**Table S1: Clinical characteristics**
XRT indicates local field radiation therapy to the brainstem; HDC indicates high-dose chemotherapy with autologous stem cell transplantation; WHO for World Health Organization, IHC for immunohistochemistry. NA for not analyzed. *These cases exhibited tumor diagnosed as WHO grade III in the pons, and WHO grade IV in the lateral ventricle subventricular lesion. Note that the clinical characteristics of cases VU-DIPG I through V were previously reported [2] and the clinical characteristics and neuroimaging for cases SU-DIPG-III and SU-DIPG-VII were previously reported [3].

**Figure S1: Neural precursor cell immunophenotype of DIPG** Confocal photomicrograph illustrating DIPG cells that express the neural stem cell markers vimentin (red) and GFAPδ (green). Nuclei are marked with DAPI (blue). Yellow cells are those expressing both vimentin and GFAPδ. (Scale bar = 50 μm).

**Supplementary methods**
The autopsy protocols, previously described in detail [4,2], were approved by the institutional medical ethical committees of Stanford and VU University Medical Centers. Patients were included if they had classic DIPG MRI findings and clinical presentation. Informed consent was obtained. Brain autopsy and neuropathological review was conducted by board-certified pathologists expert in neuropathology (HV at Stanford, PW at VU). Following standard gross examination, coronal sections were made of cerebral hemispheres, while the brainstem and cerebellum were cut perpendicular to the long axis of the brainstem, and routine blocks were taken from medulla, pons, midbrain, cerebellum, thalamus, lateral ventricles, hippocampus and frontal cortex; areas exhibiting abnormalities on radiological or gross examination were also sampled.
Hematoxylin and eosin staining and immunohistochemistry were carried out according to standard procedures on paraffin embedded sections (5 μm thickness). The following antibodies were used: GFAP (1:300, Dako), Olig-2 (1:500, Millipore), Nestin (1:250, Millipore), Ki67 (1:200, Dako), p53 (1:400, Ventana) and Vimentin (1:2000, clone V9 manufactured at the Department of Pathology, VU University, Amsterdam). Antigen retrieval was performed by microwave heating in Tris/ EDTA pH9, or in the case of GFAP and Nestin by microwave heating in citrate buffer, or in the case of Ki67 using ER2 antigen retrieval (Leica), or in the case of P53 using Ventana's proprietary antigen retrieval solution, pH 8.5, with incubation time of 24 minutes. All antibodies were incubated for one hour at room temperature except as noted above. Procedure and antibody information for GFAPδ and Vimentin immunofluorescence have been previously reported [1].

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