We describe a young patient with no known family history of cancer who presented at 18 months with 2 advanced primary tumors, choroid plexus carcinoma and adrenal cortical carcinoma. Immunohistochemical studies demonstrated high levels of nuclear p53 protein expression in both tumors, as well as in the adjacent normal-appearing adrenal cortical cell nuclei of the adrenal gland. The immunohistologic distribution of elevated p53 expression suggests that this individual has a de novo germline mutation affecting p53 gene expression.

Report of a Case

An 18-month-old Hispanic boy presented with a 1-week history of emesis, decreased activity, tactile fevers, and depressed mental status after mild head trauma. Initial computed tomographic scan of the head was remarkable for a 6-cm-diameter, uniformly enhancing mass attached to the inferior wall of the right lateral ventricle with marked hydrocephalus, right greater than left. The patient had otherwise been well and had demonstrated normal early development, walking, and speech. He had no siblings, but his parents, both in their early 20s, did not know of any cancers in first- or second-degree relatives. Subtotal resection of the ventricular mass was performed, with a diagnosis of choroid plexus carcinoma based on the marked cytologic atypia, high mitotic index, and invasion into the ventricular wall. Abdominal ultrasound was performed after insertion of a ventriculoperitoneal shunt to relieve the hydrocephalus during induction chemotherapy, and a large left adrenal mass was detected. The 7.0 × 6.5 × 4.0-cm mass was diagnosed as adrenal cortical carcinoma by fine-needle aspiration biopsy. He subsequently underwent subtotal resection, due to periaortic infiltration that prevented a total resection. The child completed a course of chemotherapy (VP-16, ifosfamide, mesna, and carboplatin). A second resection of the ventricular tumor from a posterior approach resulted in removal of 95% of the tumor, including most of the area of attachment to the ventricular wall. A follow-up magnetic resonance imaging study 6 months after the surgeries showed no interval increase in the size of the brain and abdominal tumors, although it did show significant hydrocephalus on the left. Shunt revision was performed without complications at that time. The patient is continuing on an episodic chemotherapeutic regimen and has resumed walking at age 36 months.

Pathologic Findings

Histopathologic studies included staining with hematoxylin-eosin and frozen sections of tumor with osmium processing. Immunohistochemical staining of formalin-fixed, paraffin-embedded sections was done with an avidin-biotin peroxidase labeling procedure using primary antibodies against the following antigens: p53 and MIB-1 (Dako Corporation, Carpinteria, Calif.). Light and electron microscopic examination of the child’s 2 tumors demonstrated that they were clearly 2 primary tumors rather than metastatic spread of tumor. The cytologic smear of the ventricular tumor revealed irregular papillary architecture with a surface lining of cytologically malignant cells (Figure 1, A), surrounded by sheets of pleomorphic cells (Figure 1, B) with foci of necrosis and microcalcifications. Scattered mitotic figures, including atypical mitoses, were present. The immunohistochemical proliferation index (MIB-1) was 2% to 5% focally. Immunohistochemical analysis for p53 protein was performed...
Figure 1. The cytologic smear of the ventricular tumor displayed irregular papillary architecture with a surface lining of cytologically malignant cells (hematoxylin-eosin, original magnification ×60 [A]). The bulk of the tumor showed sheets of pleomorphic cells (hematoxylin-eosin, original magnification ×120 [B]) with frequent expression of nuclear positivity for p53 (arrows) (p53, original magnification ×120 [C]). Electron microscopic examination revealed multiple basal bodies, some with sprouting cilia in the brain tumor cell, indicating an epithelial origin, such as choroid plexus (arrow) (original magnification ×3200 [D]).

Figure 2. The brown-tan cut surface of the encapsulated adrenal tumor showed hemorrhagic and cystic areas.

The resected left adrenal mass weighed 98 g and measured 7.0 × 6.5 × 4.0 cm. The cut surface of the encapsulated tumor was brown-tan with hemorrhagic and cystic changes (Figure 2). Light microscopy showed sheets of polygonal cells with round or bizarre nuclei and abundant eosinophilic and microvacuolar cytoplasm (Figure 3, A). The tumor included some larger anaplastic cells with bizarre lobated nuclei and displayed multifocal necrosis and occasional microcalcifications. The MIB-1 immunohistochemical stain for the proliferation index showed localization in 5% of tumor cells. Expression of p53 protein varied from 1+ to 4+ in some of the tumor cell nuclei (Figure 3, B). Adjacent, residual, normal-appearing adrenal gland also displayed p53 positivity in the nuclei of adrenal cortical cells (Figure 3, C). No papillary architecture or epithelial element of cells was found in the adrenal tumor. Frozen sections of tumor with osmium processing indicated that the vacuolated cytoplasm of the tumor cells was positive for lipid content. Electron microscopic examination confirmed the diagnosis of adrenal cortical carcinoma by demonstration of tubular cristae of the mitochondria and droplets of cytoplasmatic fat (Figure 3, D).

Molecular examination of tissues was performed for some known p53 gene mutations. Specimens of DNA from both tumors were isolated by proteinase K and phenol DNA extraction techniques. Probes for 4 of the most commonly identified p53 mutations4 were tested against the tissue DNA samples (exons 5–8). None of these mutations was detected in DNA extracted from either tumor. Further gene sequencing studies may reveal the specific mutation in this case, but if the mutation is in a regulatory or receptor gene for p53, rather than in exons 5 through 8, determination of the mutation may be difficult.

COMMENT

The p53 gene is located on the short arm of the human chromosome 17, band 13 (17p13) and has 11 exons that span approximately 20 kb. This gene yields a 2.8-kb messenger RNA transcript and encodes for a 53-kd nuclear phosphoprotein (hence, p53) of a 393-amino-acid sequence protein that is expressed at low levels in the wild type.5 p53 protein is a multifunctional transcription factor involved in the control of cell cycle progression and in determining the survival of cells exposed to DNA-damaging agents. DNA damage induces a transient nuclear accumulation and transcriptional activation of the p53 protein, accompanied by transcriptional activation of other target genes that are responsible for the induction of cell cycle arrest or apoptosis.5 Ineffective p53 protein would therefore result in continuous cell cycling without apoptotic disposal of defective cells, producing a tumor.
Mutations in the p53 gene are the most commonly reported genetic abnormality in human cancers. p53 mutant proteins differ in the extent to which they have lost suppressor function and in their capacity to inhibit wild-type p53 present in a dominant-negative manner. Mutant p53 protein has a relatively long half-life, compared to the wild type, and thus can be more readily detected by immunohistochemistry. Accumulation of altered p53 protein appears to be a common step in the development of many human cancers and has been reported frequently in cancers of the breast, colon, and lung.

The majority of p53 mutations are located in the DNA-binding domain of the protein; however, a recent study of a family suffering from LFS revealed a p53 mutation in exon 4, outside the core domain, which was identified as necessary for mdm2 (an oncogene that normally down-regulates p53 activity)-mediated p53 degradation. It was demonstrated that a mutation in this region is associated not only with resistance of the mutant p53 to mdm2-mediated degradation, but also with an impaired response of mutant protein to DNA damage. In addition, the p53(LFS) mutant was found to be defective in its transactivation function, which correlated with its inability to suppress cell growth and to induce apoptosis. The molecular basis for p53(LFS) functional impairment appeared to be its predominantly cytoplasmic localization caused by a faulty nuclear import mechanism, which, at least in part, resulted from the mutant protein’s decreased affinity for the importing of chaperon molecules.

There has been no consensus, however, about the reported prevalence of altered p53 protein expression in the relatively rare tumors seen in this case, that is, choroid plexus carcinomas and adrenocortical carcinoma. Some investigators have suggested that the abnormalities of p53 expression may be a late event in the development of adrenocortical tumors. One report described a pediatric patient with adrenocortical carcinoma at 19 months, with later discovery of a cerebral primitive neuroectodermal tumor at age 5 years. This case harbored a germline mutation in exon 7 of p53 in codon 248. The present case has a similar young age of presentation for the adrenal malignant tumor.

In the case we are reporting, concurrent primary tumors were manifest at 18 months of age in a child with no known family history of malignancies. Both tumors, choroid plexus carcinoma in the brain and adrenocortical carcinoma, expressed variable immunohistochemically detectable levels of p53 in cell nuclei of the tumors and in residual, histologically normal adrenal cortical cell nuclei.

Our findings are highly suggestive of a de novo p53 germline mutation, although the initial molecular biological studies to match the mutation with 4 known mutation sequence probes were negative. We suggest that immunohistochemical determination of p53 excess in simultaneous or subsequent malignant tumors may be of diagnostic importance in both children and adults, since the classic Li-Fraumeni presentation may extend into middle age. Consideration of this possibility is important because some p53-expressing lesions result in malignancies that are resistant to the most widely used therapeutic regimens and demand more aggressive treatment. Early detection of this parameter is potentially very important in determining effective patient care.

References