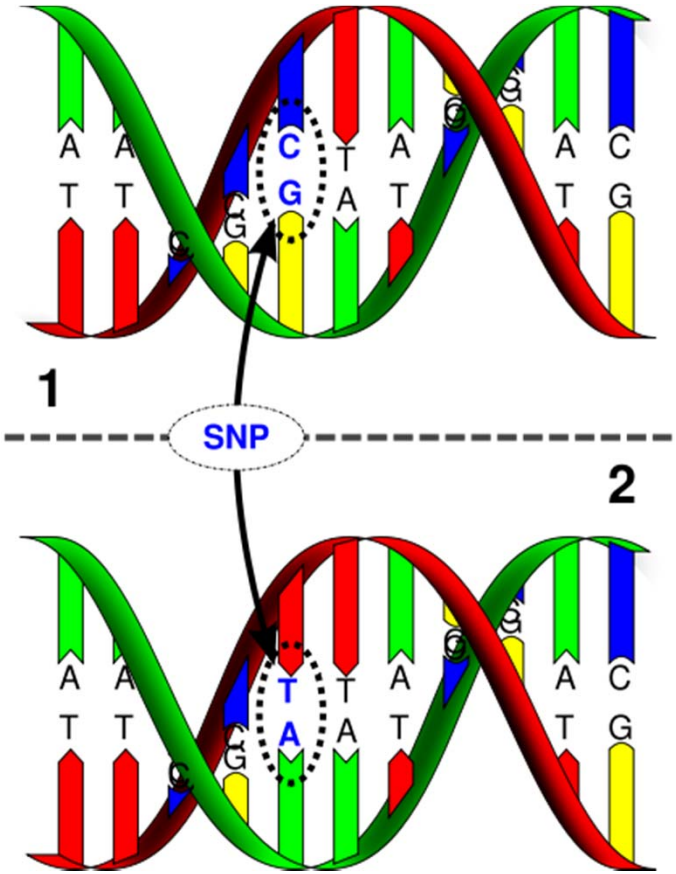
- Autism is a neurodevelopmental disorder that is characterized by deficits in certain aspects of human behavior (American Psychiatric Association) :
  - Social interaction
    - Lack of eye to eye contact
    - Failure to develop peer relationships
  - Communication
    - A delay in spoken language without attempt of another form of communication
  - Restricted/repetitive behaviors
    - Repetitive motor actions
    - Inflexibility to routine/resistance to change
    - Preoccupation with objects

Assessments

- ADI
  - (93 Items) and presents the results as a score.
  - higher the score the more severe the autism.
  - Based on the cutoff of the score of the impaired areas.
- ADOS
  - Based upon various activities such as social interaction and communication to receive a score.

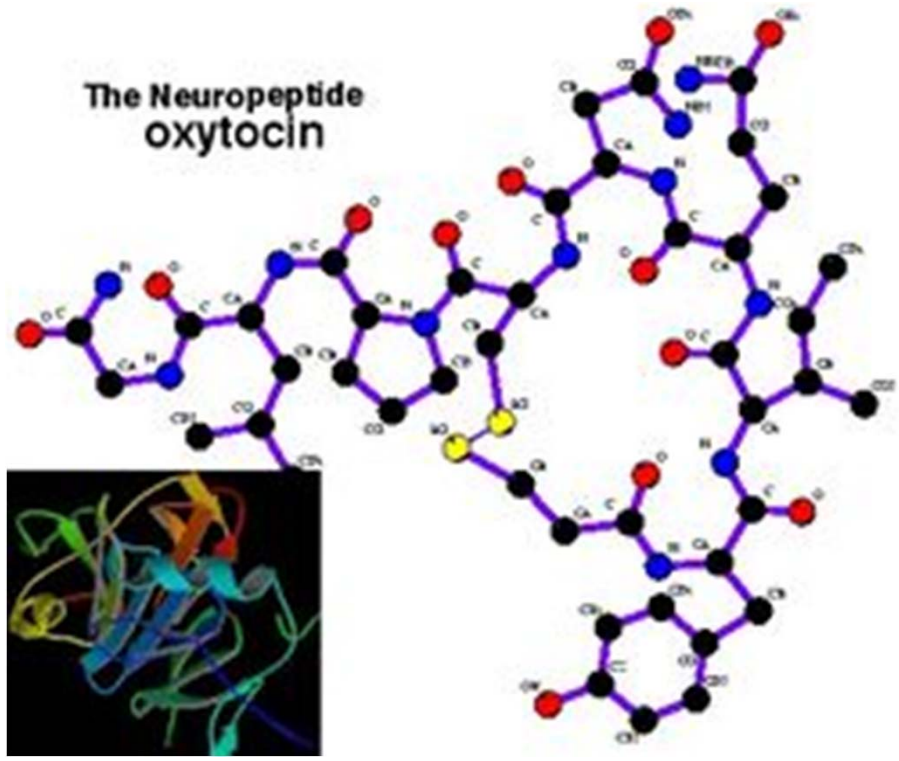
Point Mutations

- Single Nucleotide Polymorphism (SNPs)- A variation in a single base pair.



Oxytocin

- Oxytocin is a nonapeptide
- synthesized in the paraventricular and supraoptic nucleus of the hypothalamus.
- released into the blood stream through axon terminals in the posterior pituitary (Carter, 2006).



What does oxytocin have to do with autism?

- Since oxytocin regulates social behavior which is a deficit in Autism, genes coding for oxytocin as well as its receptor (OXTR) have become candidate genes (Carter, 2006).

Transmission

- Wu et. al genotyped rs2254298 and rs53576.
- Over transmission of SNPs (Wu, 2005).
- Suma Jacob et al. genotyped same SNPs with a Caucasian population.
- Association with rs2254298.
- Frequency of one allele (G allele) was less than the frequency of A allele in the Han population.
- Result of different linkage pattern (Jacob, 2007).

Genes Controlling Affiliative Behavior in ASD

- There is evidence for the association with the OXTR and OXT genes
- variety of multivariate phenotypes with rs226493 of the OXTR gene and rs2740204 of the OXT gene for:
  - Communication Skills
  - Stereotyped Behaviors

(Yrigollen, 2008)

Objectives

- Association between SNPs due to their proximity.
- no association means the SNP will have an equal chance transmission
- If there is association, the SNP will be transmitted at a higher frequency.

Hypothesis

- H_A =A positive association will appear between clinical diagnosis of ASD and at least one of the SNPs captured in this study if *p* value is less that .01 for the intensity of the alleles.

- H0=None of the seven SNPs captured in this will demonstrate any association with clinical diagnosis of autism. This will be indicated by a calculated *p* value that is greater than .01 for allele intensity.

Methods and Materials

Participants

- A sample of probands with autism and their families available through the Yale Child Study Center.
  - 525 participants
  - 177 classified as probands.
  - 151 nuclear families.
  - The majority of the sample was Caucasian
  - Approved by Yale University IRB (Yrigollen,2008)

Phenotyping

- Probands and their relatives were evaluated with:
  - ADI and ADI-Revised
  - ADOS
  - Autism Diagnostic Observational Schedule-Generic (ADOS-G); Vineland Adaptive Behavioral Scales.

Genotyping

- SNPs Studied:
  - rs237884
  - rs1042778
  - rs36062132
  - rs237893
  - rs4686302
  - rs180789
- Previously extracted DNA from blood or saliva collected from probands and their families.
- DNA concentrated or diluted to100 nanograms/ microliter.

Genotyping (continued)

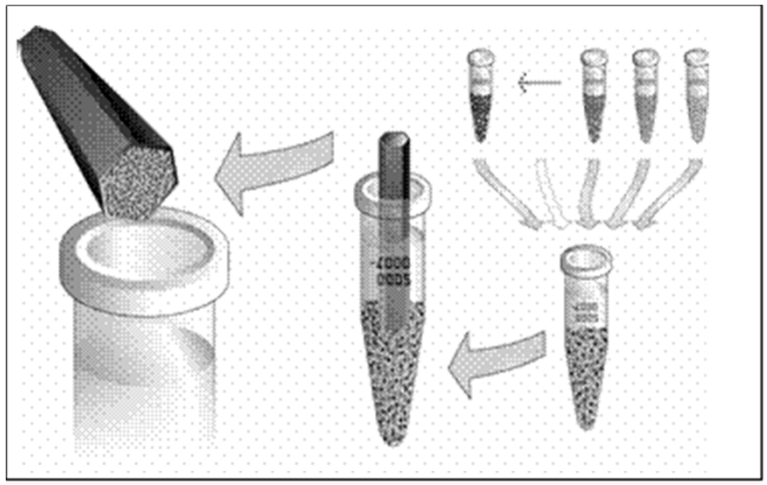
- Concentrated DNA (a total of 96 samples) ran on 4% agrose gels stained with ethidium bromide.
- This process checks the quality of the DNA.
- KECK run an array which is also run in 96 well format.

Genotyping (continued)

- KECK analyzes samples on custom Illumina bead arrays.
- Each bead has a complementary sequence to a SNP so it hybridizes to the SNP.
- Each bead also has a coding sequence which shows what beads code for what SNP, which allows us to know which SNP is present.

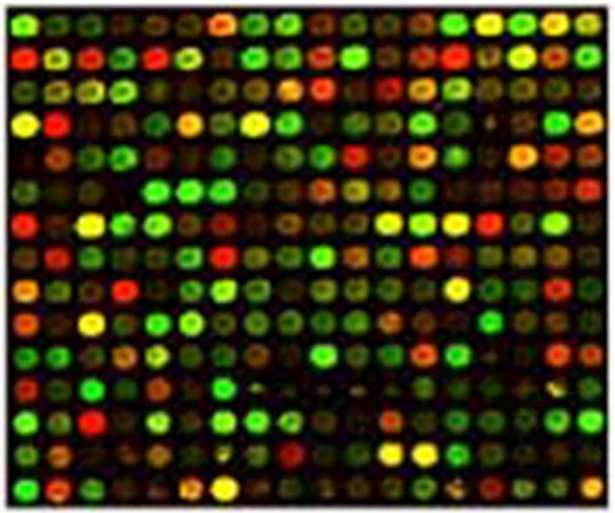
Genotyping (continued)

- Beads are assembled into microwells at the end of a fiber optic bundle.



Genotyping (continued)

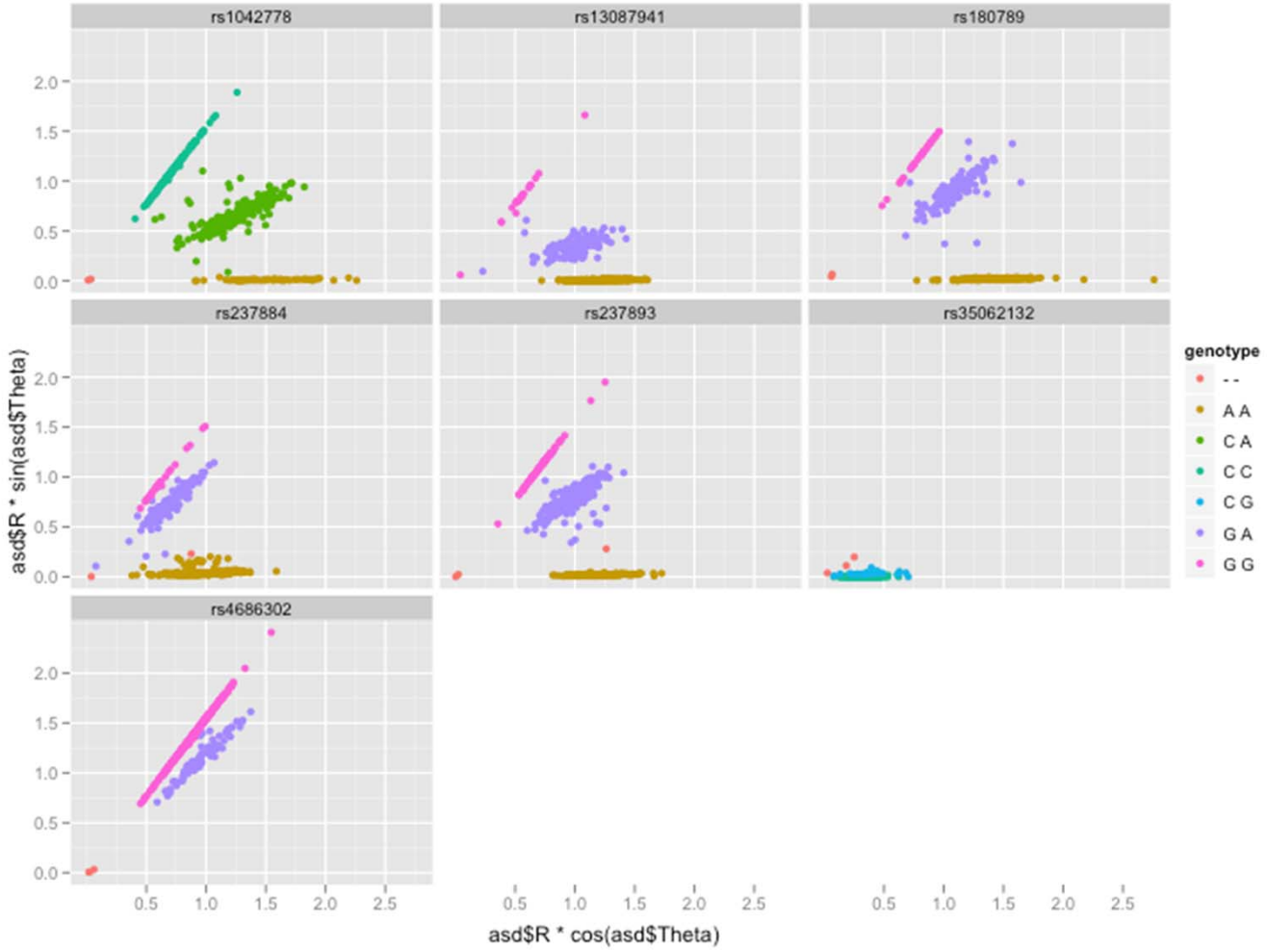
- These hybridized bundles are then used to make an array.
- Shows intensity of one florescent if one allele present, if both shows florescence the SNP is heterozygous.



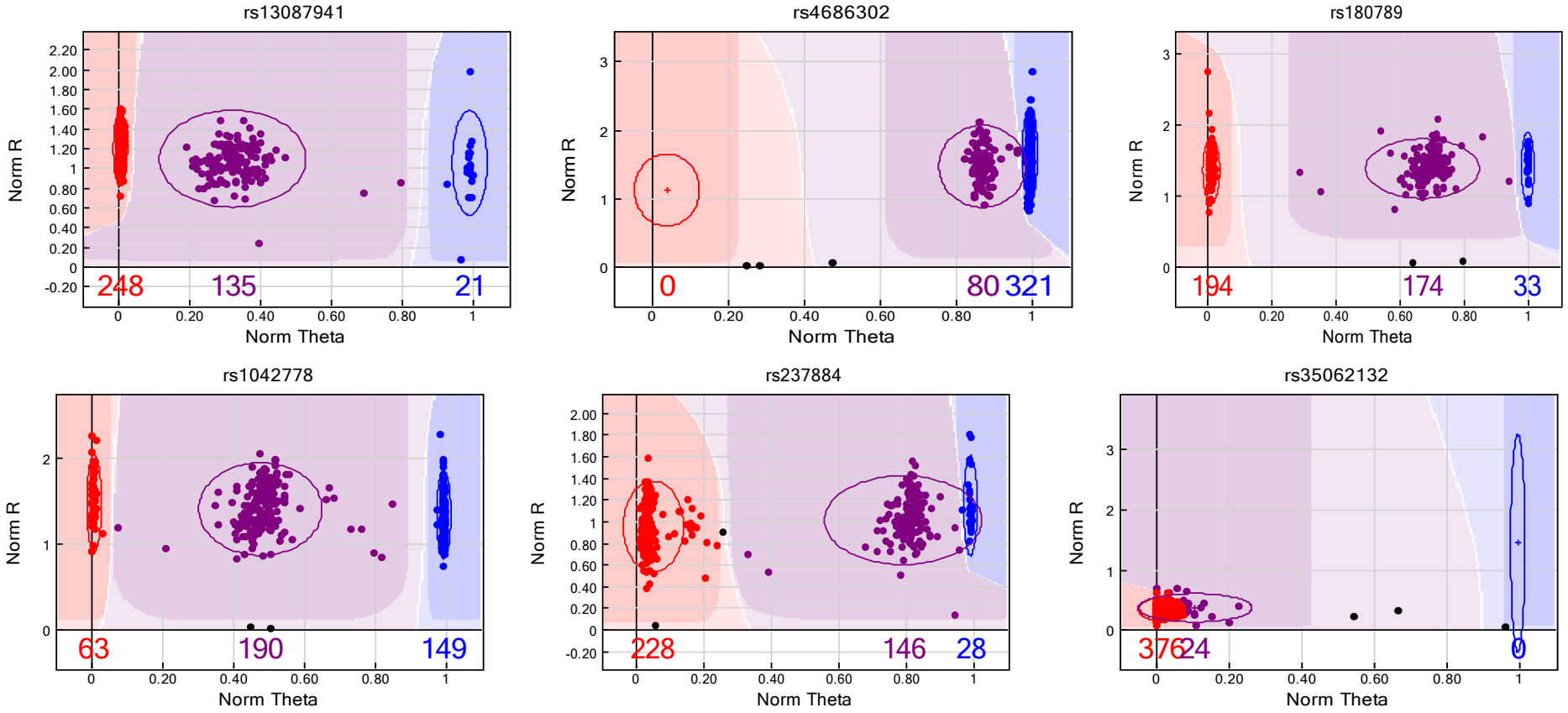
Analysis

- For data cleaning and processing we used "R".
- FBAT (Family Based Association Test) was used (Horvath, Xu, Laird, 2001) for statistical analyses.
- null hypothesis of "no association in the presence of linkage"

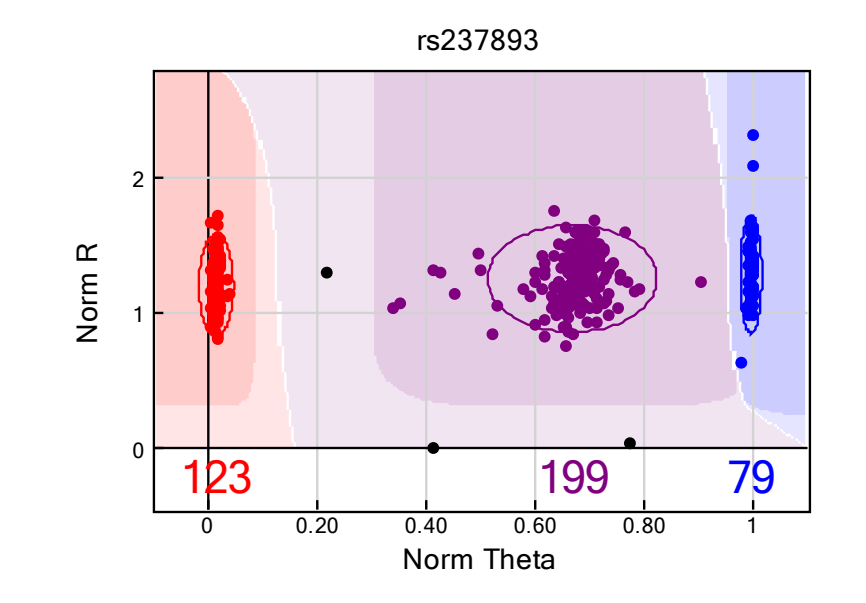
Results


rs1042778
rs13087941
rs180789
rs237884
rs237893
rs35062132
rs4686302
asd$R * sin(asd$Theta)
asd$R * cos(asd$Theta)
genotype
- -
A A
C A
C C
C G
G A
G G

RESULTS (CONTINUED)



RESULTS (CONTINUED)



Discussion

- low p value for rs237893 demonstrates an association between rs237893 and clinical diagnosis.
- rs237893 shows high transmission disequilibrium.
- excess transmission of the GA allele.
- rs237893 has a significant role in the clinical diagnosis of ASD.

Discussion Continued

- rs237893 is located on an intron (http://www.ncbi.nlm.nih.gov/).
- genetic contribution of this SNP to ASD cannot be ruled out. (Wu, 2005).
- Possibility of rs1042778 having a role in clinical diagnosis of ASD.

Future Research

- A study with a larger sample size.
- Other neurobiological studies suggest vasopressin plays a similar role in autism as oxytocin .
- future research should evaluate the genetic relationships and a possibility in autism (Wu ,2005).

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