Risk and Prognostic Factors for Malignant Glioma

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Akademisk avhandling

som med vederbörligt tillstånd av Rektor vid Umeå universitet för avläggande av medicine doktorsexamen framläggs till offentligt förvar i sal E04, byggnad 6E,
Fredagen den 21 december, kl. 09:00.
Avhandlingen kommer att förvaras på engelska.

Fakultetsopponent: Professor, Monika Hegi,
Centre Hospitalier Universitaire Vaudois CHUV, Schweiz.
Glioblastoma is the most common and aggressive type of glioma and associated with poor prognosis. Apart from ionizing radiation and some rare genetic disorders, few aetiological factors have been identified for primary brain tumours. Inverse associations to asthma and low IgG levels for varicella zoster virus have in previous studies indicated that the immune system may play a role in glioma development. Little is known about prognostic factors in glioma. Previous studies have shown an association between age, Karnofsky performance status, O6-methylguanine-DNA methyltransferase (MGMT) hypermethylation, and prognosis. Polymorphisms in different low penetrance genes have in some studies been associated with glioma prognosis.

**Material and methods:** In paper I, we analysed IgG levels for four different viruses, Epstein-Barr virus (EBV), cytomegalovirus (CMV), varicella zoster virus (VZV) and adenovirus (Ad), in prediagnostic blood samples from 197 cases with glioma and 394 controls collected from three large cohorts: the Northern Sweden Health and Disease Study; the Malmö Diet and Cancer Study; and the Diet, Cancer and Health cohort from Copenhagen. ELISA was used to measure IgG levels and for EBV response to both the nuclear antigen (EBNA1) and the viral capsid antigen (VCA) was measured, for VCA using immunofluorescence. IgG levels were divided into quartiles and binary logistic regression was used to compare the quartiles in cases and controls. All odds ratios were adjusted for age, sex, and cohort. In paper II-IV, we studied 176 glioblastoma cases from Sweden and Denmark. We collected treatment and follow-up data on the cases. We genotyped 30 tagging SNPs in EGF, 89 in EGFR, 27 in VEGFR2, and 17 in VEGF. We also studied 1458 SNPs in 136 DNA repair genes. Hazard ratios were calculated using Cox regression; the major allele was set as categorical variable and all HR were adjusted for age, sex, country, and treatment. For the DNA repair gene results, we adjusted the p-values for multiple testing. Significant findings were confirmed in separate datasets.

**Results and Discussion:** We found a trend towards higher IgG VZV levels in controls compared to glioma cases, especially when restricting the analyses to only include glioma cases with at least 2 years between blood sample and diagnosis. This finding might indicate that there is an aetiological and not a disease-related association. This confirms previous findings and support that a strong immune system can detect and inhibit growth of small cancer clusters. In EGF, we found seven SNPs in one haplotype block that were significantly associated with glioblastoma survival. Four of the SNPs were available for confirmation; however, none reached statistical significance. One explanation could be age differences in the different cohorts. In EGFR, four SNPs associated with survival were found; however, as 89 polymorphisms were tested this was the expected outcome by chance. In VEGF and VEGFR2, we found two SNPs associated with glioblastoma survival, but they could not be confirmed in the separate dataset, and due to multiple testing, were considered to be false positives. Among the DNA repair genes, we found nine SNPs in three genes-MSH2, RAD51L1 and RECQL4-associated with glioblastoma survival after confirmation and adjustment for age, sex, country, and treatment. After adjusting for multiple testing, one SNP in MSH2 and one in RECQL4 remained significant.

**Conclusions:** Our studies provide additional knowledge to the aetiological and prognostic factors important for glioma, emphasising the possible importance of immune function mechanisms. We found limited evidence for the role of genetic variants in glioma progression genes, and some for DNA repair variants as prognostic factors for glioblastoma survival.

**Keywords**
Glioma, Glioblastoma, Risk, Outcome, EGR, EGFR, VEGF, VEGFR, DNA repair, virus