Reviewer's report

Title: Breakpoint characterization of a novel large intragenic deletion of MUTYH detected in a MAP patient: Case report

Version: 2 Date: 18 August 2011

Reviewer: ROSSELLA TRICARICO

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Torrezan et al. report on a large genomic deletion, spanning exon 4-16 of the MUTYH gene, detected in the homozygous state in a MAP patient. aCGH analysis, sequencing and in silico-based evaluation, used to characterize the deletion breakpoints at base pair level, identified the presence of an Alu sequence adjacent to the distal breakpoint and of a 2 bp insertion at the junction site. These findings suggest that genomic rearrangements could represent another mechanism of the loss of function of MUTYH gene in MAP and a possibly involvement of the non-homologous end-joining (NHEJ) repair mechanisms. Very recently, the same deletion was found in a MAP patient in compound heterozygosity with one of the most common MUTYH Caucasian mutation p.Gly396Asp (Rouleau et al., Clin Genet. 2011Sep;80(3):301-3).

The report is well written and the design of molecular analyses is appropriate.

MINOR ESSENTIAL REVISIONS:

Abstract

1. The statement “Most MUTYH pathogenic variants are missense mutations, and currently, no gross genomic deletions have been described. Case Presentation: We have identified the first large deletion in the MUTYH gene: a >4.2 kb deletion encompassing exons 4-16…” is not correct because the c.348+33_*64+146del4285 deletion has already been identified in a MAP patient (Rouleau et al., Clin Genet. 2011Sep;80(3):301-3). This data should be presented, and the implications of finding the same rearrangement in (presumably) different ethnic groups should be discussed.

Background:

1. The statement in the second paragraph “Most biallelic MUTYH mutation carriers have between ten and several hundred polyps, usually with later onset compared to FAP patients” is incomplete because the clinical presentation of MAP also includes a number of patients with early-onset CRC and none or few polyps (>10) (Nielsen et al., Crit Rev Oncol Hematol 2010, 1-16.). Please refer to this data.

2. Please refer to Rouleau et al., Clin Genet. 2011Sep;80(3):301-3 (Fourth paragraph, last sentence).
Case Presentation:

1. The case report should contain a detailed description of the patient including more detailed information on ethnic origin. More detailed information should also be provided on clinical and family history, tumour histology, presence of extracolonic manifestations, any other tests that were carried out (e.g. APC screening), phenotype of the affected proband’s sister etc. It would be useful to have a figure showing the pedigree of the family.

2. The geographic/ethnic origin of the controls and of the 51 familial CRC patients should be the same as that of the proband, and should be specified.

Identification and characterization of the deletion

1. Synthetic information about the primers set and condition used for the junction fragment sequencing analysis should be provided.

2. Please specify if a region of another gene is deleted downstream of the 3’end of the MUTYH gene.

3. The authors should adhere to the Human Genome Variation Society (http://www.hgvs.org/rec.html) recommendations for the description of genomic deletions. These are some points to consider in this regard:

   - The HGVS gene nomenclature is in italic character. Please check the name of the genes in abstract, background, first sentence and in case presentation, second paragraph, second and third sentence.

   - The sequence deletion at genomic level as described as chrX:g.32,218,983_32,984,039del and using “del” after an indication of the first and last nucleotide(s) deleted. In addition, a combination of a deletion and insertion at the same site is described using the format c.112_117delinsTG. These designations should be used in the description at genomic and cDNA level of the variant “chr1: 45794768_45799052del4285” and “c.348+33_*64+146del4285” identified in the proband.

   - The description of the homozygous changes in a recessive disease are the following: c.[change allele 1];[change allele 2] (last HGVS update August 4, 2011). It should be used in the genotype description of the proband (sixth paragraph, fourth sentence).

   - Please, note that the chromosome build used should always be mentioned (e.g. NCBI Build 36.1 or UCSC Feb. 2009 (GRCh37/hg19) assembly). It should be add to Hg19 in Identification and characterization of the deletion, third paragraph, fourth sentence and Legends, figure 1, last sentence.

4. In the identification and characterization of the deletion, eight paragraph, eleventh sentence, some references should be specified.

5. The c.348+33_*64+146del deletion has already been identified in a MAP
patient by qPCR-HRM and aCGH (Rouleau et al., Clin Genet. 2011Sep;80(3):301-3). This reference should be included and discussed.

Legends

1. Figure 1, section G: The description is not clear. Please clarify and indicate what the light gray test indicates.

Figures

1. Figure 1, section D: Please indicate the location of the 16R2 primer.

In addition, the following language corrections should be introduced:
- Background, First paragraph, third sentence: "Mut Y" should be replaced with "mutY"
- Background, Third paragraph, sixth sentence: "~" should be replaced with "approximately"
- Identification and characterization of the deletion, eight paragraph, thirteenth sentence: "8-oxyG" should be replaced with "8-oxoG"
- Figure legends, figure 2, second sentence: "USCS" should be replaced with "UCSC"

Level of interest: An article of importance in its field

Quality of written English: Acceptable

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