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[Cancer Epidemiol Biomarkers Prev.](#) 2010 Jun;19(6):1409-22.

### Identification of potential serum biomarkers of glioblastoma: serum osteopontin levels correlate with poor prognosis.

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#### Abstract

**BACKGROUND:** The aim of this study is to identify serum biomarkers with classification and prognosis utility for astrocytoma, in particular glioblastoma (GBM). **METHODS:** Our previous glioma microarray database was mined to identify genes that encode secreted or membrane-localized proteins. Subsequent analysis was done using significant analysis of microarrays, followed by reverse transcription-quantitative PCR (RT-qPCR) and immunohistochemical validation in tumor tissues, ELISA and Western blot validation in sera, and correlation with survival of GBM patients. **RESULTS:** Significant analysis of microarrays identified 31 upregulated and 3 downregulated genes specifically in GBMs. RT-qPCR validation on an independent set of samples confirmed the GBM-specific differential expression of several genes, including three upregulated (CALU, CXCL9, and TIMP1) and two downregulated (GPX3 and TIMP3) novel genes. With respect to osteopontin (OPN), we show the GBM-specific upregulation by RT-qPCR and immunohistochemical staining of tumor tissues. Elevated serum OPN levels in GBM patients were also shown by ELISA and Western blot. GBM patients with high serum OPN levels had poorer survival than those with low serum OPN levels (median survival 9 versus 22 months respectively;  $P = 0.0001$ ). Further, we also show high serum TIMP1 levels in GBM patients compared with grade II/III patients by ELISA and downregulation of serum GPX3 and TIMP3 proteins in GBMs compared with normal control by Western blot analysis. **CONCLUSIONS:** Several novel potential serum biomarkers of GBM are identified and validated. High serum OPN level is found as a poor prognostic indicator in GBMs. **IMPACT:** Identified serum biomarkers may have potential utility in astrocytoma classification and GBM prognosis. Copyright 2010 AACR.

PMID: 20530493 [PubMed - in process]

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