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Genetic Predictors of Taxane-induced Neurotoxicity in a SWOG Phase III Intergroup Adjuvant Breast Cancer Treatment Trial (S0221)

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Abstract

Purpose—We assessed SNPs in the Fanconi Anemia (FA)/BRCA pathway in women being treated with taxanes for breast cancer in an effort to find a prognostic biomarker for neurological toxicities, which while improve survival remain a debilitating outcome.

Patients and Methods—We used data and samples (n=888) from SWOG0221, a phase III adjuvant trial of 4 dose/schedules of cyclophosphamide (C), doxorubicin (A) and paclitaxel (T) for high risk breast cancer. The relationship of SNPs in the (FA)/BRCA pathway with risk grade 3/4 neurotoxicities. In a separate cohort we measured the correlation of significant SNPs in this pathway with corresponding gene expression.

Results—No associations between SNPs in *BRCA1* and taxane-induced neuropathy was found. For *FANCD2*, significant associations between 4 (out of 20) SNPs and neurological toxicities, with risk estimates approaching 2. Two *FANCD2* haplotypes were also found to be significantly associated with neurological toxicity, increasing the odds in the overall population 1.8 (95%

confidence interval (CI) 1.3–2.5) and 1.7 (95% CI, 1.2–2.4) fold. Although numbers were small, there appeared to be a specific African-American haplotype that was associated with an almost 3-fold increase in risk of neurological toxicity. Expression analyses revealed that significant *FANCD2* SNPs were associated with *FANCD2* expression levels ($p=0.03$)

Conclusion—SNPs in *FANCD2*, were associated with a 70 to 80% increase in the odds of grade 3/4 neurological toxicities and increased expression of the gene. If replicated, women with these genotypes should be closely monitored for toxicities, and could be targeted for preventive measures or alternative therapeutic approaches.

INTRODUCTION

The addition of taxanes (T) to doxorubicin (Adriamycin®, A) and cyclophosphamide (C) in the adjuvant setting for patients with early stage breast cancer reduces risk of recurrence and mortality. However, treatment efficacy does not come without a price (1–5). Patients receiving taxanes may develop distal axonopathy with symmetrical sensory neuropathic pain and abnormal sensations such as dysesthesias, numbness and altered proprioception. The onset is early in therapy and can last months to years in the survivorship period (6). In a CALGB adjuvant trial of paclitaxel (C9344), 15% of patients had moderate parasthesias following treatment, and 3% had sensory neurotoxicity that interfered with daily functioning (1). Clearly, identification of biomarkers that predict taxane-induced neurotoxicity would be of value so that, balanced with efficacy, doses could be adjusted, preventive interventions could be focused, or these agents could be avoided for those at greatest risk.

The mechanisms for taxane-induced neuropathy have not been completely elucidated. Taxanes disrupt normal microtubule dynamics (7), resulting in mitotic arrest and possibly apoptosis (8). Emerging evidence supports a potential role of DNA damage as a means of taxane cytotoxicity. In cultured cancer cell lines, accumulated DNA breaks are observed after exposure to paclitaxel (9,10), and over-expression of DNA repair genes involved in chromosome instability appear to be associated with taxane resistance (11). Thus, deficiencies in known DNA repair pathways might result in increased toxicity to taxanes. The Fanconi anemia (FA)/BRCA genes are highly polymorphic, and several studies have demonstrated that SNPs in these genes result in alterations of their enzymatic products and consequently their functions (12–14). Thus, we evaluated potential associations between taxane-induced neurotoxicity and SNPs in *BRCA1* and *FANCD2* genes in a cohort of 888 women enrolled in an ongoing cooperative group clinical trial for breast cancer.

PATIENTS AND METHODS

Patient population

We used data and samples from a North American Breast Cancer Intergroup clinical trial led by the Southwest Oncology Group (SWOG) S0221. This is a phase III trial of continuous schedule AC - paclitaxel (Taxol®, T) vs. Q2 week schedule AC, followed by T given either every 2 weeks or weekly for 12 weeks as post-operative adjuvant therapy. The premise of the trial is to test if a continuous ‘metronomic’ schedule is superior to an accelerated but more conventional schedule of AC - T for breast cancer.

To be eligible for the trial, patients must have node-positive or high-risk (tumor ≥ 2 cm) node-negative operable Stage II or III invasive breast cancer with known ER/PR status and no prior cancer, have undergone breast surgery, and have no prior chemotherapy. Blood is drawn upon registration to S0221 and shipped to the Roswell Park Cancer Institute (RPCI) laboratory, where it is processed and stored at -80° . The total accrual goal for S0221 is 3,250 patients and as of August 2010, 2,666 patients had been enrolled; as of early 2009 when genotyping was performed, blood was available on 905 women. The trial schema and

patient numbers are presented in Figure 1 and methods and results are reported according to REMARK criteria (15).

Toxicities evaluated

During the trial, toxicities are monitored and reported using the NCI Common Toxicity Criteria for Adverse Events (CTCAE) Version 3.0, which contains descriptive terminology to be used for adverse event (AE) reporting, with a grading (severity) scale provided for each AE term. Primary AEs with the taxane treatment are neurological toxicities, including pain, motor coordination difficulties and myalgias. Grade 3 toxicities interfere with activities of daily living and grade 4 are life-threatening and often require hospitalization. In S0221, 12% of patients have experienced grade 3 or 4 neurotoxicities. In addition to the clinical assessment of neurotoxicities, patients are also queried at multiple time points using the Functional Assessment of Cancer Therapy-Taxane (FACT-TAX) (16). The FACT-TAX allows us to further examine associations between genotypes and patient reported neurologic toxicities. The analysis reported herein was approved by the Data Safety and Management Committee (DSMC). S0221 is still accruing and therefore we cannot evaluate toxicity or genotype with cancer outcomes, or interactions of the SNPs with randomized treatment, until the primary study results are available.

SNP selection and laboratory methods

The open-source software program SNAGGER, (<http://sourceforge.net/projects/snagger>) was used to select SNPs in *FANCD2* and *BRCA1*. In addition to a minimum minor allele frequency (MAF) of 0.05 and an r^2 threshold of 0.80, we prioritized the SNP list by chromosomal position, functionality and Illumina design scores. For the total population, 24 and 21 tag SNPs in *FANCD2* and *BRCA1*, respectively, were selected using SNAGGER. Quality control (QC) was monitored by the inclusion of 5% blind duplicates, and concordance was 99.1%. Eight SNPs were removed from analyses due to: low MAF, violation of Hardy Weinberg Equilibrium proportions (HWE) in those without toxicity, and genotyping quality <95%. After QC, 20 SNPs for *FANCD2* and 17 SNPs for *BRCA1* were evaluated among patients who self-reported as White/Other, and 9 SNPs in *FANCD2* and 12 in *BRCA1* among African American (AA) women. To control for potential confounding due to cryptic population substructure, we included SNPs to estimate individual ancestry (IA), selecting 70 uncorrelated SNPs from HapMap in both Yoruban African and unrelated CEPH Caucasian populations (17). Population admixture proportions were determined using the Bayesian clustering algorithms implemented in the program STRUCTURE v2.2 (18).

DNA was extracted from whole blood using Qiagen Flexigene kits, according to manufacturer's instructions, and quantified using both PicoGreen and Nanodrop™ technologies. DNA was plated onto 96-well plates and genotyping was conducted using the Illumina GoldenGate platform in the RPCI Genomics Core Facility. IA measures showed that European ancestry was estimated at 0.97 among the 740 patients who self-reported as White and 0.16 among those who self-reported as Black (n=67). The remaining study population was primarily Asian, and average proportion of EA ancestry was 0.93; thus, these patients (n=41) were included in analyses with women self-reporting as white. For QC, we removed individuals with > 15% genotypes missing (n=7) and patients who self-reported as black but had more than 0.85 proportion of EA ancestry (n=2), leaving 888 participants for analyses.

Statistical analysis

The proportion of EA admixture was included as a covariate in all models to control for potential confounding due to cryptic population substructure, as well as age and treatment group. We performed both single SNP and haplotype analyses using logistic models. For all

analyses, the reference population included individuals with grades 0, 1 or 2 neurological toxicities versus grades 3/4. For single SNP tests of association, we used additive (dominant) coding for variant alleles when the prevalence of the least frequent homozygote was >0.05 (<0.05). Permutation analyses were used to account for multiple testing. The method of Gabriel et al (19) was used to determine haplotypes for each gene within whites and AAs and the overall population. Each haplotype configuration, weighted by its probability, was used in the logistic regression models to test for association with grade 3/4 toxicity. Given the final population under study ($n=888$), after conservative correction for multiple testing we have 80% power to detect minimum odds ratio of grade 3/4 toxicity of 1.8 assuming MAF >0.15 .

To evaluate if SNPs that were significantly associated with CTCAE grade 3/4 toxicity were also associated with self-reported toxicities, the sixteen neuropathy components of the FACT-TAX were summed (20) with a maximum score of 64 indicating no neuropathy. We categorized the neuropathy sums into tertiles, and then used ordinal models to assess the odds of higher neuropathy compared to lower by genotype.

Ex vivo association of FANCD2 genotypes and gene expression

We evaluated if significant tag SNPs in *FANCD2* were significantly associated with gene expression using human lymphoblastoid cell lines from 63 unrelated European American women without a diagnosis of cancer in an independent cohort (21). Microarray-based gene expression was assayed using the Illumina HumanHT-12v3 Gene Expression Beadchip, with more than 48,000 probes derived from the National Center for Biotechnology Information Reference Sequence (NCBI) RefSeq (Build 36.2, Rel 22) and the UniGene (Build 199) databases. The raw intensity was scanned and extracted using BeadScan and the data corrected by local background subtraction in GenomeStudio module. The log₂ transformed intensity data was normalized with the Quantile normalization algorithm. *FANCD2* gene was expressed in all lymphoblastoid cell lines; the expression-detection p-value was less than 0.05 across all samples. Linear regression was used to examine associations between SNPs and mean log₂ *FANCD2* expression levels under dominant and additive models, adjusted for age.

RESULTS

The characteristics of the study population in this analysis are shown in Table 1. Approximately half of the patients were less than 51 years of age, and the majority of patients were white. According to CTC criteria, 107 (12%) patients experienced grade 3 or 4 neurotoxicity. The median FACT-TAX neurotoxicity score reported six months after chemotherapy initiation was 46 (range 6–64), with tertile cutpoints of 39 and 52.

Allele frequencies and HWE test p-values for SNPs evaluated by toxicity status ($<$ grade 3, grades 3/4) are shown in Table 2. Although the minor alleles for both *FANCD2* and *BRCA1* did not differ by race, with the exception of rs3172417 in *FANCD2*, the distributions of minor allele frequencies for all remaining SNPs in *FANCD2* were higher in the black population, with, on average, approximately 20% greater frequency in black women than white and Asian women (data not shown). For *BRCA1*, minor allele frequencies were slightly more common in white/Asian than black women. Although differing in frequencies, the minor alleles were the same in both populations; thus, we considered the population collectively, as well as by race, when analyzing both genes. Several of the *FANCD2* SNPs, but no *BRCA1* SNPs, violated HWE proportions assumption at the $\alpha=0.05$ among those experiencing toxicity, but not among those without grade 3 and 4 toxicities.

The block structure for *FANCD2* and associations between SNPs and toxicity are shown in Figure 2. While it is evident that linkage disequilibrium (LD) is strong across the entire gene, when examining the LD structure by race, populations differed, with less correlation across typed SNPs among blacks. As expected, the LD structure of *BRCA1* was almost identical across populations, comprising 2 haplotypes (data not shown). Given the strong and varying LD in *FANCD2*, we measured associations for both single SNPs and haplotypes in relation to clinical neurotoxicity in the overall population as well as stratified by race. We did not conduct haplotype analyses for *BRCA1* due to the similar LD structure across populations and no evidence of single SNP associations with grade 3/4 neurotoxicity for $\alpha=0.05$ (Table 3).

For *FANCD2*, the four most significant single SNP associations ($p<0.001$) were found within intronic regions (rs7648104, rs7637888, rs6786638, rs6442150, Figure 2A), yielding increases in odds of neurotoxicity by approximately two fold. In analyses stratified by race, associations were of borderline significance in both the white/Asian and black populations, likely due to small numbers. When these SNPs were examined in relation to the FACT-TAX scores, ORs, although somewhat lower with slightly wider confidence intervals than when using CTC criteria, were also significantly associated with neurotoxicity when compared to women who experienced no neurotoxicity (rs7637888, OR=1.47 (CI=1.00–2.15), rs6786638 OR=1.47 (CI=1.00–2.15), rs6442150 OR=1.47 (CI=1.00–2.15) and rs7648104 (OR=1.53, CI=1.05, 2.25).

Results of significant ($\alpha=0.05$) haplotype tests of association with grade 3/4 neurotoxicity are presented in Table 4. In the black population, 3 haplotype blocks spanning 2 SNPs each were detected; each haplotype block consisted of 2 haplotypes with a frequency greater than 1%. The most significant 2-SNP haplotype was found at rs7648104-rs7637888, with black women with A-G haplotype (28%) having an almost three fold greater odds of grade 3/4 neurotoxicity than those with C-A haplotype (72%). This haplotype was less frequent (15%) in White or Asian women. Importantly, among women with grade 3 and 4 neurotoxicity, the frequency of this haplotype in White or Asian women was approximately 1/3 that of black women (20% versus 59%). The difference by race was much less (9%) among those who did not experience toxicity. The White and Asian population was estimated to have one haplotype block composed of two 11-SNP haplotypes and a second block spanning 8 SNPs composed of 3 haplotypes with frequency greater than 1%. Two haplotypes in each block showed an approximately 1.6 fold odds of grade 3/4 toxicity when compared with all other haplotypes ($p=0.03$). In the overall population, there was strong evidence of haplotype association with clinical neurotoxicity for the least frequent haplotype in both the first ($p<0.005$) and second ($p<0.007$) block.

FANCD2 SNP association with gene expression

A total of 4 SNPs in the intronic regions of *FANCD2* were tested for association with gene expression levels of *FANCD2*. The first three markers are in strong LD ($r^2>0.99$). Individuals with at least one copy of the minor allele for all three SNPs have decreased *FANCD2* expression and significantly different mean expression across genotypes.

DISCUSSION

In this study, we found that patients who harbor SNPs and haplotypes in *FANCD2* experienced nearly a twofold increase in odds of grade 3 and 4 neurotoxicity compared to patients with common, wildtype haplotypes. There were no associations with SNPs in *BRCA1*, nor were there combined effects of *FANCD2* and *BRCA1* genotypes (data not shown). Although numbers of black patients were low, the frequency of ‘at risk’ alleles was more common and associations with risk of toxicity appeared to be stronger among them

than among white/other patients. Functional analyses to examine associations between SNPs in *FANCD2* and levels of gene expression showed that the ‘at risk’ alleles were associated with lower gene expression. Thus, individuals with minor alleles/haplotype may have greater odds of toxicity because *FANCD2* is under-expressed, conferring insufficient activity in repairing normal neuronal or myelin sheath cells after chemotherapy-induced DNA damage.

In addition to observing associations between SNPs in *FANCD2* and grades 3 and 4 toxicity according to the CTCAE reporting, we also noted relationships between these SNPs and self-reported neurotoxicities. A unique feature of this study was the ability to assess the association between these SNPs and the patients’ experience of toxicity as measured by the FACT-TAX. Using these data we found similar associations to those using CTCAE reports, adding to the validity of the results. They also demonstrate what could be interpreted as a dose-response effect. CTCAE grade 3/4 toxicity was more strongly associated with genetic variants in *FANCD2*. In contrast, weaker, although still statistically significant, associations were observed for lower extremes of adverse side effects.

It is well known that paclitaxel induces microtubule stabilization (MTS) and thereby mitotic arrest. However, emerging evidence supports a potential role of DNA damage following mitotic arrest in dictating taxanes’ cytotoxic effects. In cultured peripheral blood lymphocytes from healthy donors, paclitaxel treatment caused significant single and double DNA strand breaks quantified by alkaline comet assay, which was almost completely repaired after removal of the drug (9). Similar findings were reported in cultured human cancer cells, as DNA breaks accumulated following mitotic arrest triggered by paclitaxel treatment (10). These breaks activate a DNA damage response, which recruits DNA repair proteins and initiates DNA repair. Moreover, global gene expression analyses in cell lines in response to MTS agents, such as paclitaxel, revealed that chromosome instability determines the MTS response (11). Failure to initiate efficient mismatch repair or homologous recombination may contribute to MTS cytotoxicity. Thus, acquired DNA damage following mitotic arrest may be one of the mechanisms responsible for taxane-caused cell death.

We investigated the novel Fanconi anemia (FA)/BRCA pathway because of its central role in DNA damage response and mobilization of DNA repair machinery (12–14). *FANCD2* and *BRCA1* co-localize to the damaged chromatin foci and may act as a platform for assembly of DNA repair machinery through crosstalk with a range of DNA repair molecules (22). Thus, the FA/BRCA pathway may be predictive for response to cancer chemotherapy. In support of this, the FA/BRCA pathway has been linked to chemotherapy resistance in patients with glioma (19), ovarian cancer (23) and multiple myeloma (24), and resistance to melphalan in multiple myeloma may be circumvented by targeting this pathway with bortezomib (25). Although there are no published studies investigating the roles of FA/BRCA pathway in MTS response, it is an important relationship to evaluate, considering the massive amount of DNA damage caused by MTS.

While the high degree of correlation among SNPs in the *FANCD2* gene makes it difficult to isolate the causal variant(s) without further deep genotyping or sequencing, our observation that multiple SNPs were also associated with expression levels of *FANCD2* serves to buttress the biological plausibility of the associations between these SNPs in *FANCD2* and risk of neurotoxicities. When evaluating haplotypes by race, the differing LD structure and higher frequency of the minor variant in the black population yielded combinations of genetic variants with very different odds of toxicity between races. The significant two-SNP haplotype, A-G, spanning rs7648104-rs7637888, appears to be a unique race-specific risk haplotype, which could help explain the higher incidence of clinical toxicity in individuals self-reporting as black (19%) than those self-reporting as white/Asian (9%). However, these

findings must be interpreted with caution because the sample size for black patients is relatively small. The different LD structure of *FANCD2* and the race specific haplotype association underscores the importance of determining subgroup specific genetic risk factors when considering genotoxic exposures

We are unaware of any other reports of the association between SNPs in key genes involved in FA/BRCA pathway and taxane-induced neuropathy in women undergoing treatment for breast cancer in an adequately sized and well-defined population. We observed strong and significant associations between both CTC and self-reported toxicity and genetic variants in the *FANCD2* gene. Furthermore, the “risk allele” of *FANCD2* gene is associated with decreased expression of *FANCD2*, supporting our hypothesis that functional genetic variations in *FANCD2*, a key gene in FA/BRCA pathway, could modify response to taxanes. Our results must be validated in future studies, and the genetic mechanisms of FA/BRCA pathway in taxane response further elucidated. Nevertheless, our findings suggest the genetic variations in *FANCD2* gene could be used as an indicator for taxane toxicity, as well as a potential target for preventive measures or alternative therapeutic approaches.

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Figure 1.

Trial schema and patient numbers

Abbreviations:

A: Doxorubici

C: Cyclophosphamid

T: Paclitaxe

G: Filgrasti

PEGG: Pegfilgrastim

*897 women were genotyped; 9 women were removed for quality control purposes

Figure 2.
Block structure for FANCD2 and associations between SNPs and toxicity

Table 1

S0221 clinical trial population characteristics *

Variable	N (%)
<u>Age</u>	
23–42	228 (26)
43–50	245 (28)
51–57	197 (22)
58–80	218 (24)
<u>Self-reported Race</u> [§]	
White	740 (83)
Black	67 (8)
Other	81 (9)
<u>Treatment Arm</u>	
1	230 (26)
2	248 (28)
3	209 (24)
4	201 (23)
<u>CTCAE Grade of Neuropathy</u>	
0–2	781 (88)
3	103 (12)
4	4 (< 1)
<u>Self-reported Neuropathy (FACT-TAXANE)</u>	
39	160 (33)
39–52	174 (36)
> 52	146 (30)

* characteristics of a subset (n=888) on whom genotyping was performed;

[§] Black includes all individuals reporting as Black only, white includes women reporting as White only and Other includes women reporting as Asian, Pacific Islander, Native American or as belonging to multiple race groups

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Table 2

Marker Statistics

Gene	Marker	Location (base pairs)	Alleles*	MAF [§]	Freq (Tox >=3)	Freq (Tox <3)	HWE <i>p</i> (Tox >=3)	HWE <i>p</i> (Tox <3)
FANCD2	rs3846177	10043572	A/C	0.16	0.22	0.15	0.04	1
	rs9881859	10047814	T/A	0.16	0.23	0.15	0.04	1
	rs7648104	10048311	A/C	0.16	0.23	0.15	0.04	0.88
	rs7637888	10055722	G/A	0.16	0.23	0.15	0.04	1
	rs9875081	10058935	A/G	0.16	0.22	0.15	0.06	1
	rs9879080	10059224	A/G	0.16	0.22	0.15	0.06	1
	rs3895942	10083745	G/C	0.16	0.22	0.15	0.06	1
	rs6786638	10093075	G/C	0.16	0.23	0.15	0.04	1
	rs6805869	10094917	G/A	0.16	0.22	0.15	0.06	0.56
	rs6442150	10097575	G/A	0.16	0.22	0.15	0.04	1
	rs9849434	10108710	A/G	0.16	0.22	0.15	0.06	0.88
	rs6807485	10109320	C/G	0.08	0.06	0.09	1	1
	rs1552244	10110577	G/A	0.16	0.22	0.15	0.04	0.88
	rs7610821	10111150	A/G	0.16	0.22	0.15	0.06	0.88
	rs2272125	10113069	C/A	0.16	0.22	0.15	0.06	0.88
	rs2272124	10113189	G/A	0.16	0.22	0.15	0.06	0.88
	rs2272123	10115115	A/G	0.16	0.22	0.15	0.06	0.88
	rs7647987	10115696	G/A	0.16	0.22	0.15	0.06	0.88
	rs3172417	10117949	A/G	0.38	0.33	0.39	0.64	0.94
	rs12152512	10118770	A/G	0.16	0.22	0.15	0.06	0.88
BRCA1	rs8176305 [¶]	38454890	G/A	0.08	0.1	0.07	0.26	0.33
	rs8070179	38456214	A/G	0.37	0.39	0.39	0.88	0.08
	rs8176257	38469731	A/C	0.23	0.19	0.24	0.45	0.38
	rs8176242	38471400	A/G	0.31	0.3	0.32	0.88	0.82
	rs8176237 [¶]	38472273	A/G	0.07	0.06	0.07	0.54	0.79
	rs3737559 [¶]	38487830	A/G	0.07	0.06	0.07	0.54	0.82

Gene	Marker	Location (base pairs)	Alleles*	MAF [§]	Freq (Tox >=3)	Freq (Tox <3)	HWE <i>p</i> (Tox >=3)	HWE <i>p</i> (Tox <3)
	rs8176161	38494916	A/C	0.32	0.3	0.32	0.87	0.75
	rs8176160	38495029	G/A	0.32	0.3	0.32	0.87	0.78
	rs16942	38497526	G/A	0.31	0.29	0.32	0.92	0.81
	rs4986850 [‡]	38498997	A/G	0.08	0.1	0.07	0.26	0.36
	rs1799950 [‡]	38500007	G/A	0.06	0.05	0.06	0.1	0.09
	rs799923	38505457	A/G	0.21	0.21	0.22	0.4	0.1
	rs8176109	38519302	G/A	0.32	0.3	0.32	0.94	0.74
	rs8176098	38521732	C/A	0.32	0.3	0.32	0.83	0.63
	rs3765640	38529773	G/A	0.32	0.3	0.32	0.87	0.82
	rs8176077	38529874	G/A	0.32	0.3	0.32	0.87	0.74
	rs799905	38530713	G/C	0.4	0.42	0.39	0.61	0.43

* first allele listed is the minor allele in EA and Asian population;

[§] in all populations;

[‡] Analyzed only in overall and White/Other population due to MAF <0.05

Table 3

Single SNP association of BRCA1 with grade 3/4 neurotoxicity

Marker	P-value	OR
rs8176305	0.48	1.56 (0.92,2.67)
rs8070179	0.78	1.05 (0.68,1.62)
rs8176257	0.44	0.72 (0.46,1.12)
rs8176242	0.80	0.97 (0.63,1.49)
rs8176237	0.90	0.90 (0.47,1.70)
rs3737559	0.80	0.91 (0.48,1.72)
rs8176161	0.70	0.95 (0.62,1.46)
rs8176160	0.70	0.96 (0.63,1.47)
rs16942	0.76	0.90 (0.59,1.37)
rs4986850	0.52	1.50 (0.89,2.60)
rs1799950	0.78	0.83 (0.4,1.71)
rs799923	0.80	0.97 (0.63,1.50)
rs8176109	0.95	0.94 (0.61,1.44)
rs8176098	0.70	0.97 (0.63,1.49)
rs3765640	0.60	0.96 (0.63,1.48)
rs8176077	0.79	0.94 (0.61,1.44)
rs799905	0.77	1.10 (0.69,1.66)

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Table 4

FANCD2 Haplotype analyses by population

Population	Markers	Haplotype	Overall Freq	Freq in Tox >=3	Freq in Tox <3	OR (95% CI)	Permutation p-value
Black	rs7648104-rs7637888	A-G	0.28	0.46	0.23	2.84 (1.2, 6.9)	0.03
White/Other*	rs3846177-rs9849434	A-T-A-G-A-A-G-G-G-A	0.15	0.22	0.15	1.6 (1.1 – 2.4)	0.03
	rs1552244-rs12152512	G-A-C-G-A-G-G-A					
All	rs3846177-rs9849434	A-T-A-G-A-A-G-G-G-A	0.16	0.24	0.15	1.8 (1.3, 2.5)	0.005
	rs1552244-rs12152512	G-A-C-G-A-G-G-A	0.17	0.24	0.16	1.7 (1.2, 2.4)	0.007

* Includes all individuals self-reporting as White and Other (table 1)