Letter to the Editor

Am. J. Hum. Genet. 75:517–519, 2004

Reply to Repping et al.

To the Editor:

We welcome the enormous contribution that Repping and colleagues have made to the elucidation of the DNA sequence and organization of the Y chromosome, but many questions remain unanswered after the sequencing of the Y chromosome of one man (Skaletsky et al. 2003). It was appreciated, almost a decade ago, that the structure of the *AZFc* region is particularly variable; six independent deletion events and four duplications that affect one short section of this region (50f2/C), together representing ~8% of normal men, were identified by Jobling et al. (1996), and this study could have detected only a small proportion of the total *AZFc* variation. Yet, it provides a useful benchmark for an assessment of our current understanding. We can now define the molecular basis of one of the deletions described in 1996, the haplogroup-12 "small" 50f2/C deletion (Fernandes et al. 2004; Repping et al. 2004*b*), and possibly a second (if the "small" 50f2/C deletion in haplogroup 2 [Jobling et al. 1996] corresponds to the b2/b3 deletion in YCC haplogroup F*[xHK] or I [Repping et al. 2004*b*]), but the 50f2/C duplications all fall on haplotypic backgrounds different from those of the b2/b4 duplications (which include 50f2/C) described so far (Repping et al. 2003). Thus, researchers have still not accounted for at least 8 of 10 rearrangements reported in 1996. It seems that our current methods, whether based on SNVs/SFVs or on plus/minus STSs, allow us to describe only a small proportion of the variation present in this region.

Are plus/minus STSs, nevertheless, more useful than SNVs/SFVs for characterizing *AZFc* variation (Repping et al. 2004*a* [in this issue])? It is a matter of opinion. Even for the best-characterized variants, the gr/gr and g1/g3 (also known as "b2/b3") deletions, it is unclear whether the independent deletions on different lineages represent true recurrent mutations—taking place at the same recombination site each time—or whether the recombination events have occurred in different locations within the amplicons on different occasions. In the latter case, conflation of different structures—which could have different gene contents—by plus/minus STSs would be a weakness of this classification scheme, and discrimination between them by SNVs, a strength (fig. 1 of Repping et al. [2004*a*] [in this issue]). It would, however, seem rash to rely on either of these two methods alone—FISH, used by Repping et al. (2003); Southern blotting, used by Fernandes et al. (2002, 2004); and quantitative PCR can all be helpful in defining the structures. But most important of all, this work highlights the importance of an evolutionary understanding of the Y chromosome, and we particularly welcome Repping et al.'s acceptance of this evolutionary approach.

Evolutionary interpretations must, however, be made with caution—we should avoid the "fallacy of the contemporary ancestor" (Jobling et al. 2004). Modern inverted Y chromosomes (see fig. 3 of Repping et al. [2004*b*]) are not the ancestors of haplogroup-N chromosomes, and their frequencies do not indicate which mutational pathway was followed. The best guide to the pre-N structure may be provided by haplogroup-O chromosomes, a sister clade to N in the current Y phylogeny (Jobling and Tyler-Smith 2003) and thus the closest known outgroup. The b2/b3 inversion has indeed been reported in haplogroup O (Repping et al. 2004*b*); if it was present in the common ancestor of the two lineages, the haplogroup-N deletion would result from a g1/g3 deletion following this b2/b3 inversion, rather than a b2/b3 deletion following a g1/g3 inversion. If so, the conclusions from the SNV-based study (Fernandes et al. 2004) would be more accurate than those from the plus/minus STS–based one (Repping et al. 2004*b*).

The present discussion can take place only because our methods for characterizing *AZFc* structures are pitifully inadequate. Rather than behaving like the proverbial group of blind men who encounter an elephant from different sides and insist on describing it from their own favorite partial perspectives, we should assume that all the inversions, duplications, and deletions that are permitted by the sequence will occur, limited only by the winnowing of natural selection. The resulting structures may differ, by many rounds of rearrangement, from the modern haplogroup-R GenBank sequence, but use of the SNP-based phylogeny (Jobling and Tyler-Smith 2003) may allow us to understand the relationship between these structures. It would be even better to develop radically improved ways of elucidating the entire structure

so that we can obtain a reasonably complete view of this complex and evolutionarily labile region.

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References

Fernandes S, Huellen K, Goncalves J Dukal H, Zeisler J,, Rajpert De Meyts E, Skakkebaek NE, Habermann B, Krause W, Sousa M, Barros A, Vogt PH (2002) High frequency of *DAZ1/DAZ2* gene deletions in patients with severe oligozoospermia. Mol Hum Reprod 8:286–298

Fernandes S, Paracchini S, Meyer LH, Floridia G, Tyler-Smith C, Vogt PH (2004) A large AZFc deletion removes *DAZ3/DAZ4* and nearby genes from men in Y haplogroup N. Am J Hum Genet 74:180–187

Jobling MA, Hurles ME, Tyler-Smith C (2004) Human evolutionary genetics. Garland Science, New York and Abingdon, United Kingdom

Jobling MA, Samara V, Pandya A, Fretwell N, Bernasconi B, Mitchell RJ, Gerelsaikhan T, Dashnyam B, Sajantila A, Salo PJ, Nakahori Y, Disteche CM, Thangaraj K, Singh L, Crawford MH, Tyler-Smith C (1996) Recurrent duplication and deletion polymorphisms on the long arm of the Y chromosome in normal males. Hum Mol Genet 5:1767–1775

Jobling MA, Tyler-Smith C (2003) The human Y chromosome: an evolutionary marker comes of age. Nat Rev Genet 4: 598–612

Repping S, Korver CM, Oates RD, Silber S, van der Veen F, Page DC, Rozen S (2004*a*) Are sequence family variants useful for identifying deletions in the human Y chromosome? Am J Hum Genet 75:514–517 (in this issue)

Repping S, Skaletsky H, Brown L, van Daalen SK, Korver CM, Pyntikova T, Kuroda-Kawaguchi T, de Vries JW, Oates RD, Silber S, van der Veen F, Page DC, Rozen S (2003) Polymorphism for a 1.6-Mb deletion of the human Y chromosome persists through balance between recurrent mutation and haploid selection. Nat Genet 35:247–251

Repping S, van Daalen SK, Korver CM, Brown LG, Marszalek JD, Gianotten J, Oates RD, Silber S, van der Veen F, Page DC, Rozen S (2004*b*) A family of human Y chromosomes has dispersed throughout northern Eurasia despite a 1.8-Mb deletion in the azoospermia factor c region. Genomics 83: 1046–1052

Skaletsky H, Kuroda-Kawaguchi T, Minx PJ, Cordum HS, Hillier L, Brown LG, Repping S, et al (2003) The male-specific region of the human Y chromosome is a mosaic of discrete sequence classes. Nature 423:825–837

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0002-9297/2004/7503-0019$15.00