1. Introduction

Isocitrate dehydrogenase 1 (IDH1) and isocitrate dehydrogenase 2 (IDH2), which convert isocitrate to α-ketoglutarate while reducing nicotinamide adenine dinucleotide phosphate (NADP+ to NADPH), were identified as mutated in a large percentage of progressive gliomas. These mutations occur at IDH1R132 or the homologous IDH2R172. Melanomas share some genetic features with IDH1/2-mutated gliomas, such as frequent TP53 mutation. We sought to test whether melanoma is associated with IDH1/2 mutations. Seventy-eight human melanoma samples were analyzed for IDH1R132 and IDH2R172 mutation status. A somatic, heterozygous IDH1 c.C394T (p.R132C) mutation was identified in one melanoma metastasis to the lung. Having identified this mutation in one metastasis, we sought to test the hypothesis that certain selective pressures in the brain environment may specifically favor the cell growth or survival of tumor cells with mutations in IDH1/2, regardless of primary tumor site. To address this, we analyzed IDH1R132 and IDH2R172 mutation status 53 metastatic brain tumors, including nine melanoma metastases. Results revealed no mutations in any samples. This lack of mutations would suggest that mutations in IDH1/2 may be necessary for the formation of tumors in a cell-lineage dependent manner, with a particularly strong selective pressure for mutations in progressive gliomas; this also suggests the lack of a particular selective pressure for growth in brain tissue in general. Studies on the cell-lineages of tumors with IDH1/2 mutations may help clarify the role of these mutations in the development of brain tumors.

© 2010 Elsevier Inc. All rights reserved.
mononuclear cells from 78 patients enrolled in Institutional Review Board-approved clinical trials at the Surgery Branch of the National Cancer Institute at the United States National Institutes of Health. Pathology-confirmed melanoma cell lines were derived from mechanically or enzymatically dispersed tumor cells, which were then cultured for 5–15 passages. IDH1/2 gene mutation status was subsequently analyzed in a panel of 53 tumor resections from metastases to the brain. Genomic DNA was isolated from frozen tumor tissue samples obtained from the Tissue Bank at the Preston Robert Tisch Brain Tumor Center at Duke University. In both cases, genomic DNA was isolated using the DNeasy Blood and Tissue kit (Qiagen, Valencia, CA, USA). PCR and sequencing primers were designed using Primer 3 (http://www-genome.wi.mit.edu/cgi-bin/primer/primer3_www.cgi) and synthesized by Invitrogen (Carlsbad, CA, USA). PCR and sequencing primers were designed to amplify the region surrounding the commonly mutated codons in both IDH1 and IDH2 exons. PCR products were 300–500 bp in length. PCR primers were designed to amplify the region surrounding the commonly mutated codons in both IDH1 and IDH2. In one melanoma sample, all coding exons of TP53, CDKN2A, and CDKN2B, as well as hotspot exons of BRAF and NRas, were sequenced using these methods.

3. Results and discussion

Of 78 human melanoma samples analyzed for IDH1R132 and IDH2R172 mutation status, a somatic, heterozygous IDH1 c.C394T (p.R132C) mutation was identified in tissue derived from a melanoma lung metastasis from a 53 year old female (Fig. 1). No IDH2R172 mutations were detected in any of these samples. In the IDH1–mutated sample, we sequenced regions of BRAF, NRas, TP53, and CDKN2A/CDKN2B to identify any changes in genes that are commonly altered in melanoma. We found a BRAF c.T1799A (p.V600E) mutation, while NRas, TP53, and CDKN2A/CDKN2B were unaltered in this sample.

IDH1/2 mutations are frequent in progressive gliomas, but are very rare in other cancers besides acute myelogenous leukemia. To date, mutations in IDH1 have been identified only in one B-cell acute lymphoblastic leukemia [8], two prostate cancers [8], and one colorectal cancer [9]. Noting the presence of mutations in this small fraction of other cancers, we postulated two hypotheses. In the first hypothesis, mutations in IDH1/2 may be cell-lineage dependent, and thus only occur in the cell-of-origin for a specific subset of tumors, including progressive gliomas. In the second hypothesis, a tissue-dependent selective pressure for growth in brain tissue may select for frequent mutations in IDH1/2. If the second hypothesis is correct, we should be able to identify the presence of relatively frequent mutations in IDH1/2 in non-primary CNS metastases to the brain. Therefore, we analyzed IDH1R132 and IDH2R172 mutation status in a set of nine melanoma metastases to the brain and an additional panel of 44 metastases of other tumor types to the brain, including lung, breast, renal, uterine, ovarian, esophageal, and urothelial cancer metastases. Results revealed no mutations regardless of tumor type, consistent with a previous small-scale study looking at colon cancer metastases to the brain [10].

Here, we present the first identification of a mutation in IDH1 or IDH2 in melanoma. These results have important clinical implications regarding the role of mutations in IDH1/2 in the development of tumors. Mutations in IDH1 have now been identified in gliomas, leukemias, prostate cancer, colorectal cancers, and melanomas, supporting the possibility of a commonly altered pathway that may prove advantageous to the formation of tumors in all these cell types. The lack of mutations in a panel of non-primary CNS metastases to the brain would suggest that mutations in IDH1 or IDH2 are not necessarily required specifically for growth in brain tissue. The low frequency of mutations in IDH1R132 and IDH2R172 in primary glioblastomas, which grow extremely aggressively in brain tissue, further supports this. This leads to the conclusion that mutations in these genes may be necessary for formation of tumors in a cell-lineage dependent manner, with a particularly strong selective pressure for mutations in progressive gliomas and acute myelogenous leukemias. Studies focusing on the cell-lineages from which tumors with IDH1 or IDH2 mutations develop may help to elucidate the role of these mutations in cancer pathogenesis.

Acknowledgments

We thank Kristin E. Yates for technical assistance. This work is supported by the Intramural Research Programs of the National Human Genome Research Institute and the National Cancer Institute, National Institutes of Health, USA, and by NIH grant R01CA118822.

References


