...ve the disease-specific haplotype. DSCs 12, 10, 1, 3, and 2 were disease-specific among patients with XDP ...sted here as well as in a previous report.[9] The 1,666-bp deletion was detected by PCR among all affected and unaffected Filipinos and in the ethnic panels (data not shown) and was considered to be nonspecific. By contrast, the SVA retrotransposon was clearly disease specific in our data set (figs. 1*b* and 3). These disease-specific variants, the DSCs and the SVA retrotransposon, were located around the *TAF1* and *MTS* genes.

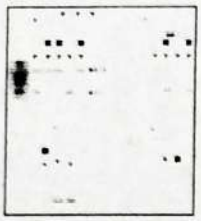

Figure 3.

The SVA insertion in an additional seven patients with XDP and their relatives, from four families. Information on these patients is given in table 1 (patients 14–20).

Table 5.

All Nucleotide Variants in the *DYT3* Critical Region on Xq13.1

Previously Proposed *MTS* Transcripts

Next, northern hybridization and long RT-PCR analysis were undertaken to confirm the structures and expression of the *TAF1* and *MTS* genes. Northern analysis showed that the hybridization signal seen at ~7 kb of *TAF1* had a tendency toward reduction in patient caudate and that the *MTS* transcript lengths reported elsewhere[9] were not detectable in RNAs from either patient or control tissues (fig. 2*b*). Long RT-PCR analysis showed that the PCR fragment pattern from *MTS* was identical to that from *TAF1* (fig. 4*a*). If the *MTS* transcripts had lacked upstream exons 1–29 and exon 38, spanning >2 kb, as shown in the previous report,[9] the northern analysis would have detected the corresponding signals at ~2–5 kb. Moreover, our long RT-PCR analysis showed that exons 3 and 4 from *MTS* (gray exons in fig. 4*b*) are attached at the 3′ end of exon 38 of *TAF1*. Quantitative RT-PCR analysis by the TaqMan assay with five probes designed to detect the previously proposed *MTS* transcripts MTS-V4—which was regarded as a candidate transcript for *DYT3* because of a deduced amino-acid change[9]—MTS-37/1, MTS-37/3, MTS-2/3, and MTS-32′/34′ (fig. 4*b*) yielded very weak and irregular amplification signals that did not allow quantification of expression levels in the caudate nucleus (table 6), whereas the probe MTS-3/4 for exons 3 and 4 attached to *TAF1* yielded a relatively weak but regular signal (table 6). These results consistently suggested that the previously proposed *MTS* transcripts may be extremely rare or unexpressed and that at least exons 3 and 4 of *MTS* may be just an additional part of the 3′ UTR of *TAF1*, rather than part of a new distinct short gene.

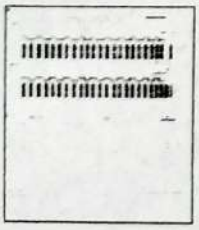

Figure 4.

Long RT-PCR and the alternative exons of *TAF1*. *a*, Long RT-PCR analysis. The broken line indicates an expected cDNA fragment of *TAF1* with the long RT primer on the end of exon 38 (*short arrow*). By subsequent PCR with the use of the long cDNA, six lanes—TA02, ...

Table 6.

Linearity of Amplification Curve from Threshold Cycle (Ct) = 25 to Ct = 35[Note]

Decreased Expression of *TAF1* in the XDP Brain

At least 10 new alternative splicing exons around *TAF1* were found by long RT-PCR analysis (fig. 4*b*), but neither the DSCs nor the SVA insertion were located in any known and predictably translated regions of the *TAF1* exons. We then