New Insights into von Hippel-Lindau Function Highlighted by Investigation of the Trichloroethylene-Induced p.P81S Hotspot Mutation

Len Neckers, Christopher J. Ricketts, W. Marston Linehan

Correspondence to: W. Marston Linehan, MD, Urologic Oncology Branch, Center for Cancer Research, National Cancer Institute, CRC Room 1W-5940, 9000 Rockville Pike, Bethesda, MD 20892–1107 (e-mail: WMLinehan@nih.gov).

The von Hippel-Lindau gene (VHL), composed of three exons, encodes a protein, pVHL, with diverse tumor suppressor activities. Germline VHL mutations are found in patients with von Hippel-Lindau (VHL), an autosomal dominant hereditary cancer syndrome in which affected individuals are at risk for the development of highly vascularized benign and malignant tumors including central nervous system hemangioblastomas, retinal hemangiomas, clear cell renal carcinomas (ccRCCs), pheochromocytomas, and pancreatic neuroendocrine tumors (1). Biallelic inactivation of VHL, whether via mutation or promoter methylation, is found in VHL-associated tumors and in nearly 90% of sporadic ccRCC tumors (2,3), the most common form of sporadic kidney cancer.

Among its activities, pVHL functions as the substrate recognition module of a multiprotein ubiquitin ligase complex, and, by recruiting hypoxia-inducible factor (HIF) 1α and 2α to this complex, mediates their oxygen-dependent degradation (4). VHL mutations that interfere with HIF-α recognition promote aerobic glycolysis and support neoangiogenesis, perhaps the most intensively studied consequences of pVHL loss. However, regulation of HIF-α expression is unlikely to be the sole mechanism underlying the tumor suppressor function of pVHL. Given its role as the substrate recognition module of a multiprotein complex, it is reasonable to assume that pVHL has additional activities that depend on protein–protein interactions unrelated to (or perhaps complementary with) HIF stabilization, and that deregulation of one or more of these activities contributes to tumor formation, likely in a tissue-specific manner. Indeed, pVHL has been implicated in a number of cellular processes that appear to be HIF independent, including cell cycle regulation, extracellular matrix assembly, and cytoskeleton stability (5).

The development of ccRCC is associated with both genetic (eg, VHL, germline mutation) and environmental factors, particularly long-term exposure to the industrial solvent trichloroethylene (TCE). Although TCE exposure does not affect the histopathology of sporadic ccRCC, its onset is earlier in TCE-exposed individuals, and the VHL gene in a substantial percentage of these tumors contains a specific mutation of a cytosine to thymidine at nucleotide 454, resulting in the substitution of serine for proline at amino acid 81 (p.P81S) (6). This hotspot mutation is found in nearly 40% of sporadic ccRCC tumors from TCE-exposed individuals, but it was not observed in sporadic tumors from unexposed individuals, is rarely found in patients with VHL disease, and was not present in the more than 400 tumors sequenced for The Cancer Genome Atlas ccRCC project (7). Interestingly, the VHL p.P81S mutation is associated with low penetrance and mild phenotype when identified in VHL families, and in cases of TCE-associated tumors this mutation has occasionally been detected in the apparently normal kidney parenchyma adjacent to the tumor, consistent with protumorigenic but not directly transforming activity (6,8). Although genotype–phenotype correlations exist in cancers with germline VHL mutations, the contribution of the TCE–associated P81S mutation to the development of sporadic ccRCC has not been extensively explored.

In the current issue of the Journal, DeSimone and colleagues use embryonic stem cells derived from Vhl−/− mice to assess the phenotypic consequences of reexpression of either the p.P81S mutation or a second hotspot mutation, p.R167Q, compared with reexpression of wild-type pVHL (9). These investigators report that although VHL p.P81S mutation does not provide a growth advantage to embryonic stem cells in vitro, teratomas derived from these cells after injection into nude mice were much larger than those derived from embryonic stem cells expressing either p.R167Q mutation or wild-type pVHL. The authors demonstrate that the cells expressing p.P81S VHL displayed much lower apoptotic markers in comparison with other embryonic stem cells and were much less likely to initiate apoptosis in response to radiation-induced, ataxia telangiectasia mutated–mediated DNA damage. Thus DeSimone and colleagues suggest that the canonically milder VHL p.P81S mutation provides a more subtle pro-tumor benefit that might normally be overshadowed by the effects of more penetrant VHL mutations on glycolysis and tumor angiogenesis. Additionally, this could explain the mutation’s milder phenotype in VHL syndrome compared with other VHL mutations. Although not directly transforming, the unique phenotypic consequences of the VHL p.P81S mutation would certainly promote tumor growth and potentially provide resistance to therapeutic radiation. This may be particularly relevant in TCE-associated tumors where it would be expected that the DNA damage would cause multiple additional mutations that could drive carcinogenesis and work cooperatively with other mutation events. Indeed, it has been reported that several patients exposed to high levels of TCE harbored three or four mutations in the VHL gene alone (6,7).

This report adds to previous observations that the wide variety of VHL mutations can have varied effects on the known functions of pVHL in addition to effects on HIF1α and HIF2α. The VHL
p.P81S mutation presents with a milder degree of HIF 1/2 stabilization and activation of HIF target genes, providing a degree of pseudohypoxia that can still be modulated by actual hypoxia in the surrounding environment. The report also demonstrates that the VHL p.P81S and p.R167Q mutations result in the loss of interaction with Elongin C. A recent publication highlighted the mutation of this protein’s gene (TCEB1) in a small number of ccRCCs that notably lack VHL gene mutations, suggesting these could be seen as equivalent events (3).

Interestingly, another recent report identified proline 81 as part of a binding motif (PXVXL) for a novel VHL binding partner, heterochromatin protein 1, whose association is disrupted by the p.P81S mutation (10). This new association recruits VHL to chromatin where it could affect gene expression via ubiquitination of chromatin-associated proteins, or perhaps by serving as an adaptor between heterochromatin protein 1 and other proteins. This is particularly relevant considering the recent discovery of the interaction of VHL–HP1 interaction and potential chromatin remodeling effects could provide important mechanistic insight into the specific tumor-promoting activity of TCE–associated VHL p.P81S mutation while also identifying potential therapeutic approaches for patients affected by this disease.

References

Funding
This work was supported by the Intramural Research Program of the NIH, National Cancer Institute, Center for Cancer Research.

Notes
The authors have no conflicts of interest to disclose. The study sponsor had no role in the writing of this editorial or the decision to submit the editorial for publication.

Affiliations of authors: Urologic Oncology Branch, Center for Cancer Research, National Cancer Institute, Bethesda, MD (LN, CJR, WML).

Inflammatory Breast Cancer: Yet Another Risk of the Obesity Epidemic?
Laleh Amiri-Kordestani, Farin Kamangar, Jo Anne Zujewski

Correspondence to: Jo Anne Zujewski, MD, National Cancer Institute, 9609 Medical Center Dr, Rockville, MD 20850 (e-mail: zujewski@mail.nih.gov).

Two of the most perplexing problems in breast cancer, inflammatory breast cancer (IBC) and obesity, are linked in the report by Schairer and colleagues in this issue of the Journal (1). In particular, these results show a striking fourfold increased risk of IBC in obese women compared with normal weight individuals (body mass index [BMI] < 25 kg/m²).

IBC constitutes approximately 1–6% of all breast cancers (2,3), which makes studies of its risk factors somewhat difficult. IBC has an aggressive clinical course, with a 5-year disease-free survival of only 34% (4). Although IBC has been recognized as a separate entity (5), we know surprisingly little about both its etiology and pathophysiology.