

## Abstract 101

### FRAGILE X SYNDROME: A SIMPLE MOLECULAR DIAGNOSIS BY MEANS OF A TRIPLE METHYLATED-SPECIFIC PCR ASSAY

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Fragile X or Martin Bell syndrome (FXS, OMIM 309550) is an X-linked dominant disorder with reduced penetrance. This syndrome is one of the most common types of inherited mental impairment. The most frequent cause of this disorder is the expansion of a CGG trinucleotide repeat in the 5' UTR of the *FMR1* gene. This expansion leads to DNA hypermethylation, and silencing of the *FMR1* gene resulting in the absence or severely reduced expression of the FMR1 protein, FMRP. Much less often, other mutational mechanisms, such as deletions of *FMR1*, can also be involved. The FXS is inherited from females, who typically carry an unstable premutation allele of the CGG-repeat tract. Premutation carriers are themselves at risk of premature ovarian failure (POF) and of fragile X-associated tremor ataxia syndrome (TAS). The objective of the present work was the optimization of a simple technique based on a methylation specific triple PCR assay for the diagnosis of the fragile X syndrome. DNA samples were previously treated with sodium bisulfite and then they were simultaneously amplified by a triple specific PCR assay using three sets of primers to amplify the non-methylated and methylated alleles as described in the literature. We studied two families, both with previous cytogenetic tests for FXS. One of the groups was represented by five members, where the youngest was positive for the cytogenetic test. The second family group included six members, where three showed evidence of mental retardation and psychomotor difficulties. This evaluation allowed us to discriminate between normal, premutation and full mutation alleles among members of the families tested. Because of its simplicity, low cost and almost immediate results, this technique appears to be an excellent alternative to incorporate into the laboratory routine for patients with symptoms suggest fragile X syndrome. In conclusion, this analysis allows us to discriminate between all the three FMR1 alleles (normal, premutated and fully mutated) in both genders, with its subsequent implications in genetic counseling for both the patients and their relatives over the risk of being a premutation carrier or transmitting the disease.

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