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SWI/SNF gene variants and glioma risk and outcome

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Abstract

BACKGROUND—The human SWItch/Sucrose Non-Fermentable (SWI/SNF) chromatin remodeling complex plays essential roles in a variety of cellular processes and has been implicated in human cancer. However, the role of germline genetic variants in this complex in relation to cancer risk is not well studied.

METHODS—We assessed the association of 16 variants in the catalytic subunits (*SMARCA2* and *SMARCA4*) of the SWI/SNF complex with the risk of glioma subtypes (lower grade astrocytoma, oligodendroglioma and glioblastoma [GBM]) and with mortality from high-grade tumors (GBM) in a multicenter US case-control study that included 561 cases and 574 controls. Associations were estimated with odds ratios (OR, for risk) or hazards ratios (HR, for mortality) with 95% confidence intervals (CI). False discovery rate (FDR-q) was used to control for multiple testing in risk associations.

RESULTS—None of the investigated SNPs was associated with overall glioma risk. However, analyses according to histological subtypes revealed a statistically significant increased risk of oligodendroglioma in association with *SMARCA2* rs2296212 (OR=4.05, 95%CI=1.11-14.80, P=0.030, q=0.08) and rs4741651 (OR=4.68, 95%CI=1.43-15.30, P=0.011, q=0.08) and *SMARCA4* rs11672232 (OR=1.90, 95%CI=1.01-3.58, P=0.048, q=0.08) and rs12232780

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(OR=2.14, 95%CI=1.06-4.33, P=0.035, q=0.08). No significant risk associations were observed for GBM or lower grade astrocytoma. Suggestive associations with GBM mortality were not validated in the Cancer Genome Atlas.

CONCLUSION—Our findings suggest that genetic variants in *SMARCA2* and *SMARCA4* influence the risk of oligodendroglioma. Further research is warranted on the SWI/SNF complex genes and epigenetic mechanisms more generally in the development of glioma in adults.

Keywords

SNP; *SMARCA2*; *SMARCA4*; chromatin remodeling; glioma; SWI/SNF

Introduction

Glioma is the most common central nervous system tumor comprising approximately 80% of malignant brain tumors. Despite improvements in diagnosis and treatment, the prognosis of glioma remains dismal with a 5-year survival rate of less than 30% [1]. There is a paucity of information on the etiology of glioma. Familial aggregation [2] and the identification of common [3, 4] and rare [5] genetic susceptibility variants suggest a genetic predisposition to this disease. However, the currently known genetic variants may collectively account for a small proportion of cases and more variants remain to be identified.

Chromatin structure plays an important role in the regulation of gene expression and perturbation in chromatin structure is important in cancer development [6]. The human SWItch/Sucrose Non-Fermentable (SWI/SNF) complex modulates the structure of chromatin and thus plays an important role in cancer development [6]. Somatic mutations in subunits of the complex including, BRM/*SMARCA2* and BRG1/*SMARCA4*, have been observed in several human cancers including brain tumors [6, 7]. BRG1 expression is elevated in human glioma tumor tissue samples compared to normal brain tissue [8]. Furthermore, *SMARCA2* is expressed in proliferating neural stem cells and the conversion of rat oligodendrocyte precursor cells to multipotent neural stem-like cells is mediated, in part, by *SMARCA2* [9].

Because of the important role of *SMARCA2* and *SMARCA4* in neural development and cancer, we investigated for the first time whether genetic variants in these genes are associated with the risk of glioma subtypes and mortality in 561 cases and 574 controls in a clinic-based case-control study in the United States.

Materials and Methods

Study population

Details of the study protocols are reported elsewhere [10]. The study population comprised Caucasian participants (United States residents only) in a clinic-based case-control study in the Southeastern United States. The present analysis included participants enrolled between 2004 and 2010 and followed up till March 2012. Cases had histologically-confirmed primary glioma and were enrolled a median of one month following tumor diagnosis. Controls were friends, in-laws and other associates of the cases and persons from the same communities as the cases with no previous history of brain tumor. Controls were frequency matched to cases on age, gender and residence. Eighty-seven percent of eligible glioma cases participated in the study and 49.6% of confirmed eligible households yielded a control participant.

The study was approved by Investigational Review Committees at each participating center and all subjects provided written informed consent.

Biospecimen and data collection and Genotyping

DNA samples were self-collected by oral rinse or the saliva method using Oragene kits (www.dnagenotek.com) and extracted using standard protocols [10]. Information on demographic characteristics and risk factors for glioma were obtained via in-person interview.

SNPs in *SMARCA2* (N=8) and *SMARCA4* (N=9) were selected based on SNP information from unrelated Caucasian samples in Hapmap using a minor allele frequency (MAF)≥0.05. Genotyping was attempted on 599 glioma cases and 619 controls using the Taqman Open Array system under previously described conditions [10]. A total of 83 participants (38 cases [6.3%] and 45 controls [7.3%]) were excluded due to low (<80%) call rates, leaving 561 cases and 574 controls in the final analysis. All of the 17 SNPs in *SMARCA2/4* were successfully genotyped. The mean sample call rate was 98.6% and the mean concordance for 70 duplicate samples was 99.7%. One SNP in *SMARCA4* (rs1801514) was monomorphic and was therefore excluded from further analysis.

The Cancer Genome Atlas Data

We attempted to validate mortality associations using independent data from The Cancer Genome Atlas (TCGA). TCGA has genotype, demographic and clinical data on various cancer sites (including GBM) that are freely available and widely used by cancer researchers [11]. For comparability to our data, we limited analysis of the TCGA data to Caucasians aged 19-89 years. Seven of the SNPs were genotyped, and 8 other SNPs had suitable proxies (r^2=0.8) in TCGA, whereas no counterpart could be identified for one SNP (rs11672232).

Statistical analysis

Genotypes among participants were used to estimate allele frequencies and departure from Hardy-Weinberg equilibrium (HWE) was assessed among control subjects using Fisher's exact test. The association between each SNP and glioma risk (overall or histological subtypes) was estimated with odds ratios (OR) and 95% confidence intervals (CI) using unconditional logistic regression. Cox proportional hazards regression was used to estimate hazard ratios (HR) and 95% CIs for the association between SNPs and mortality among patients with GBM (the most prevalent histological subtype). A total of 262 GBM-related deaths occurred in a median of 13.3 months (range: 0.79 months-62.0 months) following diagnosis. The 43 surviving patients were followed a median of 30.5 months (range: 7.4 months-55.7 months). Survival time was defined as the time from GBM diagnosis to death or last contact among surviving cases.

Three inheritance genetic models (log-additive, dominant and recessive) were tested for each outcome and the model with the minimum *P*-value was considered as the best genetic model [12]. All regression models included terms for age and gender. Statistical analysis was performed using SAS Version 9.1 (SAS Institute, Inc., Cary, NC) and statistical significance was defined as a two-sided *P*-value <0.05. False discovery rate (FDR) q-values were calculated to adjust for multiple testing.

Results

Descriptive information on study populations is shown in Table 1. None of the SNPs was associated with overall glioma risk (Supplementary Table 1). Analyses according to histological subtypes revealed a statistically significant increased risk of oligodendroglioma

in association with *SMARCA2* rs2296212 (OR=4.05, 95%CI=1.11-14.80, P=0.030, q=0.08) and rs4741651 (OR=4.68, 95%CI=1.43-15.30, P=0.011, q=0.08) and *SMARCA4* rs11672232 (OR=1.90, 95%CI=1.01-3.58, P=0.048, q=0.08) and rs12232780 (OR=2.14, 95%CI=1.06-4.33, P=0.035, q=0.08) (Table 2). The two SNPs in *SMARCA2* were perfectly correlated (r^2=1.0), but no correlation was observed for the two *SMARCA4* SNPs. No significant risk associations were observed for GBM or lower grade astrocytoma (Table 2).

SMARCA2 rs13288443 (HR=0.36, 95%CI=0.13-0.99, P=0.049) was associated with increased GBM mortality in our data (Supplementary Table 2). This SNP was not genotyped in TCGA, but results for a highly correlated (r^2=0.85) SNP (*SMARCA2* rs12003289) did not validate our finding in the TCGA data (Supplementary Table 2).

Discussion

We report for the first time results from an association study of genetic variants in the SWI/SNF complex with glioma risk and mortality, and one of the first studies of this pathway in any cancer. Findings suggest that SNPs in the component genes, *SMARCA2* and *SMARCA4*, may be associated with an increased risk for oligodendroglioma, though not other glioma subtypes. These preliminary results are consistent with a potential role for chromatin remodeling in adult-onset oligodendroglioma, consistent with the recently demonstrated role for this process in the onset of glioma in children [13].

SMARCA2 and *SMARCA4* encode catalytic subunits, which are responsible for the DNA-dependent ATPase activity of the SWI/SNF complex. This activity is required to regulate the accessibility of transcription factors to their DNA targets for expression and therefore important in cancer development [6]. Contrary to findings from other cancers showing that the catalytic units act as tumor suppressors, a recent study showed that expression of *SMARCA4* was elevated in human glioma tumor tissue samples (both benign - Grades I/II and malignant - Grades III/IV) compared to normal brain tissue and down-regulation in glioma cell lines resulted in decreased cell proliferation, migration and invasion [8]. In our study, we observed an increased risk of oligodendroglioma with *SMARCA4* variants. Both variants are intronic and their function is not known, but if they have a functional impact that elevates expression then it will result in the observed increased glioma risk. However, *in silico* analysis of the variants using F-SNP (http://compbio.cs.queensu.ca/F-SNP/) showed no regulatory relevance.

Our finding for the *SMARCA2* variants is interesting in light of the role of this gene in oligodendrocyte maturation [9]. One of the examined variants (*SMARCA2* rs2296212) is nonsynonymous, resulting in a change of aspartate in protein position 1546 to a glutamate. *In silico* analysis showed no difference in the molecular phenotype for the polymorphism. However, the variant is located in an exonic splicing enhancer/silencer and can potentially affect the function of the gene. Additional research is warranted to elucidate the functional significance of these variants and the biological mechanism underlying the observed associations.

The present analysis is one of the first studies to examine genetic variants in the SWI/SNF complex in relation to cancer risk. Only two previous studies have examined this topic. One study reported a strong association between two *SMARCA2* promoter variants and lung cancer risk in smokers [14], while a second study reported no association between coding variants in *SMARCA2* and *SMARCA4* and breast cancer risk [15].

Our study benefits from a relatively large sample considering the rarity of glioma, rapid case ascertainment, and pathological confirmation of all cases. However, we did not validate the observed associations in an independent sample and there is a need for replication.

Furthermore, only the catalytic subunits of the SWI/SNF complex were investigated necessitating a more comprehensive analysis of these genes in future studies.

In summary, the findings of this study provide suggestive evidence that genetic variants in the chromatin remodeling machinery may be associated with an increased risk for oligodendroglioma. On the basis of these findings, further studies are warranted on the contribution of germline genetic variants in the SWI/SNF complex to glioma and other cancer sites.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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References

[1]. Bondy ML, Scheurer ME, Malmer B, Barnholtz-Sloan JS, Davis FG, Il'yasova D, et al. Brain tumor epidemiology: consensus from the Brain Tumor Epidemiology Consortium. Cancer. 2008; 113(7 Suppl):1953–1968. [PubMed: 18798534]

[2]. Scheurer ME, Etzel CJ, Liu M, Barnholtz-Sloan J, Wiklund F, Tavelin B, et al. Familial aggregation of glioma: a pooled analysis. Am J Epidemiol. 2010; 172(10):1099–1107. [PubMed: 20858744]

[3]. Shete S, Hosking FJ, Robertson LB, Dobbins SE, Sanson M, Malmer B, et al. Genome-wide association study identifies five susceptibility loci for glioma. Nat Genet. 2009; 41(8):899–904. [PubMed: 19578367]

[4]. Wrensch M, Jenkins RB, Chang JS, Yeh RF, Xiao Y, Decker PA, et al. Variants in the CDKN2B and RTEL1 regions are associated with high-grade glioma susceptibility. Nat Genet. 2009; 41(8): 905–908. [PubMed: 19578366]

[5]. Stacey SN, Sulem P, Jonasdottir A, Masson G, Gudmundsson J, Gudbjartsson DF, et al. A germline variant in the TP53 polyadenylation signal confers cancer susceptibility. Nat Genet. 2011; 43(11):1098–1103. [PubMed: 21946351]

[6]. Wilson BG, Roberts CW. SWI/SNF nucleosome remodellers and cancer. Nat Rev Cancer. 2011; 11(7):481–492. [PubMed: 21654818]

[7]. Jones DT, Jager N, Kool M, Zichner T, Hutter B, Sultan M, et al. Dissecting the genomic complexity underlying medulloblastoma. Nature. 2012; 488(7409):100–105. [PubMed: 22832583]

[8]. Bai J, Mei PJ, Liu H, Li C, Li W, Wu YP, et al. BRG1 expression is increased in human glioma and controls glioma cell proliferation, migration and invasion in vitro. J Cancer Res Clin Oncol. 2012; 138(6):991–998. [PubMed: 22362300]

[9]. Kondo T, Raff M. Chromatin remodeling and histone modification in the conversion of oligodendrocyte precursors to neural stem cells. Genes Dev. 2004; 18(23):2963–2972. [PubMed: 15574597]

[10]. Egan KM, Thompson RC, Nabors LB, Olson JJ, Brat DJ, Larocca RV, et al. Cancer susceptibility variants and the risk of adult glioma in a US case-control study. J Neurooncol. 2011

[11]. Cancer Genome Atlas Research Network. Comprehensive genomic characterization defines human glioblastoma genes and core pathways. Nature. 2008; 455(7216):1061–1068. [PubMed: 18772890]

[12]. Permuth-Wey J, Kim D, Tsai YY, Lin HY, Chen YA, Barnholtz-Sloan J, et al. LIN28B polymorphisms influence susceptibility to epithelial ovarian cancer. Cancer research. 2011; 71(11):3896–3903. [PubMed: 21482675]

[13]. Schwartzentruber J, Korshunov A, Liu XY, Jones DT, Pfaff E, Jacob K, et al. Driver mutations in histone H3.3 and chromatin remodelling genes in paediatric glioblastoma. Nature. 2012; 482(7384):226–231. [PubMed: 22286061]

[14]. Liu G, Gramling S, Munoz D, Cheng D, Azad AK, Mirshams M, et al. Two novel BRM insertion promoter sequence variants are associated with loss of BRM expression and lung cancer risk. Oncogene. 2011; 30(29):3295–3304. [PubMed: 21478907]

[15]. Haiman CA, Garcia RR, Hsu C, Xia L, Ha H, Sheng X, et al. Screening and association testing of common coding variation in steroid hormone receptor co-activator and co-repressor genes in relation to breast cancer risk: the Multiethnic Cohort. BMC Cancer. 2009; 9:43. [PubMed: 19183483]

[16]. Permuth-Wey J, Thompson RC, Burton Nabors L, Olson JJ, Browning JE, Madden MH, et al. A functional polymorphism in the pre-miR-146a gene is associated with risk and prognosis in adult glioma. J Neurooncol. 2011; 105(3):639–646. [PubMed: 21744077]

Table 1

Characteristics of study participants

	GliomaSE[a]		TCGA[b]
	Cases (N=561)	**Controls (N=574)**	**Cases (N=281)**
Age, median (range)	51 (18-88)	55 (19-87)	58 (19-89)
Gender, N (%)			
Male	352 (62.8)	327 (57.0)	178 (63.4)
Female	209 (37.3)	247 (43.0)	103 (36.6)
State of Residence, N (%)			
Tennessee	151 (26.9)	187 (32.6)	-
Florida	149 (26.6)	138 (24.0)	-
Alabama	94 (16.8)	78 (13.6)	-
Kentucky	69 (12.3)	75 (13.1)	-
Georgia	64 (11.4)	60 (10.5)	-
Other[c]	34 (6.1)	36 (6.3)	-
Histological Subtype[d], N (%)			
Glioblastoma	305 (54.4)	--	281 (100.0)
Lower Grade Astrocytoma	138 (24.6)	--	-
Oligodendroglioma	83 (14.8)	--	-
Other gliomas	35 (6.2)	--	-
Vital Status for GBMs only			
Living	43 (14.1)	-	56 (19.9)
Deceased	262 (85.9)	-	225 (80.1)

[a] Southeastern study of Glioma in Adults

[b] The Cancer Genome Atlas

[c] Includes US residents in all the other States of the US

[d] Histological subtypes were defined as previously reported [16]

Table 2

Odds ratio and 95% Confidence intervals for the association between variants in *SMARCA2* and *SMARCA4* and the risk of histological subtypes of glioma

			Glioblastoma (Cases=305, Controls=574)		**Lower Grade Astrocytoma (Cases=138, Controls=574)**		**Oligodendroglioma (Cases=83, Controls=574))**		
Gene	SNP	Model[a]	OR (95%CI)[b]	*P*	OR (95%CI)[b]	*P*	OR (95%CI)[b]	*P*	*FDR-q*
SMARCA2	rs10964465	D	0.88 (0.62, 1.26)	0.48	0.77 (0.48, 1.26)	0.30	0.82 (0.46, 1.48)	0.52	0.39
	rs10964468	R	2.98 (0.95, 9.35)	0.06	2.17 (0.39, 11.99)	0.38	NA[c]	NA[c]	NA[c]
	rs13288443	A	1.24 (0.92, 1.67)	0.15	0.93 (0.61, 1.44)	0.76	0.87 (0.50, 1.50)	0.62	0.39
	rs2296212	R	1.23 (0.43, 3.55)	0.70	2.11 (0.60, 7.41)	0.24	4.05 (1.11, 14.8)	**0.030**	0.08
	rs4741636	A	0.93 (0.76, 1.14)	0.50	1.19 (0.90, 1.58)	0.22	0.92 (0.64, 1.30)	0.62	0.39
	rs4741651	R	1.10 (0.39, 3.10)	0.85	2..44 (0.78, 7.63)	0.13	4.68 (1.43, 15.3)	**0.011**	0.08
	rs7040174	D	0.90 (0.67, 1.21)	0.48	0.68 (0.45, 1.03)	0.07	1.05 (0.64, 1.71)	0.86	0.46
	rs7847382	D	1.02 (0.74, 1.41)	0.92	1.07 (0.69, 1.66)	0.76	0.53 (0.28, 1.00)	0.050	0.08
SMARCA4	rs11672232	R	0.75 (0.47, 1.19)	0.22	1.32 (0.75, 2.30)	0.34	1.90 (1.01, 3.58)	**0.048**	0.08
	rs11880865	R	0.84 (0.56, 1.25)	0.38	0.95 (0.56, 1.63)	0.85	1.38 (0.75, 2.52)	0.30	0.28
	rs12232780	R	0.91 (0.53, 1.55)	0.73	1.02 (0.51, 2.04)	0.96	2.14 (1.06, 4.33)	**0.035**	0.08
	rs17001086	D	1.05 (0.74, 1.48)	0.79	1.28 (0.82, 2.02)	0.28	1.09 (0.61, 1.95)	0.76	0.44
	rs17304534	D	0.92 (0.69, 1.23)	0.57	1.02 (0.68, 1.53)	0.93	1.03 (0.62, 1.70)	0.92	0.46
	rs6511718	R	1.19 (0.71, 1.98)	0.51	1.29 (0.64, 2.59)	0.47	1.69 (0.76, 3.74)	0.20	0.21
	rs7275	R	1.09 (0.66, 1.80)	0.75	1.17 (0.59, 2.34)	0.65	1.46 (0.66, 3.21)	0.35	0.29
	rs7935	R	0.83 (0.53, 1.30)	0.42	1.20 (0.69, 2.09)	0.52	1.75 (0.94, 3.28)	0.08	0.10

[a] Best fitting genetic model; A, log-additive; D, dominant; R, recessive

[b] Odds ratios and 95% confidence intervals obtained from multinomial logistic regression models adjusted for age (continuous) and gender (male/female)

[c] small numbers