Wilms tumor 1 gene, CD97, and the emerging biogenetic profile of glioblastoma

Aravind Somasundaram, B.S.,1 Nathan Ardanowski, B.S.,1 Charles F. Opalak, M.P.H.,1 Helen L. Fillmore, Ph.D.,2 Archana Chidambaram, M.D., Ph.D.,3 and William C. Broaddus, M.D., Ph.D.1

1Department of Neurosurgery, Virginia Commonwealth University, Richmond, Virginia; 2Department of Hematology-Oncology, Roswell Park Cancer Institute, Buffalo, New York; and 3School of Pharmacy and Biomedical Sciences, University of Portsmouth, Portsmouth, United Kingdom

Glioblastoma multiforme (GBM) is the most common type of primary brain tumor, and current treatment regimens are only marginally effective. One of the most vexing and malignant aspects of GBM is its pervasive infiltration into surrounding brain tissue. This review describes the role of the Wilms tumor 1 gene (WT1) and its relationship to GBM. WT1 has several alternative splicing products, one of which, the KTS+ variant, has been demonstrated to be involved in the transcriptional activation of a variety of oncogenes as well as the inhibition of tumor suppressor genes. Further, this paper will examine the relationship of WT1 with CD97, a gene that codes for an epidermal growth factor receptor family member, an adhesion G-protein–coupled receptor, thought to promote tumor invasiveness and migration. The authors suggest that further research into WT1 and CD97 will allow clinicians to begin to deal more effectively with the infiltrative behavior displayed by GBM and design new therapies that target this deadly disease.

(key wordS • glioblastoma • Wilms tumor 1 gene • CD97

Abbreviations used in this paper: DAF = decay accelerating factor; EGF-TM7 = epidermal growth factor–7 transmembrane; GBM = glioblastoma multiforme; shRNA = short hairpin RNA; WTAP = Wilms tumor 1–associated protein (pre-mRNA-splicing regulator WTAP); WT1 = Wilms tumor 1.
Wilms Tumor 1 Gene

Though WT1 is absent in normal neural tissue, it has been described in cultured neoplastic glial cell lines, and further work has indicated high WT1 expression in acute myeloid leukemia and acute lymphoid leukemia.7,13 WT1 encodes for a zinc finger family transcription factor located at 11p13 whose physiological expression is associated with normal genitourinary embryologic development.1 It has been hypothesized that WT1 may exert its oncological effects in a multifaceted modality via an alternative mRNA splicing variant that involves the insertion of the 3–amino acid sequence of lysine, threonine, serine (KTS) into the active zinc finger region.28 This is in contrast to the KTS− variant, which transcriptionally regulates suppression of at least 7 putative tumor suppressor genes and increases expression of 11 reputed oncogenes.3 The KTS+ product, on the other hand, has only a limited role in direct transcriptional regulation; instead it participates in putative splicing factor interactions and has an association with olfactory neuron development.14

GBM and Wilms Tumor

When WT1 expression was first reported in brain tumors, it was hypothesized that testing for WT1 expression might not have clinical significance.21 The first laboratory investigation that focused on WT1 expression in central nervous system malignancies, including GBM, found WT1 expression in most of these tumors; yet there were no mutations affecting the zinc fingers of the gene product in tumors expressing WT1.9 Though this appeared to indicate that WT1 expression might not have clinical relevance to the molecular etiology of brain tumors, these results nonetheless ignited a novel interest in the relationship of WT1 to central nervous system tumors. A few years later, Oji et al. were able to show that the WT1 protein expression was significantly greater in high-grade tumors than in low-grade tumors and that treatment with WT1 antisense oligomers specifically inhibited GBM cell lines.24 This study, coupled with later confirmation from immunohistochemical tests that demonstrated WT1 expression in GBM cells, highlighted its potential as a target for immunotherapy.12,22 This differential expression of WT1 in high-grade brain tumors has recently been validated in an in vivo study analyzing human gliomas, which included 442 glioblastomas, 303 astrocytomias, 41 oligodendrogliomas, and 43 oligoastrocytomias.23 The results showed that WT1 expression in brain tumors increased with WHO grade, older age, and absence of IDHI mutation.22

The introduction of a WT1-based vaccine in clinical trials was first described by Oka et al. in 2004.13,23 The authors reported the effects of the vaccine on breast cancer, lung cancer, and leukemia in 26 patients, reporting tumor regression and no toxicity as a result of the drug. The same group recently published data on a Phase II clinical trial that included 21 patients who received WT1 peptide vaccination for recurrent GBM.15 Preliminary data showed that patients with WT1/HLA-A*2402 positivity who received WT1 immunotherapy showed a favorable outcome when compared with patients treated with a standard chemotherapy protocol.15 In a follow-up study analyzing the same GBM patient population, the authors reported that WT1 expression can be used as a prognostic marker in determining progression-free survival after immunotherapy and that patients with intermediate WT1 expression levels may have the best outcomes.7

One possible mechanism of action of a WT1-based vaccination could lie in its suppression of WT1’s inactivation of tumor suppressor gene p53 (TP53). This hypothesis was initially tested in p53-null Saos-2 osteosarcoma cells, and it was revealed that WT1 inhibits p53-mediated apoptosis, which is normally induced by chemotherapy, radiation, and overexpression of wild-type p53.19,20 Clark et al. continued to investigate this relationship between WT1 and p53 in GBM by examining the effect of WT1 expression and WT1 silencing on p53-mediated cell death and response to radiotherapy.28 Results showed that WT1 silencing led to GBM cells becoming susceptible to radiation-induced death, suggesting a potential target to improve responses to radiotherapy. The relationship of WT1 silencing and chemotherapy was tested in a separate study by Chen et al. that revealed similar results; WT1 silencing resulted in increased chemotherapeutic response.3 It was reported in follow-up work that there was an interaction between the KTS+ isoform of WT1 and wild-type p53, revealing that the potential oncogenic function of WT1 in GBM may depend on this splicing variant.7

Additional avenues whereby WT1 promotes tumorogenesis was found in WT1’s relation with Wilms tumor 1–associated protein (WTAP, also known as pre-mRNA-splicing regulator WTAP). WTAP is a nuclear protein that was isolated through a yeast 2-hybrid screen and has been reported to be overexpressed in GBM cell lines.16,19 Jin et al. suggested that WTAP may regulate migration and invasion of GBM cells by controlling epidermal growth factor signaling.16 The link between WT1 and cancer cell invasion was initially reported by Jomgeow et al. in TYK ovarian cancer cell lines, where in vitro experiments demonstrated that WT1 overexpression led to an increase in cell invasion.17 Kijima et al. demonstrated that GBM cell lines with downregulated WT1 expression had decreased tumorigenicity in an intracranial in vivo assay.15 Further, they suggested that WT1 may play a role in apoptosis as cell lines transfected with anti-WT1 shRNA showed upregulation of apoptosis-related genes when compared with control cell lines—supporting a previous report that WT1 may be involved in apoptosis in GBM.34 These preliminary studies suggest that the oncogenic role WT1 may play in GBM cell proliferation, invasiveness, and survival involves suppressed apoptosis.

CD97

Recent research at our institution has focused on another molecule that appears to be involved in promoting the invasiveness of GBM through a relationship with WT1, as an apparent target of WT1 transcriptional activation.3 The CD97 molecule, also known as TM7LNI, is a member of the epidermal growth factor–7 transmembrane (EGF-TM7) cell receptor subfamily that mediates cell-cell interactions. CD97 is a part of a 6-mem-
Wilms tumor 1 gene and GBM

The expression of CD97 has been reported in various tumors, including thyroid carcinomas, pancreatic cancers, colon cancers, and oral squamous cell carcinomas. This gene is involved in cell-to-cell adhesion, migration, and signaling, and its expression has been correlated with the prognosis of patients with GBM. A recent study by Chidambaram et al. demonstrated that down-regulation of Wilms' tumor 1 (WT1) in GBM cell lines was associated with a shorter survival time compared to WT1-overexpressing cells.

**References**


**Disclosure**

The authors report no conflict of interest concerning the materials or methods used in this study or the findings specified in this paper.

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A. Somasundaram et al.

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