AN INHERITED P53 POINT MUTATION IN A CANCER PRONE FAMILY
WITH LI-FRAUMENI SYNDROME

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ABSTRACT

Somatic cells derived from members of a cancer-prone family representing three generations were used to assess mutations in selected regions of p53. Fibroblast DNAs from four family members--the proband, his brother, their father and a paternal aunt, yielded an identical point mutation in codon 245 in only one allele of the p53 gene. This mutation, involving G to A transition (GQC → GAC) leads to substitution of aspartic acid for glycine at that codon in p53 protein and is not present in NSF DNAs of the proband’s mother or his paternal grandfather, neither of whom are in the cancer-prone lineage. Despite the observed mutation, the level of p53 protein detected in these fibroblasts is comparable to low levels observed in normal control fibroblasts. This is in contrast to the high levels of mutant p53 usually found in tumor cell lines. Thus the mutant p53 in these fibroblasts appears to behave differently as compared to the mutant p53 previously detected in transformed cells. Given the inherited nature of this p53 mutation, the demonstrated role of p53 in tumorigenesis and the location of mutation in a region of the gene known to be critical for its function, it appears that we have identified a primary genetic alteration in this Li-Fraumeni family, a defect which may predispose them to increased susceptibility to cancer.
Familial cancer syndromes provide opportunities to examine the mechanisms of inherited susceptibility to cancer as well as more general processes involved in the development of malignancy. Tumor suppressor genes have been implicated in many inherited as well as in sporadic form of malignancies (for reviews see ref. 1-3). A large body of experimental evidence supports the concept of tumor formation by loss-of-function mutations in suppressor genes as predicted by the two-hit model of Knudson (4) and DeMars (5) involving inactivation of both alleles for manifestation of the tumorigenic phenotype. The tumor suppressor gene, p53, has been shown to have sustained numerous genetic alterations in diverse neoplasm, usually exhibiting loss of one allele and point mutation in the other. We have been studying predisposing genetic factors in a specific cancer-prone family diagnosed as having Li-Fraumeni syndrome, which is characterized by the early onset of diverse neoplasms, as well as occurrence of multiple primaries in single individuals (6,7). Although p53 mutations in other studies are reported to be tumor specific (for reviews see refs. 8 and 9), we reasoned that if a defect in the p53 gene was central to the tumorigenesis in this cancer-prone family, the alterations in p53 gene may be detected in at least one allele in noncancerous somatic cells. Normal skin fibroblast (NSFs) derived from members of this family, representing three generations, were analyzed for alterations in the mutational hot spots of the p53 gene by polymerase chain reaction (PCR) amplification and direct sequencing of the PCR product. Recently we (10) and others (11) have described germ-line p53 mutations in Li-Fraumeni cancer-prone families. Here, we briefly summarize our findings on the inherited codon 245 mutation in the p53 gene of fibroblasts derived from members of a specific cancer-prone family.

**GERM-LINE P53 MUTATION IN MEMBERS OF A CANCER-PRONE FAMILY:**

NSF cell lines derived from members of three generations of a cancer-prone family (Fig. 1) were utilized to assess the status of p53 gene. Utilizing p53 cDNA as a probe, we did not detect any major alteration in the p53 gene by Southern or Northern blot analyses (data not shown).

We, therefore, analyzed the family NSFs for subtle