Reviewer's report

**Title:** Evaluation of microdeletions and microduplications associated with cognitive impairment in a large cohort of subjects with autism spectrum disorders identifies duplications at 15q11-q13, 22q11 and Xp11/TM4SF2

**Version:** 2  **Date:** 27 April 2008

**Reviewer:** James S Sutcliffe

**Reviewer's report:**

Cai et al report an analysis of 279 unrelated autism probands for evidence of structural variation (SV) at loci previously associated with cognitive impairment (CI). The authors use commercial MLPA kits to survey their cohort for these previously identified CI sites, then proceed to confirm potential positives using FISH and qPCR. Recent studies provide ample precedent for involvement of SV at CI loci in cases of autism, however, the extent of this phenomenon is not clear. This is a straightforward report that presents very important further information on the involvement of SV at specific loci in subjects carrying an autism diagnosis. These are important observations (e.g. TM4SF2) and will add to the emerging picture of autism genetic architecture. A few questions that arise from this reviewer’s reading of the manuscript:

1) There is no discussion of testing or comparing control subjects for SV at the loci tested. How are we to evaluate disease association by only assessing a disease population? Even if for some variants, a de novo origin is determined, de novo SV by itself is not conclusive. There is a baseline rate of de novo variation in the genome. The authors should comment on whether SV at any of the loci they identify is seen in non-clinical samples. (minor essential revision)

2) Have any of the samples assessed in this work been analyzed in previous studies of SV in autism? Several reports have now been published in which overlapping sample sets are involved, particularly involving the AGRE resource to which the authors are key contributors. The readers may be left with the impression that different publications involving different but complementary analyses reflect unique observations when in fact common samples were analyzed. The authors should indicate which if any of the families with SV findings were included in previously published reports, such as those by Sebat et al and Szatmari et al. For families included in a repository, database identifiers should be provided, such as AGRE or NIMH IDs.

3) In a related matter, if any of these SV events have been previously reported, they may be reflected in the Database of Genomic Variants for autism (“Autism CNV Database”) maintained by The Centre for Applied Genomics in Toronto. It would be helpful if the authors can be sure to indicate which SV presented here has been reported elsewhere and/or is reflected in the TCAG database. (I think this is generally the case) (minor essential revision)
4) In this reviewer’s view, it would have been nice to see representative examples of MLPA results indicating putative SV, which was subsequently confirmed (or refuted). While the paper has three figures already, MLPA is a very powerful method and I think it adds something for the reader to see representative data. In this regard, the authors careful work attributing initially positive MLPA results to altered MLPA probe binding caused by SNP variation provides a nice illustration of a potential complicating factor for this method. (discretionary revision)

Minor comments (discretionary revisions):

5) In the conclusions section on p21, the authors write “15q11-q13 duplications, including two novel microduplications, might indicate an etiological pathway of aberrant glutamate signaling interacting with epigenetic factors in some ASD individuals.” The rationale for this conclusion is clear. In my reading of the manuscript, there was no foundation laid that connects 15q11-q13 gene or genes to glutamate signaling. It would be helpful to the reader if the authors could make this clear.

6) The very beginning of the FISH methods section starts with “MLPA”. This appears to be an editing error and should be deleted.

Level of interest: An article of outstanding merit and interest in its field

Quality of written English: Acceptable

Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests:

I declare that I have no competing interests