Published in final edited form as:
Cancer Causes Control. 2014 April ; 25(4): 437–450. doi:10.1007/s10552-014-0346-x.

Single nucleotide polymorphisms in nucleotide excision repair genes, cancer treatment, and head and neck cancer survival

Annah B. Wyss,
Department of Epidemiology, Gillings School of Global Pubic Health, University of North Carolina at Chapel Hill, 2101B McGavran-Greenberg Hall, CB 7435, Chapel Hill, NC 27599, USA

Mark C. Weissler,
Department of Otolaryngology/Head and Neck Surgery, School of Medicine, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA

Christy L. Avery,
Department of Epidemiology, Gillings School of Global Pubic Health, University of North Carolina at Chapel Hill, 2101B McGavran-Greenberg Hall, CB 7435, Chapel Hill, NC 27599, USA

Amy H. Herring,
Department of Biostatistics, Gillings School of Global Public Health, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA, Carolina Population Center, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA

Jeannette T. Bensen,
Department of Epidemiology, Gillings School of Global Pubic Health, University of North Carolina at Chapel Hill, 2101B McGavran-Greenberg Hall, CB 7435, Chapel Hill, NC 27599, USA, Lineberger Comprehensive Cancer Center, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA

Jill S. Barnholtz-Sloan,
Case Comprehensive Cancer Center, Case Western Reserve University, Cleveland, OH, USA

William K. Funkhouser, and
Department of Pathology and Laboratory Medicine, School of Medicine, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA

Andrew F. Olshan
Department of Epidemiology, Gillings School of Global Pubic Health, University of North Carolina at Chapel Hill, 2101B McGavran-Greenberg Hall, CB 7435, Chapel Hill, NC 27599, USA, Carolina Population Center, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA, Lineberger Comprehensive Cancer Center, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA

Annah B. Wyss: alayman@email.unc.edu; Andrew F. Olshan: andy_olshan@unc.edu

Correspondence to: Andrew F. Olshan, andy_olshan@unc.edu.

Conflict of interest
The authors declare that they have no conflict of interest.

Electronic supplementary material The online version of this article (doi:10.1007/s10552-014-0346-x) contains supplementary material, which is available to authorized users.

Abstract

Purpose—Head and neck cancers (HNC) are commonly treated with radiation and platinum-based chemotherapy, which produce bulky DNA adducts to eradicate cancerous cells. Because nucleotide excision repair (NER) enzymes remove adducts, variants in NER genes may be associated with survival among HNC cases both independently and jointly with treatment.

Methods—Cox proportional hazards models were used to estimate race-stratified (White, African American) hazard ratios (HRs) and 95 % confidence intervals for overall (OS) and disease-specific (DS) survival based on treatment (combinations of surgery, radiation, and chemotherapy) and 84 single nucleotide polymorphisms (SNPs) in 15 NER genes among 1,227 HNC cases from the Carolina Head and Neck Cancer Epidemiology Study.

Results—None of the NER variants evaluated were associated with survival at a Bonferroni-corrected alpha of 0.0006. However, rs3136038 [OS HR = 0.79 (0.65, 0.97), DS HR = 0.69 (0.51, 0.93)] and rs3136130 [OS HR = 0.78 (0.64, 0.96), DS HR = 0.68 (0.50, 0.92)] of *ERCC4* and rs50871 [OS HR = 0.80 (0.64, 1.00), DS HR = 0.67 (0.48, 0.92)] of *ERCC2* among Whites, and rs2607755 [OS HR = 0.62 (0.45, 0.86), DS HR = 0.51 (0.30, 0.86)] of *XPC* among African Americans were suggestively associated with survival at an uncorrected alpha of 0.05. Three SNP-treatment joint effects showed possible departures from additivity among Whites.

Conclusions—Our study, a large and extensive evaluation of SNPs in NER genes and HNC survival, identified mostly null associations, though a few variants were suggestively associated with survival and potentially interacted additively with treatment.

Keywords

Head and neck cancer DNA repair; Nucleotide excision repair; Chemotherapy; Radiation; Survival

Background

An estimated 53,640 incident head and neck cancer (HNC) cases and 11,520 associated deaths occured in the United States during 2013 [1]. Comprising tumors of the oral cavity, pharynx, and larynx, HNC is a relatively fatal disease [2, 3]. Among individuals with oral and pharyngeal cancers in the United States, five-year survival rates are 61.7 and 63.2 % for White men and women, respectively, and 37.2 and 51.2 % for African American men and women, respectively [2, 3]. HNC was historically treated with surgery and/or radiation [4, 5]. However, following a series of clinical trials in the 1990s, advanced tumors (stages 3 and 4) are increasingly treated with concurrent or induction radiation and chemotherapy [4, 5]. Other tumor characteristics (e.g., location and size) and the patients' demographics (e.g., age) can also influence treatment decisions and outcomes [6].

Emerging literature suggests that genetic factors may also impact treatment response and survival among cancer patients [7, 8]. In order to initiate cell death (apoptosis) of cancerous cells, radiation and platinum-based chemotherapy are known to cause bulky DNA adducts, among other types of DNA damage [7, 9]. Since nucleotide excision repair (NER) is the pathway primarily responsible for removing DNA adducts, functional NER processes may lessen the efficacy of cancer treatment [7]. This hypothesis has led some researchers to

describe DNA repair, including NER, as a ‘‘double-edged sword’’ or ‘‘Janus, the two-faced Roman god,’’ since functional genes are thought to protect against cancer incidence, but may mitigate the effectiveness of cancer treatments thus decreasing survival [7].

Although it is hypothesized that the effects of NER variants on survival may be dependent on treatment, previous epidemiologic studies on the effects of single nucleotide polymorphisms (SNPs) in NER genes and treatment on HNC mortality have been inconsistent [9–17]. For example, some studies conducted among patients receiving radiation reported null associations for rs13181 in excision repair cross-complementing 2 (*ERCC2*) and survival [9, 12]. Other studies showed evidence for significant differences in survival across genotypes of rs13181 [10, 14, 15], including a study which found the referent genotype (AA) was associated with worse survival among individuals treated with radiation and better survival among those not receiving radiation [17]. However, previous studies have been based on small sample sizes, predominantly European-descent populations, and a limited number of variants in NER genes [9–17]. The present study extends the literature by estimating main and joint effects of treatment (combinations of surgery, radiation, and chemotherapy) and 84 SNPs across 15 NER genes on survival in a large, racially diverse group of HNC cases.

Methods

Study population

The Carolina Head and Neck Cancer Epidemiology (CHANCE) Study is a population-based case–control study of 1,389 cases and 1,396 controls from North Carolina (NC) [18–22]. For the present analysis, we compared survival among cases by treatment and genotype. All cases were 20–80 years of age and were identified from the NC Central Cancer Registry between 1 January 2002 and 28 February 2006 using rapid case ascertainment [18–22]. Cases with tumors in the oral cavity, oropharynx, hypo-pharynx, larynx, and HNC not otherwise specified (NOS) were included, while tumors of the salivary glands, naso-pharynx, nasal cavity, and nasal sinuses were excluded [18–22]. Self-reported demographic and behavioral information and biologic samples (~90 % blood, ~10 % buccal cells) were collected during a nurse-administered interview [18–22]. We excluded cases who self-reported race other than White or African American (n = 26, 1.9 %) because of sparse data, as well as lip cancers (n = 21, 1.3 %) because of etiologic differences. Cases who did not provide a biologic sample were also excluded; this comprised 52 cases (3.7 %) who were deceased at time of interview (i.e. proxy interviews) and eight cases (0.6 %) who provided in-person interviews but no biologic sample. Finally, cases whose samples were insufficient for genotyping or whose samples did not otherwise meet quality control criteria (n = 55, 4.0 %) were excluded. Our analysis included 1,227 HNC cases (922 White cases and 305 African American cases).

SNP selection and genotyping

Illumina GoldenGate assay with Sentrix Array Matrix and 96-well standard microtiter plates was used for genotyping [20–23]. As described previously [22], 71 tag SNPs in eight NER genes were selected based on a case–control study of HNC at The University of Texas MD

Anderson Cancer Center ($r^2 \geq 0.80$, minor allele frequency (MAF) ≥ 0.05, 1–2 kb flanking region, CEU population) and 58 SNPs in 12 NER genes were selected based on other cancer studies and/or potential function (Online Resource 1). Of the 129 NER SNPs, variants with poor signal intensity or genotype clustering (14 SNPs) or a MAF less than 0.05 (30 SNPs among Whites and 36 SNPs among African Americans) were excluded (Online Resource 1) [21-22]. The majority of excluded SNPs were candidate SNPs selected based on previous literature or function (Online Resource 1). Genotype frequencies for the remaining SNPs were consistent with Hardy–Weinberg equilibrium (HWE) among CHANCE controls at a Bonferroni-corrected 0.0006 alpha level, and scatter plots showed reasonable clustering; therefore, no SNPs were excluded for HWE violations [22]. Our analysis included 84 SNPs in 14 NER genes among Whites and 79 SNPs in 15 NER genes among African Americans.

Treatment

First-course treatment information was abstracted from patients' medical records. Information included whether the patient received surgery, radiation, and chemotherapy, including types of chemotherapy drugs: carboplatin, paraplatin, cisplatin, 5-FU, taxol, taxotere, docetaxel, paclit-axel, ifosfamide, and other [21]. Information on treatment start and end dates and whether radiation and chemotherapy were administered concurrently was not available for a large proportion of individuals. Therefore, combinations of treatment were generated from dichotomous variables for surgery, radiation, and chemotherapy regardless of timing. Information on tumor histology and stage was also abstracted from medical records [21]. Tumor grade was not uniformly available for all cases and therefore not considered.

HNC survival

CHANCE data were linked to the National Death Index (NDI) based on name, social security number, date of birth, sex, race, and state of residence to identify deaths through 2009, including date of death, location of death, and cause of death [21, 24]. Death records with HNC listed as an underlying cause of death were considered disease-specific deaths. For overall survival models, follow-up started at date of diagnosis for all cases and ended at date of death for individuals who died, or censoring on 31 December 2009 for individuals who were still alive [21]. For disease-specific survival models, follow-up started at date of diagnosis for all cases and ended at date of death for individuals who died of HNC, or censoring at date of death for individuals who died from causes other than HNC or 31 December 2009 for individuals who were still alive [21].

Statistical analysis

Cox proportional hazards models were used to estimate hazard ratios (HRs) and 95 % confidence intervals (CIs) for the independent and joint effects of treatment and SNPs on HNC survival among Whites and African Americans separately. To evaluate the proportionality of hazards, we examined adjusted log-negative log plots by treatment and by genotype separately. In addition, we assessed the significance of including an interaction term for time and treatment or genotype in models. If log-negative log plots indicated a violation of the proportional hazards assumption and interaction terms with time were

significant (p <>0.05), accelerated failure time (AFT) models were fit to explore robustness of results. This was the case for four SNPs in Whites (rs3731068, rs744154, rs3136085, and rs3136172) and three SNPs in African Americans (rs4150360, rs2020955, and rs13181). However, because p values for the AFT models were similar to those obtained from Cox models (i.e., the same set of significant SNP-HNC survival associations resulted from both approaches), results from the Cox models without an interaction term between SNPs and time are presented for simplicity. Absolute differences in HNC survival by treatment or genotype were also assessed via Kaplan–Meier plots, with cumulative survival calculated as the proportion of cases alive at each time point and log rank tests used to assess differences in survival.

Treatment—Treatment was modeled as a categorical variable with six groups: surgery only; radiation only; surgery and radiation; radiation and chemotherapy; surgery, radiation, and chemotherapy; and other (no treatment, chemotherapy only, or surgery and chemotherapy without radiation). Surgery only was used as the referent category because few individuals received no treatment (n = 9, 0.7 %). Even fewer individuals received chemotherapy only or chemotherapy with surgery without radiation (n = 4, 0.3 %), so these individuals were combined with individuals receiving no treatment into a single category labeled ‘‘other treatment.’’ In a separate model, we also considered receiving platinum-based chemotherapy drugs (carboplatin, paraplatin, or cisplatin, n = 464) versus not receiving platinum-based chemotherapy drugs (i.e., not receiving chemotherapy, n = 754, or only receiving non-platinum-based chemotherapy drugs, including 5 FU, taxol, taxotere, docetaxel, paclitaxel, or ifosfamide, n = 9). All treatment models were stratified by race and adjusted for sex, age (categorical), tumor stage (stages I, II, III, IV), tumor site (oral cavity, oropharynx, hypopharynx, larynx, NOS), education (high school or less, some college, and college or more), duration of cigarette smoking (years), and lifetime consumption of alcohol (categorical milliliters of ethanol).

SNPs—In agreement with previous CHANCE publications [22], SNPs were defined using a dominant genetic model and the referent allele for both Whites and African Americans was assigned to be the major allele based on controls from the overall study population. Race-stratified models included a single SNP at a time, with p values corrected using the Bonferroni method (0.05/84 = 0.0006 among Whites and 0.05/79 = 0.0006 among African Americans). The false discovery rate (FDR) approach to correcting for multiple comparisons was also considered as a supplementary analysis [25]. Further, SNP-survival associations with p values below 0.05 but not significant at a Bonferroni- or FDR-corrected alpha level were considered as ‘‘suggestive’’ associations. SNP models were adjusted for sex and age (including their interaction), as well as ancestry (proportion African ancestry). As described in previous studies of cancer among Whites and African Americans in NC, 145 ancestral informative markers (AIMS) were used to estimate the proportion of African and European ancestry of each participant based on Fisher’s information criterion (FIC) [20–22, 26–28]. Models did not include education, treatment, smoking or drinking because these variables were not considered confounders (i.e. these variables are not believed to effect germline variation), and models did not include tumor stage or site because these variables were considered causal intermediates.

Joint effects—Joint effects models included nine indicator variables for the interaction between treatment and a single SNP at a time: (1) surgery only, variant genotype; (2) radiation only, referent genotype; (3) radiation only, variant genotype; (4) surgery and radiation, referent genotype; (5) surgery and radiation, variant genotype; (6) radiation and chemotherapy, reference genotype; (7) radiation and chemotherapy, variant genotype; (8) surgery, radiation and chemotherapy, referent genotype; and (9) surgery, radiation and chemotherapy, variant genotype. Individuals receiving other treatment were excluded from this model due to small cell counts. Only joint effect estimates among Whites are presented because small cell counts among African Americans prohibited reliable estimation. Since both genetic and treatment exposures were assessed, models were adjusted for sex, age, tumor stage, tumor site, education, cigarette smoking, alcohol drinking, and ancestry. We also considered joint effects models for SNPs and platinum-based chemotherapy which included three disjoint indicator variables (no platinum-based chemotherapy, variant genotype; platinum-based chemotherapy, referent genotype; and platinum-based chemotherapy, variant genotype) and were adjusted for the covariates previously mentioned plus surgery (yes/no) and radiation (yes/no). Interactions between SNPs and treatments were assessed on the additive scale using the relative excess risk for interaction (RERI) with 95 % CIs calculated using the Hosmer and Lemeshow method [29]. Statistical analyses were performed using SAS 9.3 (Cary, NC) [30].

Results

Demographics

Of the 1,227 HNC cases in CHANCE, 545 (44.4 %) linked with the NDI through 2009 (Table 1). The remaining 682 (55.6 %) were assumed to be alive as of 31 December 2009. The median and mean follow-up times were 919.7 and 764.0 days, respectively, among individuals who died and 2,137.4 and 2,086.0 days, respectively, among those whowere alive. Among the 545 cases who died, just under half (n = 227, 41.7 %) had HNC listed as an underlying cause of death. Among these disease-specific deaths, the median and mean follow-up times were 729.7 and 594.0 days, respectively.

Modest variation by sex was observed when comparing cases who were living and dead after study follow-up (Table 1). However, a higher proportion of cases who died were diagnosed between 65 and 80 years of age (38.5% vs. 25.9%),were African American (29.7 vs. 21.0 %) or had a high school education or less (71.9 vs. 53.1 %) compared to living cases. With respect to tumor site, similar proportions had a diagnosis of laryngeal cancer (36.1 %). In contrast, 23.5 % of cases who died had oropharyngeal cancer compared to 30.1 % of cases who were living. As expected [31, 32], the distribution of tumor stage also varied by survival status, with living cases tending to have lower tumor stage.

Treatment

Among Whites, individuals who received only radiation tended to have worse overall (HR = 1.59, 95 % CI = 1.08, 2.34) and disease-specific survival (HR = 2.47, 95 % CI = 1.34, 4.56) compared to individuals who were treated with surgery alone (Table 2). Individuals receiving ‘‘other’’ treatment (i.e. no treatment, chemotherapy only or chemotherapy and

surgery) also appeared to have poorer overall and disease-specific survival, though estimates were imprecise since they were based on few individuals (data not shown). In a separate model, receiving platinum-based chemotherapy appeared to be associated with better overall (HR = 0.71, 95 % CI = 0.52, 0.95 among Whites and HR = 0.77, 95 % CI = 0.48, 1.20 among African Americans) and disease-specific survival (HR = 0.63, 95 % CI = 0.41, 0.97 among Whites and HR = 0.45, 95 % CI = 0.19, 1.02 among African Americans) (Table 2).

SNPs

Among Whites, four SNPs were modestly associated with only overall survival and three SNPs were modestly associated with both overall and disease-specific survival at an uncorrected 0.05 alpha level (Table 3). However, after correcting the alpha level using the Bonferroni and FDR methods, no SNPs were statistically significantly associated with either outcome. Among the SNPs associated with both outcomes at an uncorrected 0.05 alpha level, two tag SNPs were in linkage disequilibrium (LD) (r^2 = 0.92, CEU population) on *ERCC4*, also known as *xeroderma pigmentosum F* (*XPF*) [33]. Specifically, rs3136038 (TT + TC vs. CC) and rs3136130 (TT + GT vs. GG) were suggestively associated with a similarly reduced hazards of overall (HR = 0.79, uncorrected 95 % CI = 0.65, 0.97 and HR = 0.78, uncorrected 95 % CI 0.64, 0.96, respectively) and disease-specific death (HR = 0.69, uncorrected 95 % CI = 0.51, 0.93, and HR = 0.68, uncorrected 95 % CI = 0.50, 0.92, respectively). In addition, rs50871 (TT + TC vs. CC), a tag SNP on *ERCC2* also known as *XPD*, was suggestively associated with decreased hazards of overall (HR = 0.80, uncorrected 95 % CI = 0.64, 1.00) and disease-specific death (HR = 0.67, uncorrected 95 % CI = 0.48, 0.92). Stratifying by tumor stage, associations for rs3136038 or rs3136130 and survival were strongest among stage four cases and rs50871 among stage three cases (Online Resource 2). Figure 1 shows Kap-lan–Meier plots for these SNPs.

Among African Americans, two SNPs were associated with overall survival and four SNPs were associated with disease-specific survival at an uncorrected 0.05 alpha level, but none were significantly associated with survival at Bonferroni-corrected or FDR-corrected levels (Table 4). Only one tag SNP was associated with both overall and disease-specific survival at an uncorrected 0.05 alpha level. Specifically, rs2607755 (CC + CT vs. TT) on *XPC* was suggestively associated with reduced hazards of overall (HR = 0.62, uncorrected 95 % CI = 0.45, 0.86) and disease-specific death (HR = 0.51, uncorrected 95 % CI = 0.30, 0.86). This association was strongest among cases with stage 4 tumors (Online Resource 2). Figure 2 shows Kaplan–Meier plots for this SNP.

Joint effects

At an uncorrected 0.05 alpha level, four SNPs appeared to interact super-additively with radiation only, six SNPs appeared to interact super-additively with radiation and chemotherapy, and one SNP appeared to interact sub-additively with surgery, radiation, and chemotherapy, with respect to overall survival among Whites (Online Resource 3). Of these suggestive interactions, one SNP-radiation and two SNP-radiation, chemotherapy interactions were significant at a Bonferroni-corrected 0.0006 alpha level. Specifically, rs2972388 of *cyclin-dependent kinase 7* (*CDK7*) interacted super-additively with radiation only (RERI = 1.07, uncorrected 95 % CI = 0.55, 1.60) and with radiation and chemotherapy

(RERI = 0.72, uncorrected 95 % CI = 0.33, 1.10). In addition, rs2974752 of *RAD23 homolog A* (*RAD23A*) interacted super-additively with radiation and chemotherapy (RERI = 0.80, uncorrected 95 % CI = 0.36, 1.24). However, when disease-specific survival was considered, no SNP-treatment interactions were significant at a Bonferroni or FDR level among Whites (data not shown). Among African Americans, no SNP-treatment interactions appeared to be significant at a Bonferroni-corrected alpha level with respect to overall survival, though a super-additive interaction between rs1902658 of *XPC* and radiation, chemotherapy was significant when FDR was considered (RERI = 0.75, uncorrected 95 % CI = 0.29, 1.21). However, interaction estimates among African Americans were considered unreliable due to relatively low cell counts and are therefore not presented. With platinum-based chemotherapy, 10 SNPs suggested additive interactions at an uncorrected alpha level with respect to overall survival among Whites, but none were significant after correction for multiple comparisons (Online Resource 4).

Discussion

We detected mostly null associations between 84 SNPs in 15 NER genes and survival among White and African American HNC cases. Identifying null associations is important for following-up early positive associations, avoiding publication bias, and informing future meta-analyses [34]. To account for multiple comparisons, we principally used the Bonferroni approach, which though widely used in genetic epidemiology assumes independence of tests [25, 35, 36]. Given the correlated nature of SNPs, including some SNPs in our study, using the Bonferroni correction may be overly conservative potentially resulting in false negatives [25, 35, 36]. Therefore, we also considered the FDR approach as well as highlighted SNP-survival associations with *p* values below an uncorrected 0.05 alpha level as suggestive associations warranting further investigation.

Among Whites, we found that rs3136038 (near the 5' end) and rs3136130 (intron 5) of *ERCC4* were suggestively associated with improved overall and disease-specific survival [37, 38]. These SNPs are in LD with each other as well as several other untyped SNPs near or in introns or the 3′UTR of *ERCC4* (r^2 >0.80, CEU population) [33]. rs50871 of *ERCC2* intron 11 was also suggestively associated with improved survival among Whites [37, 38]. Among African Americans, rs2607755, which is located in intron 2 of *XPC* and is in LD with other intronic SNPs and the missense SNP rs2227998 (Arg687Ser, r^2 = 0.86, YRI population), was suggestively associated with improved overall and disease-specific survival [33, 37, 38]. The ERCC4 enzyme helps create an incision at the 5′ end of DNA adducts, while ERCC2 operates as a component of the transcription factor II H (TFIIH) subunit to denature the double helix in preparation for incisions [39, 40]. The XPC enzyme acts early in the NER pathway to recognize and bind with DNA adducts [39, 40]. Assuming that it is minor alleles which mitigate functional NER effects, thereby facilitating cancer treatment effects, one may expect variant genotypes of intronic SNPs, especially SNPs in regulatory regions or in LD with SNPs in coding regions, to be associated with improved survival, as was suggested by our findings for rs3136038, rs3136130, and rs50871 among Whites and rs2227998 among African Americans.

Although no previous HNC studies examined rs3136038 or rs3136130 on *ERCC4*, two studies assessed nine other *ERCC4* SNPs (rs1799799, rs1799801, rs3136105, rs3136146, rs3136152, rs3136155, rs3136166, rs3136189, rs3136202), many of which were in LD with the SNPs in our study (r^2 >0.80, CEU population) [10, 16, 33]. While five of these SNPs were not associated with progression-free survival among HNC cases, four SNPs appeared to be associated with worse progression-free survival contrary to our study [10, 16]. Among esophageal cancer cases, a study by Lee et al. [41] did assess rs3136038 reporting better overall survival associated with the genotype TT, though HRs were not statistically significant, similar to our study. Further, ERCC4 protein expression has been found to be elevated in HNC cell lines and displayed cisplatin resistance [42]. With respect to *ERCC2*, no previous studies have considered the effects of rs50871 on HNC survival. Rather, rs13181 and rs1799793 (which are not in LD with rs50871, CEU population) are the most commonly studied SNPs in *ERCC2*, with some studies reporting near null associations between these SNPs and survival among HNC cases [9, 10, 12, 14, 15, 33]. In our study, rs13181 was not associated with survival. Finally, no previous studies have considered associations between rs2607755 of *XPC* and survival, nor have any studies considered association between any NER variants and survival among African American HNC cases. Only one previous study has investigated a single variant in *XPC*, rs2228001 (which is not in LD with rs2607755, YRI population), noting no association with overall survival [9, 33]. Likewise, we did not find an association between rs2228001 and survival.

Since radiation and platinum-based chemotherapy are known to cause DNA damage repaired by NER genes [7, 9], we also considered associations between SNPs and survival among HNC cases in the context of treatment. Accounting for multiple comparisons using the Bonferroni method, we found that interactions between rs2972388, a synonymous SNP in *CDK7*, and radiation only, as well as radiation and chemotherapy, were more than additive with respect to overall survival among Whites [37, 38]. In addition, rs2974752, located near *RAD23A* and in LD with other SNPs near or in introns of this gene (r^2 >0.80, CEU population), interacted super-additively with radiation and chemotherapy [33, 37, 38]. However, these SNP-treatment interactions were not significant at a Bonferroni level when disease-specific survival was considered and should therefore be interpreted with some caution. Further, genotype frequencies for rs2972388 and rs2974752 were consistent with HWE at a Bonferroni-corrected 0.0006 alpha level, but not at a 0.05 alpha level. 0.05 alpha level. No previous studies have considered *CDK7* or *RAD23A* SNPs in relation to treatment and HNC survival. Only one previous study has compared NER SNP-survival associations across strata of treatment regimens [17]. Specifically, Zhong et al. [17] analyzed the effect of rs13181 in *ERCC2* on survival among 275 HNC cases receiving radiotherapy and 210 cases not receiving radiotherapy. Among cases with stage 3 and 4 tumors, the referent genotype (AA) was associated with poorer overall survival among those treated with radiation, but better survival among those who did not receive radiation [17]. Among cases with stage 1 and 2 tumors who did not receive radiation, rs13181 was not associated with survival [17].

With a population-based study of 1,227 HNC cases, the present analysis included more than double the number of HNC cases of the next largest study [10]. Study populations of

previous publications were mostly hospital-based and ranged from 47 to 531 HNC cases [10, 13]. Further, the present study population included 922 White cases and 305 African Americans cases which allowed for estimation of race-specific HRs. Linkage disequilibrium is known to vary by ancestral populations and distinct differences in survival by race occur in the United States [2, 3, 33]. Yet, prior to this study, no studies had considered NER SNP-survival associations among African American HNC cases. Another contribution of our study was the broad evaluation of NER variants which included a large number of SNPs that have not been previously evaluated. Previous studies have collectively examined approximately 18 SNPs in six NER genes and survival among HNC cases [9–17]. Our study included 84 SNPs across 15 NER genes.

Although our study included the largest study population and broadest array of NER SNPs to date to our knowledge, a few limitations should be noted. We were unable to include proxy interviews (52 cases, 3.7 %) in our analysis since these occurred for individuals who died prior to interview and therefore did not provide a biologic sample. If SNPs were related to aggressive tumors, then estimates for SNP-survival associations may be slightly attenuated to the null [21]. When SNP-survival associations were stratified by stage, associations were strongest among cases with stage 3 and 4 tumors. Further, follow-up was started at date of diagnosis, rather than date of interview (i.e., date of blood draw), since diagnosis is a more clinically informative time point. To assess the potential immortal person-time bias this may have introduced [43], we conducted sensitivity analyses with follow-up starting at date of interview. No material differences in results were noted across models, though rs50871 was suggestively associated with overall survival among Whites with an unadjusted p value of 0.05 in the primary analysis, but with a p value of 0.06 in the sensitivity analysis, and the interaction between rs1902658 and radiation, chemotherapy with respect to overall survival among African Americans was significant at an FDR level in the primary analysis, but at a Bonferroni level in the sensitivity analysis.

Other potential limitations include the following. First, because both tagging and candidate SNPs were included and selected based on European-descent populations, variation captured across some genes was limited, especially among African Americans. For this reason, haplotypes were not explored. Second, treatment was considered solely as the first-course combinations of dichotomous variables for surgery, radiation, and chemotherapy abstracted from medical records. Although treatment is fairly standardized based on tumor stage and site as well as other patient demographics [4, 5], information on duration of treatment (e.g., start and end dates) and timing of treatments combinations (e.g., concurrent chemotherapy) may have been informative. Third, treatment-SNP joint effect estimates were imprecise among Whites and unreliable among African Americans due to small cell counts. Fourth, models were adjusted for cigarette and alcohol information that was ascertained at baseline based on behaviors prior to diagnosis since information on behavioral risk factors following diagnosis was not uniformly available. Further, we did not have information on human papillomavirus infection, a known predictor of survival among cases with oropharyngeal tumors [44]. Finally, we did not have access to information on recurrent tumors and therefore did not consider disease-free or relapse-free survival.

In summary, most NER variants did not appear to be associated with survival among HNC cases. However, three SNPs in Whites (rs3136038 and rs3136130 of *ERCC4* and rs50871 of *ERCC2*) and one SNP among African Americans (rs2607755 of *XPC*) were suggestively associated with better overall and disease-specific survival. Therefore, it is recommended that future genetic epidemiology studies of HNC survival include these SNPs for replication. In addition, two SNPs appeared to possibly interact additively with radiation with or without chemotherapy among Whites. While our study is the largest to date, it is only the second to consider NER SNP-treatment joint effects on HNC survival. Therefore, additional studies with even larger sample sizes are needed to evaluate gene–environment interactions more precisely. Further studies focusing on African American and other diverse populations are recommended.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

This work was supported by the US National Institutes of Health (NIH), National Cancer Institute (NCI) [R01-CA90731-01; 2T32CA009330-21-26], and National Institute of Environmental Health Sciences (NIEHS) [T32ES007018; P30ES010126]. Dr. Avery was supported in part by grant R00-HL-098458 from the National Heart, Lung, and Blood Institute (NHBLI). The authors thank Dr. Robert Millikan for his substantial contributions to the conceptual development and analyses of this paper. We also thank Dr. Anne Hakenewerth for assistance in research development and Ms. Kathy Wisniewski for programming support.

References

1. American Cancer Society. [Accessed on 9 Feb 2013] Cancer facts and figures 2013. 2013. http://www.cancer.org/acs/groups/content/@epidemiologysurveilance/documents/document/acspc-031941.pdf
2. National Cancer Institute. [Accessed on 9 Dec 2013] Relative survival by survival time by race/ethnicity oral cavity and pharynz, all age, male, 1992–2009. 2013. http://seer.cancer.gov/faststats/selections.php?#Output
3. National Cancer Institute. [Accessed on 9 Dec 2013] Relative survival by survival time by race/ethnicity oral cavity and pharynz, all age, female, 1992–2009. 2013. http://seer.cancer.gov/faststats/selections.php?#Output
4. Vokes EE, Weichselbaum RR, Lippman SM, Hong WK. Head and neck cancer. N Engl J Med. 1993; 328(3):184–194. [PubMed: 8417385]
5. Forastiere A, Koch W, Trotti A, Sidransky D. Head and neck cancer. N Engl J Med. 2001; 345(26): 1890–1900. [PubMed: 11756581]
6. National Cancer Institute. [Accessed on 10 Dec 2013] General information about lip and oral cavity cancer. 2013. http://www.cancer.gov/cancertopics/pdq/treatment/lip-and-oral-cavity/Patient
7. Gazdar AF. DNA repair and survival in lung cancer—the two faces of Janus. N Engl J Med. 2007; 356(8):771–773. [PubMed: 17314336]
8. Braakhuis BJ, Brakenhoff RH, Leemans CR. Treatment choice for locally advanced head and neck cancers on the basis of risk factors: biological risk factors. Ann Oncol. 2012; 23(Suppl 10):173–177.
9. Carles J, Monzo M, Amat M, Jansa S, Artells R, Navarro A, et al. Single-nucleotide polymorphisms in base excision repair, nucleotide excision repair, and double strand break genes as markers for response to radiotherapy in patients with Stage I to II head-and-neck cancer. Int J Radiat Oncol Biol Phys. 2006; 66(4):1022–1030. [PubMed: 16979838]

10. Azad AK, Bairati I, Samson E, Cheng D, Mirshams M, Qiu X, et al. Validation of genetic sequence variants as prognostic factors in early-stage head and neck squamous cell cancer survival. Clin Cancer Res. 2012; 18(1):196–206. [PubMed: 22076708]
11. De Castro G Jr, Pasini FS, Siqueira SA, Ferraz AR, Villar RC, Snitcovsky IM, et al. ERCC1 protein, mRNA expression and T19007C polymorphism as prognostic markers in head and neck squamous cell carcinoma patients treated with surgery and adjuvant cisplatin-based chemoradiation. Oncol Rep. 2011; 25(3):693–699. [PubMed: 21206986]
12. Gal TJ, Huang WY, Chen C, Hayes RB, Schwartz SM. DNA repair gene polymorphisms and risk of second primary neoplasms and mortality in oral cancer patients. Laryngoscope. 2005; 115(12): 2221–2231. [PubMed: 16369171]
13. Grau JJ, Caballero M, Campayo M, Jansa S, Vargas M, Alos L, et al. Gene single nucleotide polymorphism accumulation improves survival in advanced head and neck cancer patients treated with weekly paclitaxel. Laryngoscope. 2009; 119(8):1484–1490. [PubMed: 19504558]
14. Mahimkar MB, Samant TA, Kannan S, Tulsulkar J, Pai PS, Anantharaman D. Polymorphisms in GSTM1 and XPD genes predict clinical outcome in advanced oral cancer patients treated with postoperative radiotherapy. Mol Carcinog. 2012; 51(Suppl 1):E94–E103. [PubMed: 22213390]
15. Quintela-Fandino M, Hitt R, Medina PP, Gamarra S, Manso L, Cortes-Funes H, et al. DNA-repair gene polymorphisms predict favorable clinical outcome among patients with advanced squamous cell carcinoma of the head and neck treated with cis-platin-based induction chemotherapy. J Clin Oncol. 2006; 24(26):4333–4339. [PubMed: 16896002]
16. Vaezi A, Wang X, Buch S, Gooding W, Wang L, Seethala RR, et al. XPF expression correlates with clinical outcome in squamous cell carcinoma of the head and neck. Clin Cancer Res. 2011; 17(16):5513–5522. [PubMed: 21737503]
17. Zhong S, Nukui T, Buch SC, Diergaarde B, Weissfeld LA, Grandis JR, et al. Effects of ERCC2 Lys751Gln (A35931C) and CCND1 (G870A) polymorphism on outcome of advanced-stage squamous cell carcinoma of the head and neck are treatment dependent. Cancer Epidemiol Biomark Prev. 2011; 20(11):2429–2437.
18. Divaris K, Olshan AF, Smith J, Bell ME, Weissler MC, Funkhouser WK, et al. Oral health and risk for head and neck squamous cell carcinoma: the Carolina Head and Neck Cancer Study. Cancer Causes Control. 2010; 21(4):567–575. [PubMed: 20049634]
19. Stingone JA, Funkhouser WK, Weissler MC, Bell ME, Olshan AF. Racial differences in the relationship between tobacco, alcohol, and squamous cell carcinoma of the head and neck. Cancer Causes Control. 2013; 24(4):649–664. [PubMed: 22674225]
20. Hakenewerth AM, Millikan RC, Rusyn I, Herring AH, North KE, Barnholtz-Sloan JS, et al. Joint effects of alcohol consumption and polymorphisms in alcohol and oxidative stress metabolism genes on risk of head and neck cancer. Cancer Epidemiol Biomark Prev. 2011; 20(11):2438–2449.
21. Hakenewerth AM, Millikan RC, Rusyn I, Herring AH, Weissler MC, Funkhouser WK, et al. Effects of polymorphisms in alcohol metabolism and oxidative stress genes on survival from head and neck cancer. Cancer Epidemiol. 2013; 37(4):479–491. [PubMed: 23632049]
22. Wyss AB, Herring AH, Avery C, Weissler MC, Bensen JT, Barnholtz-Sloan JS, et al. Single Nucleotide Polymorphisms in Nucleotide Excision Repair Genes, Cigarette Smoking, and the Risk of Head and Neck Cancer. Cancer Epidemiol Biomark Prev. 2013; 22(8):1428–1445.
23. Illumina. [Accessed on 25 Oct 2011] GoldenGate assay workflow. 2006. http://www.illumina.com/documents/products/workflows/workflow_goldengate_assay.pdf
24. Centers for Disease Control and Prevention. [Accessed on 10 Dec 2013] National death index. 2013. http://www.cdc.gov/nchs/ndi.htm
25. Benjamini Y, Hochberg Y. Controlling the false discovery rate: a practical and powerful approach to multiple testing. J R Stat Soc Ser B (Methodol). 1995; 1995:289–300.
26. Pfaff CL, Barnholtz-Sloan J, Wagner JK, Long JC. Information on ancestry from genetic markers. Genet Epidemiol. 2004; 26(4):305–315. [PubMed: 15095390]
27. Barnholtz-Sloan JS, McEvoy B, Shriver MD, Rebbeck TR. Ancestry estimation and correction for population stratification in molecular epidemiologic association studies. Cancer Epidemiol Biomark Prev. 2008; 17(3):471–477.

28. Barnholtz-Sloan JS, Shetty PB, Guan X, Nyante SJ, Luo J, Brennan DJ, et al. FGFR2 and other loci identified in genome-wide association studies are associated with breast cancer in African-American and younger women. Carcinogenesis. 2010; 31(8):1417–1423. [PubMed: 20554749]
29. Hosmer DW, Lemeshow S. Confidence interval estimation of interaction. Epidemiology. 1992; 3(5):452–456. [PubMed: 1391139]
30. SAS Institute Inc. SAS 9.3. SAS Institute Inc; Cary, NC: 2011.
31. National Cancer Institute. [Accessed on 10 Dec 2013] SEER stat fact sheets: oral cavity and pharynx cancer. 2013. http://seer.cancer.gov/statfacts/html/oralcav.html
32. National Cancer Institute. [Accessed on 10 Dec 2013] SEER stat fact sheets: larynx cancer. 2013. http://seer.cancer.gov/statfacts/html/laryn.html
33. Johnson AD, Handsaker RE, Pulit S, Nizzari MM, O'Donnell CJ, de Bakker PIW. SNAP: a web-based tool for identification and annotation of proxy SNPs using HapMap. Bioinformatics. 2008; 24(24):2938–2939. [PubMed: 18974171]
34. Ioannidis JP, Ntzani EE, Trikalinos TA, Contopoulos-Ioannidis DG. Replication validity of genetic association studies. Nat Genet. 2001; 29(3):306–309. [PubMed: 11600885]
35. Benjamini Y, Drai D, Elmer G, Kafkafi N, Golani I. Controlling the false discovery rate in behavior genetics research. Behav Brain Res. 2001; 125(1):279–284. [PubMed: 11682119]
36. Rice TK, Schork NJ, Rao DC. Methods for handling multiple testing. Adv Genet. 2008; 60:293–308. [PubMed: 18358325]
37. National Center for Biotechnology Information, National Library of Medicine. [Accessed on Feb 2013] Database of single nucleotide polymorphisms (dbSNP). 2013. http://www.ncbi.nlm.nih.gov/SNP/
38. Meyer LR, Zweig AS, Hinrichs AS, Karolchik D, Kuhn RM, Wong M, et al. The UCSC genome browser database: extensions and updates 2013. Nucleic Acids Res. 2013; 41(D1):D64–D69. [PubMed: 23155063]
39. Friedberg EC. How nucleotide excision repair protects against cancer. Nat Rev Cancer. 2001; 1(1): 22–33. [PubMed: 11900249]
40. Friedberg, EC.; Walker, GC.; Siede, W.; Wood, RD.; Schultz, RA. DNA repair and mutagenesis. ASM Press; Washington, DC: 2006.
41. Lee JM, Yang PW, Yang SY, Chuang TH, Tung EC, Chen JS, et al. Genetic variants in DNA repair predicts the survival of patients with esophageal cancer. Ann Surg. 2011; 253(5):918–927. [PubMed: 21490450]
42. Koberle B, Ditz C, Kausch I, Wollenberg B, Ferris RL, Albers AE. Metastases of squamous cell carcinoma of the head and neck show increased levels of nucleotide excision repair protein XPF in vivo that correlate with increased chemoresistance ex vivo. Int J Oncol. 2010; 36(5):1277–1284. [PubMed: 20372803]
43. Lash TL, Cole SR. Immortal person-time in studies of cancer outcomes. J Clin Oncol. 2009; 27(23):e55–e56. [PubMed: 19597013]
44. Ragin CC, Taioli E. Survival of squamous cell carcinoma of the head and neck in relation to human papillomavirus infection: review and meta-analysis. Int J Cancer. 2007; 121(8):1813–1820. [PubMed: 17546592]

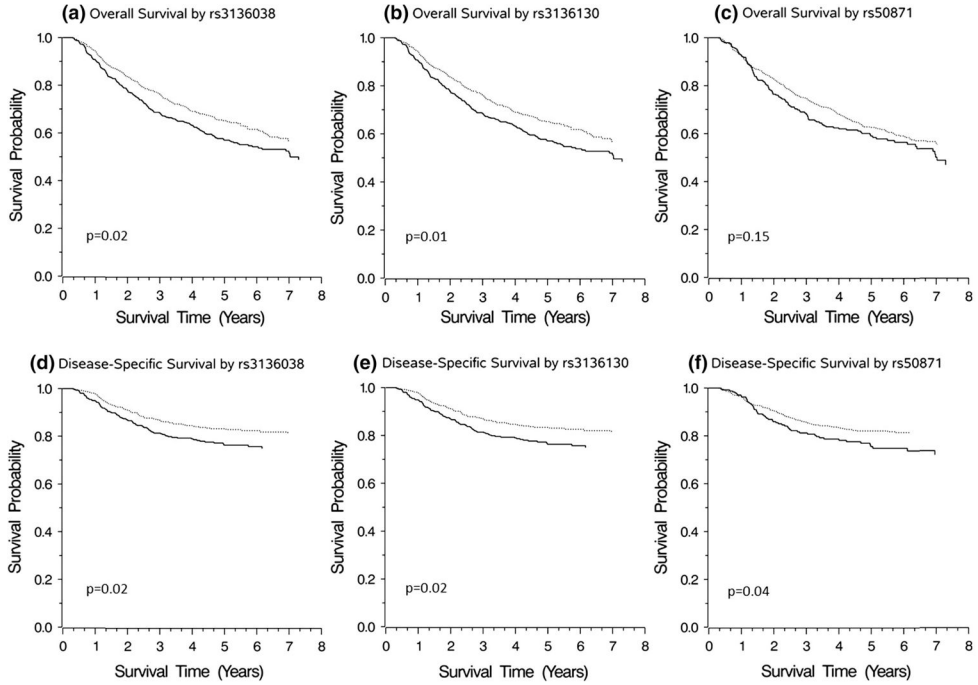

Fig. 1.

Kaplan–Meier plots for overall (OS) and disease-specific (DS) survival by select genotype among White HNC cases in the Carolina Head and Neck Cancer Epidemiology (CHANCE) study. *Solid line* represents individuals with referent genotype, while *dashed line* represents individuals with variant genotype

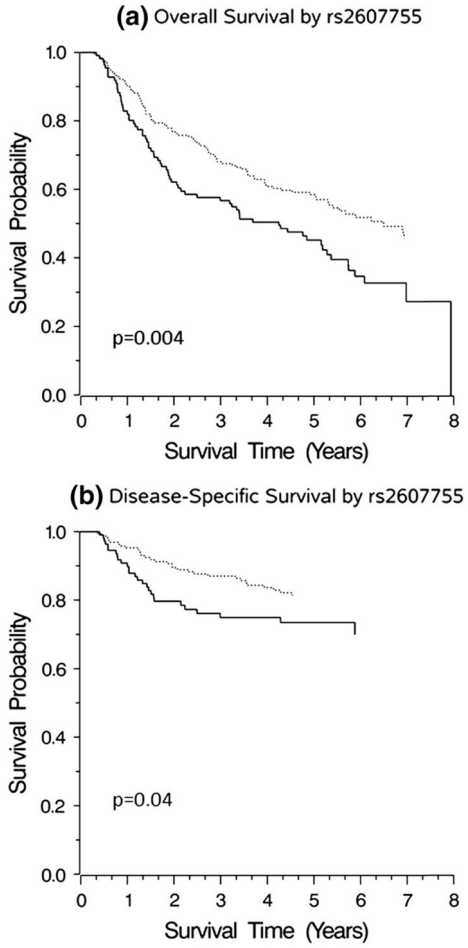

Fig. 2.

Kaplan–Meier plots for overall (OS) and disease-specific (DS) survival by select genotypes among African American HNC cases in the Carolina Head and Neck Cancer Epidemiology (CHANCE) study. *Solid line* represents individuals with referent genotype, while *dashed line* represents individuals with variant genotype

Table 1

Demographic characteristics of head and neck cancer cases, Carolina Head and Neck Cancer Epidemiology (CHANCE) study, 2002–2009

Characteristic	Alive		Overall deaths		Disease- specific deaths	
	n	%	*n*	%	*n*	%
Total	682	55.6	545	44.4	227	
Sex						
Male	519	76.1	419	76.9	169	74.4
Female	163	23.9	126	23.1	58	25.6
Race/ethnicity						
White	539	79.0	383	70.3	169	74.4
African American	143	21.0	162	29.7	58	25.6
Age at diagnosis						
20–49	152	22.3	87	16.0	42	18.5
50–54	110	16.1	79	14.5	30	13.2
55–59	125	18.3	82	15.0	35	15.4
60–64	118	17.3	87	16.0	37	16.3
65–69	78	11.4	90	16.5	33	14.5
70–74	64	9.4	71	13.0	27	11.9
75–80	35	5.1	49	9.0	23	10.1
Education						
High school or less	362	53.1	392	71.9	157	69.2
Some college	195	28.6	99	18.2	48	21.1
College or more	125	18.3	54	9.9	22	9.7
Tumor site						
Oral cavity	81	11.9	91	16.7	38	16.7
Oropharynx	205	30.1	128	23.5	54	23.8
Hypophaynx	16	2.3	39	7.2	15	6.6
NOS	134	19.6	90	16.5	42	18.5
Larynx	246	36.1	197	36.1	78	34.4
Stage						
I	195	28.6	84	15.4	17	7.5

Cancer Causes Control. Author manuscript; available in PMC 2014 July 15.

Characteristic	Alive		Overall deaths		Disease- specific deaths	
	n	%	*n*	%	*n*	%
II	119	17.4	101	18.5	38	16.7
III	118	17.3	93	17.1	40	17.6
IV	250	36.7	267	49.0	132	58.1
Surgery						
No	269	39.4	268	49.2	119	52.4
Yes	413	60.6	277	50.8	108	47.6
Radiation						
No	177	26.0	105	19.3	37	16.3
Yes	505	74.0	440	80.7	190	83.7
Chemotherapy						
No	428	62.8	326	59.8	130	57.3
Yes	254	37.2	219	40.2	97	42.7
Mean follow-up time (days)	2,137.4		919.7		729.7	
Median follow-up time (days)	2,086.0		764.0		594.0	

Table 2

Hazard ratios for cancer treatment and head and neck cancer survival in the Carolina Head and Neck Cancer Epidemiology (CHANCE) study

Cancer treatment	Whites					African Americans				
	Overall deaths	HNC deaths	Alive	Overall survival HR (95 % CI)[a,b]	Disease-specific survival HR (95 % CI)[a,b]	Overall deaths	HNC deaths	Alive	Overall survival HR (95 % CI)[a,b]	Disease-specific survival[a,b] HR (95 % CI)
Surgery only	68	21	144	1.00 (Referent)	1.00 (Referent)	26	9	31	1.00 (Referent)	1.00 (Referent)
Radiation only	77	36	83	1.59 (1.08, 2.34)	2.47 (1.34, 4.56)	24	7	31	1.00 (0.52, 1.92)	0.90 (0.26, 3.05)
Surgery and radiation	85	35	106	1.19 (0.81, 1.73)	1.28 (0.69, 2.36)	38	16	32	1.10 (0.63, 1.94)	1.13 (0.44, 2.86)
Radiation and chemotherapy	102	50	118	1.19 (0.78, 1.81)	1.48 (0.78, 2.81)	55	19	36	1.00 (0.56, 1.80)	0.52 (0.19, 1.39)
Surgery, radiation, chemotherapy	43	22	87	0.98 (0.61, 1.59)	1.26 (0.61, 2.58)	16	5	12	0.77 (0.36, 1.64)	0.36 (0.09, 1.39)
Platinum-based chemotherapy										
Did not receive platinum-based chemotherapy	239	98	336	1.00 (Referent)	1.00 (Referent)	93	34	95	1.00 (Referent)	1.00 (Referent)
Did receive platinum-based chemotherapy	144	71	203	0.71 (0.52, 0.95)	0.63 (0.41, 0.97)	69	24	48	0.77 (0.48, 1.22)	0.45 (0.19, 1.02)

HR hazards ratio, *CI* confidence interval

[a] HRs adjusted for age and sex (including pairwise interaction), tumor stage, tumor site, education, cigarette smoking, and alcohol drinking

[b] Platinum-based chemotherapy HRs adjusted for age and sex (including pairwise interaction), tumor stage, tumor site, education, cigarette smoking, alcohol drinking, and surgery and/or radiation. Fifty-seven White cases and 20 African American cases missing information on alcohol drinking and therefore dropped from models

Table 3

Hazard ratios for single nucleotide polymorphisms (SNPs) in nucleotide excision repair (NER) genes and survival among head and neck cancer cases in the Carolina Head and Neck Cancer Epidemiology (CHANCE) study, Whites

Gene	SNP	Coded allele		Overall deaths/deaths from HNC/ alive						Overall survival		Disease-specific survival	
		Referent (A)	Variant (B)	AA			AB + BB			HR (95 % CI)[a]	*p* value	HR (95 % CI)[a]	*p* value
ERCC3 (XPB)	rs4150496	G	A	168	67	233	215	102	305	0.94 (0.77, 1.15)	0.55	1.10 (0.81, 1.51)	0.53
	rs1011019	C	T	191	84	271	192	85	268	1.02 (0.83, 1.24)	0.88	1.03 (0.76, 1.39)	0.87
	rs4150434	G	A	230	110	318	153	59	221	0.99 (0.80, 1.21)	0.91	0.80 (0.58, 1.11)	0.18
	rs4150416	T	G	165	69	245	217	99	292	1.10 (0.90, 1.35)	0.34	1.20 (0.88, 1.64)	0.24
	rs4150407	A	G	131	47	187	252	122	352	0.99 (0.80, 1.23)	0.94	1.32 (0.94, 1.85)	0.11
	rs4150403	G	A	303	139	433	80	30	106	1.02 (0.79, 1.31)	0.88	0.81 (0.55, 1.21)	0.31
	rs4150402	G	A	191	84	271	192	85	268	1.02 (0.83, 1.24)	0.88	1.03 (0.76, 1.39)	0.87
XPC	rs2228001	A	C	135	58	202	248	111	336	1.13 (0.92, 1.40)	0.24	1.16 (0.84, 1.60)	0.36
	rs3731143	T	C	333	147	485	50	22	54	1.18 (0.87, 1.59)	0.29	1.16 (0.73, 1.82)	0.53
	rs2228000	C	T	213	94	311	168	74	228	1.04 (0.85, 1.28)	0.68	1.04 (0.76, 1.41)	0.82
	rs3731124	A	C	215	94	306	168	75	233	1.02 (0.83, 1.26)	0.83	1.08 (0.79, 1.47)	0.63
	rs13099160	A	G	335	146	479	48	23	60	1.13 (0.83, 1.53)	0.44	1.17 (0.75, 1.82)	0.50
	rs3731093	T	C	321	141	455	59	26	79	1.03 (0.78, 1.36)	0.83	0.98 (0.64, 1.49)	0.91
	rs3731089	G	A	321	141	457	62	28	82	1.05 (0.80, 1.38)	0.74	1.02 (0.68, 1.53)	0.94
	rs2733537	A	G	167	73	249	216	96	290	1.07 (0.87, 1.31)	0.53	1.06 (0.78, 1.44)	0.71
	rs3731068	C	A	257	111	367	126	58	172	1.09 (0.88, 1.35)	0.45	1.16 (0.84, 1.60)	0.36
	rs2607755	T	C	100	46	142	283	123	397	0.99 (0.79, 1.25)	0.95	0.96 (0.68, 1.35)	0.80
	rs1902658	G	A	99	46	136	284	123	402	0.97 (0.77, 1.22)	0.78	0.92 (0.65, 1.29)	0.61
ERCC8	rs3117	T	C	126	51	211	257	118	328	1.22 (0.98, 1.51)	0.07	1.37 (0.99, 1.91)	0.06
CDK7	rs2972388	A	G	110	50	156	273	119	383	0.99 (0.79, 1.24)	0.94	0.95 (0.68, 1.33)	0.76
XPA	rs3176757	C	T	256	114	353	127	55	186	0.96 (0.77, 1.19)	0.71	0.93 (0.67, 1.29)	0.68
	rs3176753	T	C	381	169	537				0.47 (0.06, 3.70)	0.47		
	rs2808667	C	T	345	153	469	36	16	70	0.70 (0.50, 1.00)	0.05	0.74 (0.44, 1.25)	0.26
	rs2805835	G	C	301	133	426	82	36	113	1.00 (0.78, 1.28)	0.97	0.94 (0.65, 1.36)	0.74
	rs3176689	A	T	267	118	355	116	51	184	0.90 (0.72, 1.12)	0.34	0.91 (0.65, 1.26)	0.57
	rs3176683	T	C	342	152	476	41	17	63	0.85 (0.61, 1.18)	0.33	0.80 (0.48, 1.33)	0.40

Gene	SNP	Coded allele		Overall deaths/deaths from HNC/ alive						Overall survival		Disease-specific survival	
		Referent (A)	Variant (B)	AA			AB + BB			HR (95 % CI)[a]	*p* value	HR (95 % CI)[a]	*p* value
	rs3176658	C	T	294	137	405	89	32	134	0.93 (0.73, 1.18)	0.55	0.72 (0.49, 1.07)	0.10
	rs1800975	G	A	184	87	236	185	77	280	0.88 (0.71, 1.08)	0.22	0.78 (0.57, 1.07)	0.12
RAD23B	rs1805330	C	T	306	136	458	77	33	81	1.34 (1.04, 1.72)	0.03	1.28 (0.87, 1.88)	0.21
	rs1805329	C	T	257	113	333	126	56	206	0.81 (0.65, 1.00)	0.05	0.83 (0.60, 1.14)	0.25
ERCC6	rs2228529	A	G	252	108	345	127	59	186	0.94 (0.76, 1.17)	0.58	1.03 (0.75, 1.42)	0.86
	rs2228527	A	G	251	107	347	132	62	192	0.96 (0.78, 1.19)	0.70	1.06 (0.78, 1.46)	0.70
	rs4253132	T	C	294	132	429	89	37	110	1.08 (0.85, 1.37)	0.55	1.01 (0.69, 1.46)	0.98
	rs2228528	G	A	265	115	372	117	53	167	1.05 (0.84, 1.31)	0.67	1.09 (0.78, 1.51)	0.61
DDB2 (XPE)	rs2029298	A	G	170	68	255	213	101	284	1.05 (0.86, 1.29)	0.63	1.31 (0.96, 1.79)	0.09
	rs4647709	C	T	317	137	449	66	32	90	1.06 (0.81, 1.39)	0.68	1.21 (0.82, 1.79)	0.34
	rs2291120	T	C	283	126	402	100	43	137	1.11 (0.88, 1.39)	0.39	1.06 (0.75, 1.50)	0.73
	rs1685404	G	C	172	71	246	211	98	293	1.08 (0.88, 1.32)	0.48	1.20 (0.88, 1.64)	0.24
	rs2957873	A	G	257	109	386	126	60	153	1.11 (0.90, 1.38)	0.33	1.29 (0.94, 1.78)	0.11
	rs326224	G	A	279	119	404	104	50	135	1.05 (0.84, 1.32)	0.65	1.24 (0.89, 1.74)	0.20
	rs2306353	G	A	281	121	415	102	48	124	1.10 (0.87, 1.38)	0.43	1.25 (0.89, 1.75)	0.20
	rs326222	C	T	190	81	294	193	88	245	1.09 (0.89, 1.34)	0.39	1.21 (0.89, 1.64)	0.22
	rs901746	A	G	190	81	295	193	88	244	1.10 (0.90, 1.34)	0.37	1.22 (0.89 1.65)	0.21
ERCC5 (XPG)	rs2296147	T	C	130	45	150	250	123	387	0.78 (0.62, 0.97)	0.02	1.11 (0.79, 1.58)	0.54
	rs4771436	T	G	227	99	336	156	70	203	1.09 (0.89, 1.35)	0.39	1.13 (0.83, 1.55)	0.43
	rs1047768	C	T	122	62	197	261	107	342	1.20 (0.97, 1.50)	0.10	0.96 (0.70, 1.32)	0.82
	rs3818356	C	T	227	99	336	155	69	202	1.09 (0.89, 1.34)	0.41	1.12 (0.82, 1.53)	0.46
	rs4150351	A	C	254	114	341	129	55	198	0.84 (0.68, 1.04)	0.12	0.81 (0.59, 1.13)	0.22
	rs4150355	C	T	177	66	225	206	103	314	0.86 (0.70, 1.05)	0.14	1.15 (0.84, 1.57)	0.37
	rs4150360	T	C	106	55	169	277	114	370	1.18 (0.94, 1.48)	0.15	0.93 (0.67, 1.29)	0.67
	rs4150383	G	A	255	112	375	128	57	164	1.11 (0.90, 1.38)	0.34	1.13 (0.82, 1.56)	0.47
	rs4150386	A	C	304	133	420	79	36	119	1.03 (0.80, 1.33)	0.81	1.05 (0.72, 1.53)	0.80
	rs17655	C	G	223	112	332	160	57	207	1.15 (0.94, 1.41)	0.18	0.81 (0.59, 1.11)	0.19
	rs873601	A	G	190	93	274	193	76	265	1.05 (0.86, 1.28)	0.65	0.84 (0.62, 1.14)	0.25
	rs4150393	A	G	296	132	406	87	37	133	0.89 (0.70, 1.13)	0.34	0.88 (0.61, 1.26)	0.48
	rs876430	C	T	191	94	274	192	75	265	1.04 (0.85, 1.27)	0.72	0.82 (0.60, 1.11)	0.19

Gene	SNP	Coded allele		Overall deaths/deaths from HNC/ alive						Overall survival		Disease-specific survival	
		Referent (A)	Variant (B)	AA			AB + BB			HR (95 % CI)[a]	*p* value	HR (95 % CI)[a]	*p* value
	rs1051677	T	C	303	129	432	80	40	106	1.06 (0.83, 1.36)	0.63	1.28 (0.89, 1.82)	0.18
	rs1051685	A	G	302	141	434	81	28	104	1.04 (0.81, 1.34)	0.74	0.79 (0.52, 1.19)	0.26
ERCC4 (XPF)	rs3136038	C	T	184	86	218	199	83	321	0.79 (0.65, 0.97)	0.03	0.69 (0.51, 0.93)	0.02
	rs1799798	G	A	320	143	437	63	26	102	0.90 (0.69, 1.19)	0.47	0.83 (0.55, 1.27)	0.40
	rs744154	C	G	208	96	272	175	73	267	0.88 (0.72, 1.08)	0.21	0.77 (0.57, 1.05)	0.10
	rs3136085	G	C	205	96	270	178	73	269	0.88 (0.72, 1.08)	0.23	0.75 (0.55, 1.02)	0.06
	rs3136130	G	T	184	86	216	199	83	323	0.78 (0.64, 0.96)	0.02	0.68 (0.50, 0.92)	0.01
	rs1800067	G	A	322	144	456	61	25	83	0.99 (0.75, 1.31)	0.96	0.89 (0.58, 1.37)	0.61
	rs3136172	A	G	195	91	263	188	78	276	0.92 (0.75, 1.13)	0.43	0.79 (0.58, 1.08)	0.14
RAD23A	rs2974752	A	G	135	58	198	235	106	326	1.04 (0.84, 1.29)	0.72	1.10 (0.80, 1.52)	0.57
ERCC2 (XPD)	rs13181	T	G	154	73	227	224	94	310	1.07 (0.87, 1.31)	0.53	0.94 (0.69, 1.28)	0.69
	rs238418	C	A	156	74	226	227	95	313	1.05 (0.86, 1.29)	0.63	0.92 (0.68, 1.25)	0.59
	rs1799787	C	T	196	87	276	187	82	263	1.02 (0.83, 1.25)	0.85	1.00 (0.74, 1.35)	0.99
	rs3916874	G	C	209	96	268	174	73	271	0.85 (0.69, 1.04)	0.12	0.75 (0.55, 1.02)	0.07
	rs238416	G	A	152	68	217	231	101	321	1.00 (0.81, 1.23)	1.00	0.99 (0.73, 1.35)	0.96
	rs50872	C	T	226	91	305	157	78	232	0.95 (0.77, 1.16)	0.59	1.16 (0.86, 1.58)	0.34
	rs50871	T	G	110	55	132	273	114	407	0.80 (0.64, 1.00)	0.05	0.67 (0.48, 0.92)	0.01
	rs238407	A	T	121	54	142	262	115	396	0.81 (0.65, 1.01)	0.06	0.83 (0.60, 1.15)	0.27
	rs3810366	C	G	79	31	99	304	138	439	0.89 (0.69, 1.15)	0.36	1.09 (0.74, 1.62)	0.66
ERCC1	rs735482	A	C	278	131	410	105	38	129	1.13 (0.90, 1.42)	0.28	0.86 (0.60, 1.23)	0.40
	rs2336219	G	A	278	131	410	105	38	129	1.13 (0.90, 1.42)	0.28	0.86 (0.60, 1.23)	0.40
	rs3212964	G	A	280	132	412	103	37	127	1.13 (0.90, 1.41)	0.31	0.84 (0.58, 1.21)	0.34
	rs3212955	A	G	209	94	319	174	75	220	1.15 (0.94, 1.41)	0.18	1.10 (0.81, 1.50)	0.54
	rs3212948	C	G	154	73	228	229	96	311	1.06 (0.86, 1.30)	0.58	0.93 (0.68, 1.26)	0.64
	rs3212930	T	C	241	108	335	142	61	204	0.95 (0.77, 1.18)	0.66	0.93 (0.68, 1.27)	0.64
LIG1	rs156641	G	A	151	74	219	232	95	320	1.02 (0.83, 1.26)	0.84	0.86 (0.63, 1.17)	0.33
	rs20580	C	A	97	46	140	286	123	399	1.03 (0.82, 1.30)	0.78	0.94 (0.66, 1.32)	0.70
	rs20579	C	T	294	127	397	89	42	142	0.90 (0.71, 1.15)	0.41	1.01 (0.71, 1.44)	0.94

Confidence intervals presented not corrected for multiple comparisons. Significant associations using a dominant genetic model (p <>0.05) highlighted in bold. None significant at a Bonferroni-corrected level (p <0.0006)

HR hazards ratio, *CI* confidence interval

[a] HR for dominant genetic model (AB + BB vs. AA). HRs adjusted for age and sex (including pairwise interaction) and ancestry (proportion African ancestry)

Table 4

Hazard ratios for single nucleotide polymorphisms (SNPs) in nucleotide excision repair (NER) genes and survival among head and neck cancer cases in the Carolina Head and Neck Cancer Epidemiology (CHANCE) study, African Americans

Gene	SNP	Coded allele		Overall deaths/deaths from HNC/alive						Overall survival		Disease-specific survival	
		Referent (A)	Variant (B)	AA			AB + BB			HR (95 % CI)[a]	*p* value	HR (95 % CI)[a]	*p* value
ERCC3 (XPB)	rs4150496	G	A	95	30	83	66	28	59	0.98 (0.71, 1.36)	0.93	1.40 (0.82, 2.38)	0.22
	rs4150459	G	A	101	35	89	61	23	54	1.08 (0.78, 1.50)	0.65	1.14 (0.66, 1.98)	0.63
	rs1011019	C	T	94	34	89	68	24	54	1.06 (0.77, 1.47)	0.72	1.08 (0.62, 1.86)	0.78
	rs4150434	G	A	126	46	106	36	12	37	0.84 (0.57, 1.24)	0.38	0.79 (0.41, 1.55)	0.50
	rs4150416	T	G	39	13	46	122	44	97	1.32 (0.91, 1.90)	0.14	1.47 (0.78, 2.80)	0.23
	rs4150407	A	G	43	13	41	119	45	102	1.13 (0.79, 1.62)	0.49	1.35 (0.72, 2.53)	0.35
	rs4150402	G	A	94	34	89	68	24	54	1.06 (0.77, 1.47)	0.72	1.08 (0.62, 1.86)	0.78
XPC	rs2228001	A	C	91	32	89	71	26	54	1.18 (0.85, 1.63)	0.32	1.36 (0.79, 2.32)	0.26
	rs2228000	C	T	137	50	114	25	8	29	0.77 (0.50, 1.21)	0.26	0.65 (0.30, 1.42)	0.28
	rs3731124	A	C	136	49	116	26	9	27	0.91 (0.60, 1.40)	0.67	0.75 (0.37, 1.53)	0.43
	rs3731093	T	C	140	54	123	19	3	19	0.86 (0.51, 1.45)	0.57	0.44 (0.14, 1.43)	0.17
	rs3731089	G	A	140	54	123	22	4	20	0.91 (0.56, 1.48)	0.70	0.52 (0.19, 1.47)	0.22
	rs2733537	A	G	115	46	97	47	12	46	0.86 (0.60, 1.23)	0.40	0.59 (0.30, 1.13)	0.11
	rs2607755	T	C	70	27	41	92	31	102	0.62 (0.45, 0.86)	0.004	0.51 (0.30, 0.86)	0.01
	rs1902658	G	A	30	10	23	132	48	120	0.94 (0.62, 1.43)	0.78	0.87 (0.43, 1.76)	0.71
ERCC8	rs3117	T	C	72	21	54	90	37	89	0.81 (0.59, 1.12)	0.20	1.15 (0.66, 1.99)	0.62
CDK7	rs2972388	A	G	77	30	83	85	28	60	1.36 (0.98, 1.87)	0.06	1.04 (0.61, 1.77)	0.88
CCNH	rs2266691	A	G	140	52	117	22	6	26	0.83 (0.53, 1.32)	0.44	0.54 (0.23, 1.28)	0.16
	rs2266692	G	T	133	46	104	29	12	39	0.68 (0.45, 1.03)	0.07	0.80 (0.41, 1.53)	0.49
XPA	rs3176757	C	T	126	42	110	36	16	33	1.00 (0.68, 1.46)	0.98	1.42 (0.78, 2.59)	0.25
	rs3176753	T	C	123	40	102	39	18	41	0.89 (0.62, 1.29)	0.54	1.17 (0.66, 2.08)	0.58
	rs3176748	A	G	132	53	118	30	5	25	1.03 (0.68, 1.56)	0.88	0.40 (0.16, 1.02)	0.05
	rs3176658	C	T	141	51	119	21	7	24	0.89 (0.55, 1.44)	0.64	0.88 (0.38, 2.00)	0.76
	rs1800975	G	A	100	32	87	55	54	51	1.05 (0.74, 1.47)	0.80	1.44 (0.82, 2.55)	0.21
RAD23B	rs1805330	C	T	93	32	90	69	26	53	1.05 (0.76, 1.47)	0.76	1.15 (0.67, 1.99)	0.61
ERCC6	rs2228529	A	G	123	48	111	39	10	30	1.17 (0.81, 1.69)	0.41	0.74 (0.37, 1.49)	0.40

Gene	SNP	Coded allele		Overall deaths/deaths from HNC/alive						Overall survival		Disease-specific survival	
		Referent (A)	Variant (B)	AA			AB + BB			HR (95 % CI)[a]	*p* value	HR (95 % CI)[a]	*p* value
	rs2228527	A	G	115	44	103	47	14	40	1.04 (0.74, 1.48)	0.82	0.81 (0.44, 1.50)	0.51
	rs4253132	T	C	97	35	94	65	23	49	1.13 (0.82, 1.56)	0.46	1.17 (0.69, 1.99)	0.57
	rs2228528	G	A	107	40	105	54	18	38	1.46 (1.04, 2.04)	0.03	1.22 (0.69, 2.14)	0.50
DDB2 (XPE)	rs2029298	A	G	43	16	47	119	42	96	1.23 (0.86, 1.76)	0.26	1.27 (0.71, 2.30)	0.42
	rs1685404	G	C	86	31	78	76	27	65	1.07 (0.77, 1.47)	0.70	0.97 (0.56, 1.66)	0.90
	rs2957873	A	G	49	20	41	112	38	102	0.95 (0.67, 1.35)	0.78	0.79 (0.46, 1.38)	0.41
	rs326224	G	A	43	14	37	119	44	106	1.12 (0.78, 1.60)	0.54	1.21 (0.66, 2.22)	0.55
	rs2306353	G	A	60	21	46	102	37	97	0.88 (0.64, 1.22)	0.45	0.93 (0.54, 1.59)	0.78
	rs326222	C	T	26	12	28	136	46	115	1.17 (0.76, 1.80)	0.47	0.88 (0.46, 1.68)	0.70
	rs901746	A	G	35	15	30	127	43	113	0.96 (0.66, 1.41)	0.85	0.79 (0.43, 1.44)	0.44
ERCC5 (XPG)	rs2296147	T	C	101	32	92	60	25	51	1.07 (0.77, 1.48)	0.70	1.42 (0.84, 2.42)	0.19
	rs2296148	C	T	122	45	106	39	13	37	0.96 (0.66, 1.39)	0.81	0.93 (0.49, 1.75)	0.82
	rs4771436	T	G	110	35	94	52	23	49	0.90 (0.64, 1.26)	0.54	1.24 (0.73, 2.12)	0.43
	rs1047768	C	T	66	21	50	96	37	93	0.87 (0.63, 1.20)	0.39	1.06 (0.61, 1.84)	0.83
	rs2020915	G	A	103	42	102	59	16	41	1.22 (0.88, 1.70)	0.24	0.82 (0.45, 1.49)	0.52
	rs3818356	C	T	110	35	96	52	23	47	0.93 (0.66, 1.31)	0.68	1.27 (0.75, 2.18)	0.37
	rs4150355	C	T	119	39	99	43	19	44	0.93 (0.64, 1.34)	0.68	1.16 (0.65, 2.05)	0.62
	rs4150360	T	C	11	5	8	151	53	135	0.79 (0.42, 1.50)	0.48	0.65 (0.25, 1.70)	0.38
	rs4150383	G	A	131	42	111	31	16	32	0.88 (0.59, 1.31)	0.53	1.34 (0.75, 2.39)	0.33
	rs17655	C	G	44	19	46	118	39	97	1.09 (0.76, 1.56)	0.65	0.84 (0.47, 1.50)	0.56
	rs873601	A	G	14	8	16	148	50	127	1.16 (0.66, 2.02)	0.61	0.76 (0.35, 1.62)	0.47
	rs876430	C	T	15	8	17	147	50	126	1.17 (0.68, 2.00)	0.58	0.84 (0.39, 1.79)	0.65
	rs1051677	T	C	116	43	116	46	15	27	1.31 (0.92, 1.86)	0.14	1.15 (0.63, 2.11)	0.64
	rs1051685	A	G	77	30	61	85	28	82	0.85 (0.62, 1.15)	0.29	0.72 (0.43, 1.20)	0.20
ERCC4 (XPF)	rs3136038	C	T	43	17	49	119	41	94	1.26 (0.88, 1.81)	0.20	1.13 (0.64, 2.01)	0.67
	rs744154	C	G	114	40	107	48	18	36	1.16 (0.82, 1.64)	0.42	1.27 (0.72, 2.26)	0.41
	rs3136085	G	C	92	34	81	70	24	62	1.06 (0.76, 1.46)	0.75	1.01 (0.59, 1.75)	0.96
	rs3136091	C	G	131	45	124	31	13	19	1.27 (0.84, 1.90)	0.25	1.55 (0.82, 2.94)	0.18
	rs3136130	G	T	39	16	36	123	42	107	1.04 (0.72, 1.51)	0.84	0.90 (0.50, 1.63)	0.74
	rs3136172	A	G	112	39	104	50	19	39	1.12 (0.80, 1.59)	0.51	1.24 (0.70, 2.19)	0.46

Gene	SNP	Coded allele		Overall deaths/deaths from HNC/alive						Overall survival		Disease-specific survival	
		Referent (A)	Variant (B)	AA			AB + BB			HR (95 % CI)[a]	*p* value	HR (95 % CI)[a]	*p* value
	rs2020955	T	C	101	37	92	61	21	51	1.11 (0.80, 1.55)	0.53	1.05 (0.60, 1.83)	0.86
RAD23A	rs2974752	A	G	39	12	38	116	42	100	1.11 (0.77, 1.62)	0.57	1.29 (0.67, 2.48)	0.45
	rs11558955	A	G	135	47	121	27	11	22	0.99 (0.65, 1.51)	0.97	1.14 (0.58, 2.22)	0.71
ERCC2 (XPD)	rs13181	T	G	86	31	87	76	27	55	1.24 (0.90, 1.70)	0.19	1.18 (0.70, 2.00)	0.54
	rs238418	C	A	5	4	3	157	54	139	0.87 (0.34, 2.27)	0.78	0.27 (0.09, 0.82)	0.02
	rs1799787	C	T	120	40	115	42	18	28	1.36 (0.95, 1.95)	0.09	1.65 (0.93, 2.91)	0.09
	rs3916874	G	C	142	53	126	20	5	17	0.94 (0.58, 1.54)	0.82	0.59 (0.23, 1.52)	0.27
	rs238416	G	A	129	44	114	32	14	29	1.04 (0.70, 1.55)	0.83	1.33 (0.72, 2.45)	0.36
	rs50872	C	T	116	42	107	46	16	36	1.08 (0.76, 1.54)	0.66	0.98 (0.54, 1.76)	0.94
	rs50871	T	G	120	36	109	42	22	34	1.07 (0.74, 1.54)	0.73	1.93 (1.11, 3.35)	0.02
	rs238407	A	T	114	39	112	48	19	31	1.22 (0.86, 1.72)	0.27	1.36 (0.77, 2.38)	0.29
	rs3810366	C	G	108	39	104	54	19	39	1.13 (0.81, 1.59)	0.47	1.04 (0.59, 1.83)	0.89
ERCC1	rs735482	A	C	83	31	73	79	27	70	0.94 (0.69, 1.29)	0.72	0.86 (0.51, 1.46)	0.58
	rs2336219	G	A	84	31	73	78	27	70	0.93 (0.67, 1.27)	0.63	0.87 (0.51, 1.47)	0.59
	rs3212964	G	A	111	43	96	49	15	47	0.98 (0.69, 1.38)	0.89	0.76 (0.41, 1.39)	0.37
	rs3212955	A	G	82	31	77	80	27	66	0.92 (0.67, 1.27)	0.62	0.92 (0.54, 1.55)	0.75
	rs3212948	C	G	5	3	4	157	55	139	0.64 (0.24, 1.73)	0.38	0.36 (0.09, 1.37)	0.13
	rs3212935	A	G	81	32	62	81	26	81	0.84 (0.61, 1.16)	0.28	0.65 (0.38, 1.10)	0.11
	rs3212930	T	C	131	44	119	31	14	24	1.09 (0.73, 1.62)	0.68	1.43 (0.77, 2.64)	0.26
LIG1	rs156641	G	A	129	43	105	33	15	38	0.76 (0.51, 1.14)	0.19	1.13 (0.61, 2.09)	0.70
	rs20580	C	A	33	8	29	129	50	113	1.00 (0.68, 1.48)	0.99	1.62 (0.76, 3.45)	0.21
	rs20579	C	T	83	26	67	79	32	76	0.91 (0.66, 1.26)	0.58	1.32 (0.77, 2.25)	0.31
	rs439132	A	G	88	27	84	74	31	59	1.10 (0.79, 1.52)	0.57	1.48 (0.86, 2.55)	0.15

Confidence intervals presented not corrected for multiple comparisons. Significant associations using a dominant genetic model (p <>0.05) highlighted in bold. None significant at a Bonferroni-corrected level (p <>0.0006)

HR hazards ratio, *CI* confidence interval

[a]HR for dominant genetic model (AB + BB vs. AA). HRs adjusted for age and sex (including pairwise interaction) and ancestry (proportion African ancestry)