Clinical Cancer Research

Genome-Wide Association Study for Anthracycline-Induced Congestive Heart Failure

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Abstract

Purpose: Anthracycline-induced congestive heart failure (CHF) is a rare but serious toxicity associated with this commonly employed anticancer therapy. The ability to predict which patients might be at increased risk prior to exposure would be valuable to optimally counsel risk-to-benefit ratio for each patient. Herein, we present a genome-wide approach for biomarker discovery with two validation cohorts to predict CHF from adult patients planning to receive anthracycline.

Experimental Design: We performed a genome-wide association study in 3,431 patients from the randomized phase III adjuvant breast cancer trial E5103 to identify single nucleotide polymorphism (SNP) genotypes associated with an increased risk of anthracycline-induced CHF. We further attempted candidate validation in two independent phase III adjuvant trials, E1199 and BEATRICE.

Results: When evaluating for cardiologist-adjudicated CHF, 11 SNPs had a P value $<10^{-5}$, of which nine independent chromosomal regions were associated with increased risk. Validation of the top two SNPs in E1199 revealed one SNP *rs28714259* that demonstrated a borderline increased CHF risk ($P = 0.04$, OR = 1.9). *rs28714259* was subsequently tested in BEATRICE and was significantly associated with a decreased left ventricular ejection fraction ($P = 0.018$, OR = 4.2).

Conclusions: *rs28714259* represents a validated SNP that is associated with anthracycline-induced CHF in three independent, phase III adjuvant breast cancer clinical trials. *Clin Cancer Res; 23(1); 43–51. ©2016 AACR.*

Introduction

Anthracyclines are a widely used chemotherapeutic class. Despite substantial antitumor activity, anthracyclines are not without toxicity. Cardiotoxicity, including congestive heart failure (CHF), is a dose-limiting side effect (1). Patients that received an anthracycline were five times more likely to develop cardiac symptoms (2). The overall incidence of CHF in patients receiving a cumulative dose of 240 to 360 mg/m^2 of doxorubicin is 1.7% to 2.1% (3). The optimal definition of cardiac dysfunction is debatable, but the most common mode of evaluation includes assessment of left ventricular ejection fraction (LVEF). Unfortunately, even with expert interpretation, there is variability in its estimation (4).

Anthracycline-induced cardiotoxicity can occur in two distinct forms: acute toxicity that develops early and presents as arrhythmias or depressed LVEF, or chronic toxicity, which occurs years later. The mechanism of cardiotoxicity is likely related to oxidative stress, although not completely elucidated (5). Known risk factors for developing clinical cardiomyopathy include cumulative dose, age, history of cardiac conditions, hypertension, liver disease, and mediastinal radiotherapy (5, 6). Risk of a cardiac event is further increased when anthracyclines are combined with other therapies, including trastuzumab (7) and bevacizumab (8, 9).

The risk of serious, irreversible cardiac damage has spurred the implementation of competing non-anthracycline regimens to obviate this concern (10), although many patients still benefit from anthracyclines based on risk or biology. The severity of the side effect necessitates a means to identify high-risk patients. Germline genetic biomarkers for CHF have been studied, but their ability to reliably predict risk remains unclear, as populations studied have been heterogeneous and results inconsistent (11–22).

We report a comprehensive evaluation of a large phase III, adjuvant breast cancer trial, E5103 (9), to identify common single nucleotide polymorphisms (SNP) that are associated with the risk of anthracycline-induced CHF. We performed a genome-wide association study (GWAS), which assessed SNPs throughout the

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Translational Relevance

Anthracyclines are a widely used chemotherapeutic class but can cause life-threatening congestive heart failure (CHF). The risk of serious cardiac damage has spurred the implementation of competing non-anthracycline regimens, although many patients still benefit from anthracyclines based on risk or underlying biology of the tumor. Germline genetic biomarkers for CHF have been studied, but their ability to reliably predict risk remains unclear as populations studied have been underpowered and heterogeneous, with inconsistent results to date. We have identified a SNP rs28714259 that is associated with CHF risk. This SNP was identified through a GWAS from the randomized phase III adjuvant breast cancer trial E5103 and was subsequently validated in two independent phase III adjuvant breast cancer trials, E1199 and BEATRICE. rs28714259 is associated with increased risk of anthracycline-induced CHF and might be useful to guide appropriate usage of anthracyclines for patients with breast cancer in the curative setting.

genome. Furthermore, we tested our most promising SNPs in two independent, randomized phase III trials, E1199 (23) and BEATRICE (8), which used a similar anthracycline backbone.

Materials and Methods

GWAS to identify common variants in ECOG-5103

E5103 overview. E5103 (9) was a phase III adjuvant breast cancer trial that randomized 4,994 patients to doxorubicin (60 mg/m^2) and cyclophosphamide (AC) for four cycles, followed by 12 weeks of weekly paclitaxel (Arm A) or to the same chemotherapy with either concurrent bevacizumab (Arm B) or concurrent plus sequential bevacizumab (Arm C). Patients were allowed to receive AC every 2 or 3 weeks at the discretion of the treating physician, and patients from all arms were scheduled to receive the same cumulative dose of doxorubicin (Table 1). Analyses were performed across all arms to identify SNPs associated with CHF. This correlative study was approved by the Indiana University IRB.

E5103 genotyping and imputation. Because of the availability of germline DNA, genotyping was performed at two independent time points in nonoverlapping study subsets (Fig. 1). DNA from an initial 2,209 and an additional 1,222 patients was genotyped by Illumina Genotyping Services using the HumanOmni1-Quad (>1 million SNPs) and the HumanOmniExpress (730,525 SNPs) array, respectively. Both sample sets used the Illumina BeadChip microarray platform for genotyping and the Illumina GenomeStudio software for initial genotyping calls. Of note, SNPs on the HumanOmniExpress were a subset of those on the HumanOmni1-Quad. Those SNPs not on the HumanOmniExpress were imputed in the second sample subset (Supplementary Data).

E5103 case and control definitions for CHF. To be selected for inclusion in E5103, patients must have not had a history of clinically significant cardiovascular disease. Cardiovascular health was monitored at the start and during the trial by MUGA or echocardiograms. In addition, a cardiac symptoms assessment was performed 2 years postregistration. Cardiac events, including CHF, decrease in LVEF, acute coronary syndrome, supraventricular tachycardia, and myocardial dysfunction, were diagnosed by a cardiologist. Cases to be considered for this correlative study included individuals that had centrally reviewed, cardiologist-adjudicated CHF. Controls were strictly defined and included patients who did not experience CHF or other reported events of cardiotoxicity, including LVEF <50% or drop in LVEF $\geq$20% from baseline. All controls received the full intended dose of AC with no modification.

Statistical analysis. A case–control GWAS was performed in the genetically defined European American (EA) subsamples (Supplementary Material) to identify SNPs associated with the presence or absence of CHF. Age, menopausal status, experimental arm, tumor grade, body surface area (BSA), hypertension during therapy, and use of antihypertension medications at baseline or antihypertensive therapy added during treatment were considered as covariates in analyses. The P value threshold was <0.05 for inclusion of covariates in the regression model. SNPs available on the HumanOmni1-Quad array were used as the common basis for all statistical analyses. An additive model of SNP effect was used when testing all hypotheses. The case–control analysis was conducted using SNPTEST v2.4 (https://mathgen.stats.ox.ac.uk/genetics_software/snptest/snptest.html). SNP and sample quality control (QC) were performed as described previously (24) and outlined in Supplementary Material.

Validation in E1199

Overview of E1199. E1199 (23) was a phase III adjuvant breast cancer trial that randomized 5,052 patients with node-positive or high-risk, node-negative breast cancer to one of four treatment arms. First, all patients received doxorubicin (60 mg/m^2) and cyclophosphamide (AC) for four cycles, followed by paclitaxel every 3 weeks for four cycles, or paclitaxel weekly for 12 cycles, docetaxel every 3 weeks for four cycles, or docetaxel weekly for 12 cycles. Tumor-derived DNA (formalin-fixed paraffin-embedded) was available from 2,906 patients (Fig. 1).

Table 1. Summary of CHF definitions and events across the clinical trials

Trial	Definition of CHF	Cumulative dose of anthracycline	Frequency of events	Bevacizumab administered
E5103	Centrally reviewed, cardiologist-adjudicated (composite of symptoms and lowered LVEF)	All patients received 240 mg/m^2 of doxorubicin	2.0%	Two of the three study arms
E1199	Common toxicity criteria v. 2.0	All patients received 240 mg/m^2 of doxorubicin	Parent trial: 1.5% GWAS subgroup: 1.6%	No
BEATRICE	Parent trial: NYHA class III/IV CHF Current GWAS: LVEF < 45%	94.8% of patients received an anthracycline at various doses per physician's choice	Parent trial: 1.6% GWAS subgroup: 2.9%	One of the two study arms

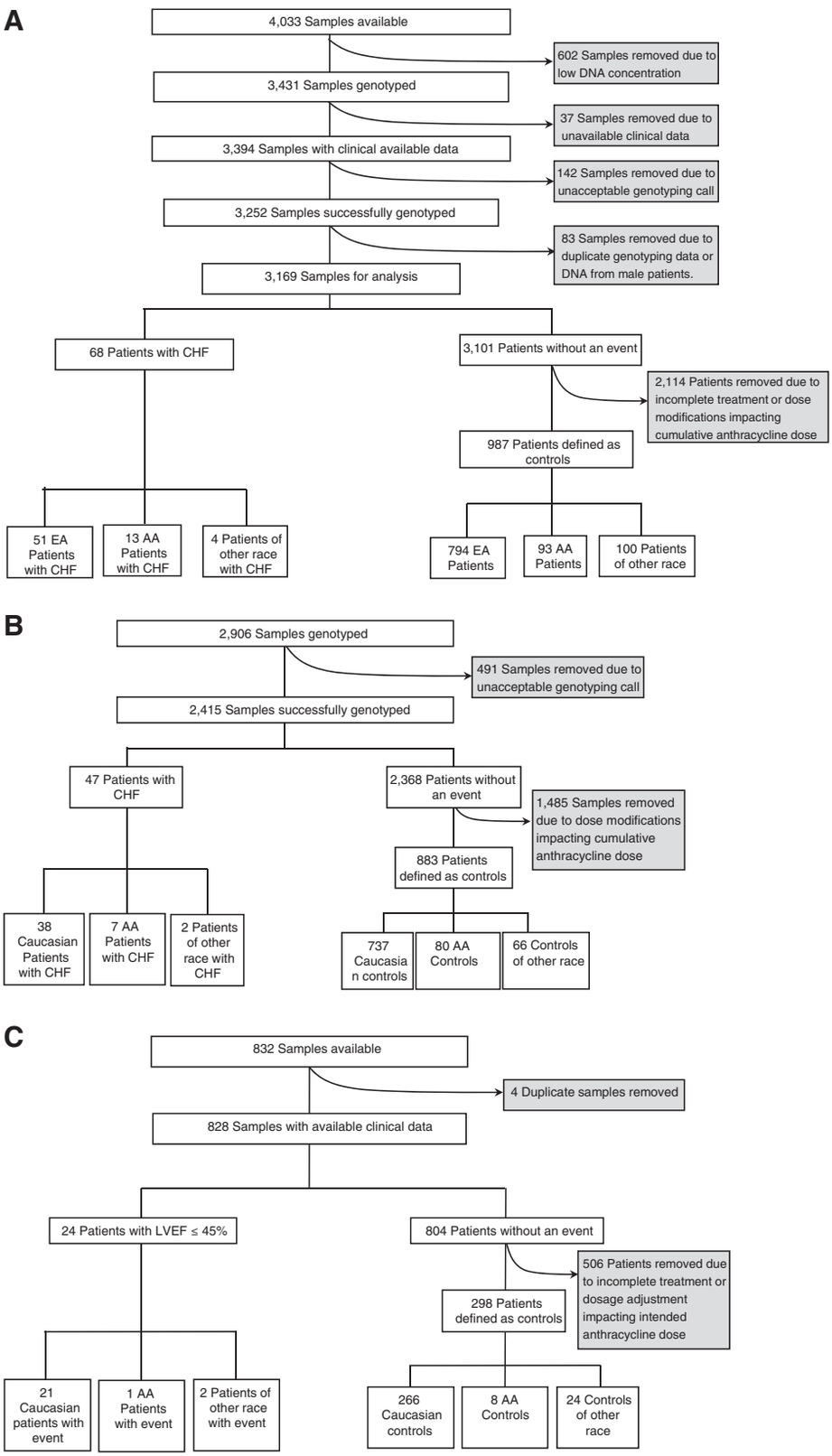

Figure 1.
Consort diagram. **A,** E5103 (the genome-wide discovery cohort); **B,** E1199 (first candidate SNP validation); **C,** BEATRICE (second candidate SNP validation). AA, African American.

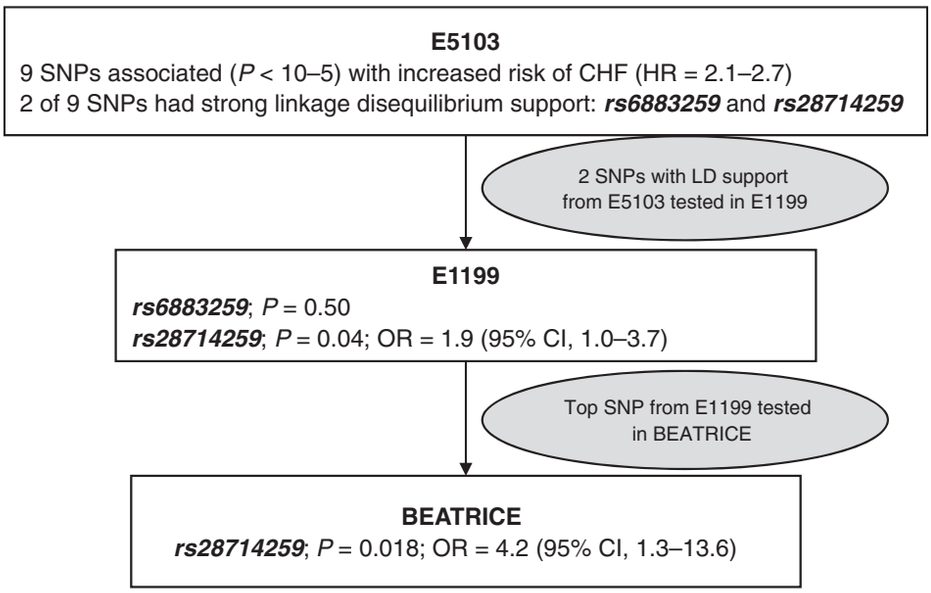

Figure 2.
Flow diagram for candidate SNP selection.

Candidate validation of top SNPS in E1199. All association results were reviewed and priority was given to two SNPs with further evidence of association from other top SNPs ($P < 10^{-5}$) in linkage disequilibrium (LD) to reduce the risk of false-positive signals from technical genotyping errors (Fig. 2). These two SNPs were genotyped using the QuantStudio 12K Flex OpenArray AccuFill System (Thermo Fisher Scientific) platform.

E1199 case and control definitions for CHF. To be included in E1199, patients must have not had a history of myocardial infarction, CHF, or significant ischemic or valvular heart disease. Cases included patients with reported CHF as defined by the Common Toxicity Criteria version 2.0. Raw LVEF data points were not available. Patients were considered controls if they had no reported cardiac events and received the full dose of intended AC with no dose modification.

Statistical analysis. Comparing cases and controls in the EA subpopulation, logistic regression analysis was performed to identify SNPs associated with CHF. Age, BSA, and treatment arm were considered as covariates. The *P* value threshold for significance was set at 0.025 after Bonferroni correction was made.

Validation in BEATRICE

Overview of BEATRICE. BEATRICE (8) was a phase III adjuvant breast cancer trial that randomized 2,591 patients to a minimum of four cycles of standard adjuvant chemotherapy (including an anthracycline, taxane, or both) at the investigator's discretion with or without bevacizumab for 1 year. Germline DNA was available from 828 patients (Fig. 1).

Candidate SNP validation in BEATRICE. The SNP with borderline association with CHF in the E1199 was further tested for association with decreased LVEF in BEATRICE (Fig. 2). The SNP was genotyped using a TaqMan SNP assay.

BEATRICE case and control definitions for CHF. To be included in BEATRICE, patients had a baseline LVEF >55%. In the parent trial, CHF was defined as New York Heart Association (NYHA) class III or IV CHF accompanied by a drop in LVEF >10% to below 50. There were only 16 cases of NYHA CHF or cardiac death across the parent trial and therefore insufficient cases in the cohort of genotyped patients. Thus, we enriched statistical power by including cases based on low LVEF. Prior to genetic analysis, we selected LVEF <45% to be considered an event. Because of inconsistency and inaccuracy of LVEF measurement, we did not select <50%, a commonly used criterion for mild dysfunction, to minimize false-positive events. We also chose 45% based on prior data suggesting that LVEF <45% was a strong contributor to future cardiovascular risk (25). Patients were considered controls if they had no reported cardiac events, had no hypertension, and received the full dose of intended anthracycline without dose modification.

Statistical analysis. Comparing cases and controls in the EA subpopulation, logistic regression analysis was performed to identify SNPs associated with decreased LVEF. Age, BSA, and treatment arm were considered as covariates. As only one statistical comparison was performed, statistical significance was set at $P < 0.05$.

Results

Phenotypes: rate of CHF and clinical predictors

E5103. Of 4,033 patients who had available DNA (Fig. 1), phenotype data were available only from 3,394 genotyped patients. In this subgroup, 2% experienced cardiologist-adjudicated CHF (Table 1). This included 68 cases: 51 EA, 13 African Americans (AA), and 4 of other race. There was no increased risk across the genetically determined racial groups (EA vs. AA vs. other; $P = 0.09$). Because of the insufficient number of non-EA cases, genetic association analysis was limited to the EA subgroup. Nongenetic demographic predictors were compared for the entire group as well as for EAs and AAs separately (Table 2). Older age was a significant risk factor, with a 43% relative increased risk with each decade of life ($P = 0.009$). There was an increased risk in Arm C ($P = 0.04$) but not in Arm B ($P = 0.87$) when compared with

Table 2. Comprehensive comparisons (*P* values) of nongenetic demographic predictors of CHF

	Samples with clinical data available					
	All		AA		EA	
Clinical trial	Age	BSA	Age	BSA	Age	BSA
E5103[a] (genotyped)	0.009	0.10	0.76	0.01	0.002	0.79
$N = 3,394$ (68 cases)						
AA = 386 (13 cases)						
EA = 2,525 (51 cases)						
Others = 484 (4 cases)						
E1199 (parent trial)	0.005	0.04	0.62	0.10	0.005	0.48
$N = 4,735$ (71 cases)						
AA = 401 (11 cases)						
EA = 4,042 (58 cases)						
Others = 292 (2 cases)						
E1199 (genotyped)	0.044	0.07	0.50	0.07	0.08	0.61
$N = 2,906$ (47 cases)						
AA = 267(38 cases)						
EA = 2,616 (7 cases)						
Others = 23 (2 cases)						
BEATRICE[a] (genotyped)	0.774	0.91	0.99	0.99	0.87	0.72
$N = 828$ (24 cases)						
AA = 31 (1 case)						
EA = 705 (21 cases)						
Others = 92 (2 cases)						

[a]Clinical data was only available for those patients who had companion DNA samples.

Arm A. There was an increased risk for patients who were on antihypertensives at baseline ($P = 7.2 \times 10^{-4}$), but not for those patients who experienced therapy-induced hypertension. BSA was not associated with an increased risk in the entire cohort ($P = 0.10$) or in EA patients ($P = 0.79$) but was associated with increased risk in AA patients ($P = 0.01$).

E1199. Phenotypic data were available from a majority of patients in the parent trial ($n = 4,735$) as well as the entire genotyped group ($n = 2,906$; Fig. 1). The risk of CHF was 1.5% in the parent trial and 1.6% in the genotyped group (Table 1). In the genotyped group, there were 47 cases of CHF, 38 among self-defined EA and 7 cases in AA and 2 cases among other races. There was no significant increase in risk based on race (EA vs. AA vs. other; $P = 0.38$). Again, genetic association analyses were limited to the EA subgroup due to an insufficient number of non-EA cases. Nongenetic demographic predictors were compared for the entire group as well as for EAs and AAs separately (Table 2). Older age demonstrated a 39% relative increased risk with each decade of life in the parent population ($P = 0.005$). BSA was not associated with risk in the EA population ($P = 0.61$) but a trend for increased risk in the AA population ($P = 0.07$). Neither hypertension nor use of antihypertensive data was available in this trial.

BEATRICE. Phenotypic data were available only from the 828 genotyped patients (Fig. 1). In this subgroup of patients, 2.9% experienced a decline in LVEF <45%. This value is higher than the 1.6% of population reported to experience NYHA class III or IV CHF in the parent trial (Table 1; ref. 8). The subgroup included a total of 24 cases, 21 among self-defined EA and the remaining 3 were of other race. There was no increased risk of CHF across the self-defined racial groups (EA vs. other; $P = 0.45$). Because of insufficient numbers of cases, the genetic association analyses were limited to the EA subgroup. Nongenetic demographic predictors were compared for the entire group as well as for EAs and AAs separately (Table 2). Older age ($P = 0.77$) and increased BSA ($P = 0.91$) were not associated with increased likelihood of decreased LVEF. There was also no increased risk for those who experienced significant therapy-induced hypertension ($P = 0.98$).

GWAS results for CHF in E5103

After genotyping and QC, 810,907 SNPs were used for the GWAS in E5103. The strength of association between genotype and CHF (Fig. 3) is demonstrated in Table 2. A total of 112 SNPs had $P < 1 \times 10^{-4}$, of which 99 were associated with increased risk of CHF (OR = 1.4–3.7; Supplementary Table S1). Nine independent chromosomal regions represented by 11 top SNPs were associated with the risk of CHF ($P < 10^{-5}$), and in two of the nine regions, there are multiple SNPs that are in LD with top SNPs and also provide evidence of association; therefore, the likelihood of a false-positive association is substantially reduced. One SNP from each of these two regions was selected for validation in independent datasets (Fig. 2). SNPs previously reported to be associated with anthracycline-induced CHF did not provide evidence of association in this study (Table 3).

Evaluation of candidate SNPs in E1199 and BEATRICE

One of the two SNPs genotyped in the E1199, rs28714259, provided evidence of borderline association with CHF ($P = 0.041$, OR = 1.9), whereas the other, rs6883259, was not ($P = 0.50$). *rs28714259* was subsequently genotyped in BEATRICE and was significantly associated with decreased LVEF <45% ($P = 0.018$, OR = 4.2; Fig. 2).

Discussion

In the current study, we aimed to identify a population at risk for developing CHF from anthracyclines using the dose and schedule commonly employed for adult solid tumors. We performed a GWAS in a large randomized phase III adjuvant trial. As a follow-up, we performed limited genotyping in two large independent phase III adjuvant trials. In total, the three large trials enrolled more than 12,500 breast cancer patients. All patients in E5103 and E1199 received a uniformed dose of doxorubicin (60 mg/m^2) and more than 95% patients in BEATRICE received an anthracycline. We provide evidence that the SNP *rs28714259* is associated with cardiac dysfunction across trials using slightly different phenotype definitions. This SNP was among the top SNPs associated with increased risk of CHF (OR > 1) and also had LD support from another top SNP in our discovery sample set of E5103 trial. In all the three trials, the A allele was associated with an increased risk of cardiac damage (E5103: OR = 2.1; $P = 9.25 \times 10^{-6}$; E1199: OR = 1.9; $P = 0.04$; and BEATRICE: OR = 4.2; $P = 0.02$).

Age was the most consistent demographic predictor for increased risk, as it was associated in two of the three trials evaluated here. There was also a correlation between the use of antihypertensive agents at baseline in E5103, but an increased risk was not seen for those who only had treatment-induced hypertension. This would support the notion that underlying and potentially long term, cardiovascular disease increased the risk of this toxicity. Unfortunately, details of comorbidities were not available for further evaluation. There was also a signal that BSA may be an important contributor but was only detected in the AA patients.

Previous studies have evaluated germline predictors. This included evaluation of candidate SNPs or gene panels in the

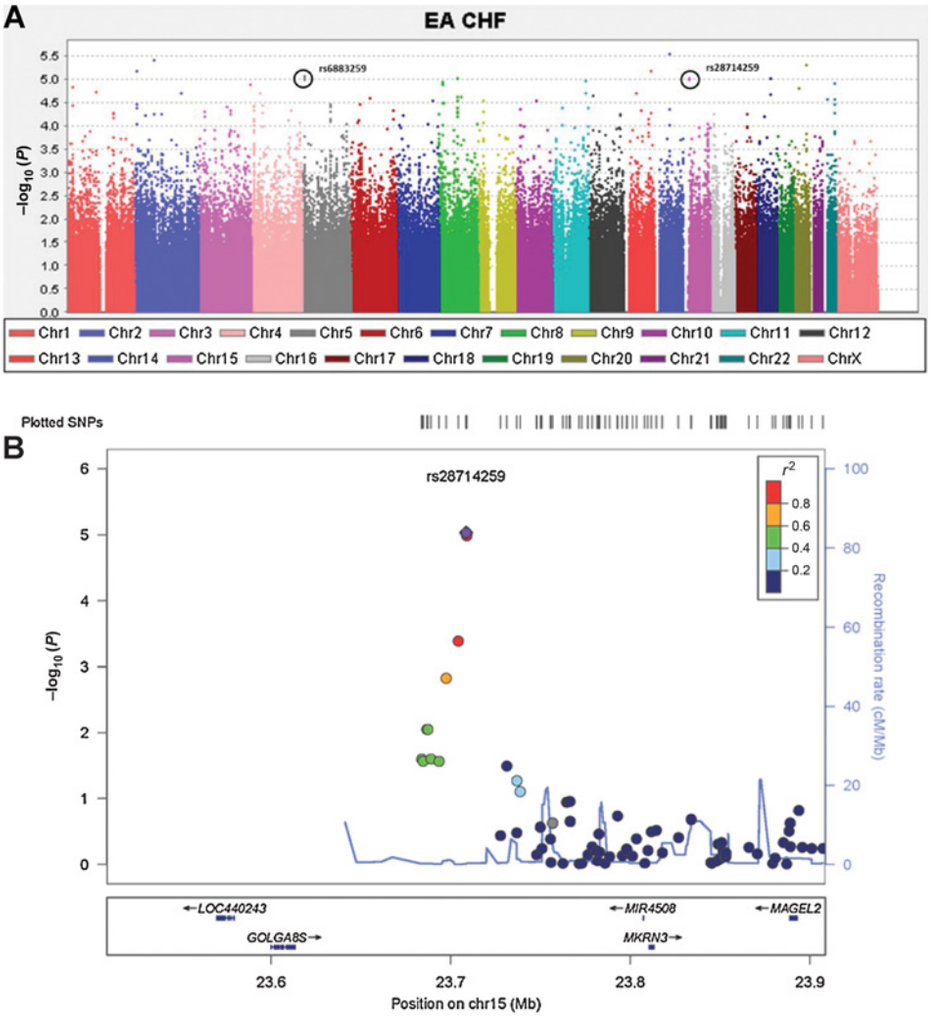

Figure 3.
GWAS of anthracycline-induced CHF in EA cohort in E5103. **A,** Manhattan plot for centrally reviewed, cardiologist-adjudicated CHF. *x-axis*, chromosomal position of each SNP analyzed; *y-axis*, magnitude of the evidence for association, show as $-\log_{10}$ (*P* value). SNPs genotyped in the two independent samples are circled; **B,** *P* values ($-\log_{10}$) of SNPs near *rs28714259* from SNP-associated analysis with CHF. SNPs are color to reflect their LD with *rs28714259*. EA, European American.

metabolism and transport pathway or in the proposed mechanistic pathways of anthracycline-induced cardiomyopathy (12–22). Several of these studies have demonstrated associations, but none of these candidates were identified in the recent GWAS by Aminkeng and colleagues (11) or in our study (Table 3). Aminkeng and colleagues focused on a pediatric population of EAs with 34 cases of decreased LVEF and 248 controls (11). The study identified a SNP in *RARG* (OR = 4.7) that was confirmed in two other independent cohorts. Our current study found the opposite effect (decreased rather than increased risk) for the *RARG* SNP ($P = 4.1 \times 10^{-6}$; OR = 0.11). There are multiple explanations for the incongruence of findings across studies. One likely reason is due to the general heterogeneity in studies, including:drug type, drug exposure, phenotype definition, and population. With regard to the latter, many of the prior studies to date have focused on the pediatric patient population.

This study adheres to stringent criteria for biomarker discovery, and the correlative study has several strengths as outlined (26). First, the discovery cohort was conducted in the context of a randomized, phase III adjuvant trial, using a standard number of doses and cycles of doxorubicin. The phenotype collection mandated close follow-up of cardiac function with serial echocardiography and events defined by cardiologist adjudication based on both imaging and clinical picture. The genomic evaluation implemented a comprehensive genome-wide approach, which allowed for unbiased discovery. The top finding was further supported in two independent trials that had the same (E1199) and similar (BEATRICE) doses and schedules of anthracycline.

There were also several weaknesses. First, two of the three trials included an experimental arm that delivered bevacizumab. Thus, an association with cardiac toxicity due to the combination of an anthracycline plus bevacizumab cannot be excluded. This concern is substantially minimized as we adjusted for arm of study for both bevacizumab-containing trials, E5103 and BEATRICE, and the third trial, E1199, only included anthracycline, with the SNP being associated with an increased risk and similar effect size across all three trials. Second, the DNA from E1199 was derived from tumor DNA, and thus, point mutations at the SNP site of interest and loss of heterozygosity cannot be excluded. This limitation was minimized by assuring meticulous QC of genotyping and that all analyzed SNPs did not violate HWE. Finally, we used slightly different definitions of cardiac dysfunction across the three trials. E5103 utilized a clinically useful definition of CHF as defined by a centrally reviewed cardiologist adjudication and considered a composite of both symptoms and depressed LVEF. E1199 utilized the CTC criteria for CHF, and BEATRICE employed

Table 3. Selected SNPs previously reported to be associated with CHF

Study	Number of patients (sample size)	Study set	Dose and schedule of anthracyclines	Phenotype	Genotype	Genes of top association	Validation with number of independent cohort
Wojnowski et al. (22)	450	DSHNHL-B1/B2	Doxorubicin/uniform dose & schedule	WHO grade 3 and 4 cardiotoxicity	Candidate SNPs in 82 genes in anthracycline pharmacodynamics	*NADPH, BCC1, ABCC2*	No
Blanco et al. (13)	145	Retrospective pediatric cohort	Any anthracycline/ variable dose & schedule	Self-reported symptoms	Candidate genes *NQO1* and *CBR3*	*CBR3*	No
Rajić et al. (16)	76	Retrospective pediatric cohort	Any anthracycline/ variable dose & schedule	Cardiologist-defined cardiac damage	Candidate SNPs in *SOD2, CAT, GSTT1, GSTM1*	*CAT*	No
Blanco et al. (14)	487	COG-ALTE03N1	Any anthracycline/ variable dose & schedule	AHA guidelines	Candidate SNPs in *CBR3* and *CBR1*	*CBR3*	No
Semsei et al. (18)	235	Retrospective pediatric cohort	Doxorubicin & daunorubicin/ uniform dose & schedule	Cardiac function measured by LVFS	Candidate SNPs in *ABCC1*	*ABCC1*	No
Visscher et al. (20)	156	Retrospective pediatric cohort	Doxorubicin & daunorubicin/ variable dose & schedule	CTCAE v3	Candidate SNPs in 220 genes in anthracycline pharmacodynamics	*CBR3, CB4, HMNT, ABCC1, SLC28A3*	Yes- two cohorts
Volkan-Salanci et al. (21)	70	Prospective adult cohort	Doxorubicin/variable dose & schedule	Various measures of cardiac function	Candidate SNPs in *CBR3* and *GSTP1*	*CBR3, STP1*	Yes
Armenia et al. (12)	255	Retrospective adult cohort	Any anthracycline/ variable dose & schedule	ACC/AHA guidelines	Candidate genes in anthracycline metabolism and pharmacodynamics	*NADPH, HFE, ABCC2*	No
Lubieniecka et al. (15)	91	Retrospective adult cohort	Daunorubicin/variable dose & schedule	Acute cardiotoxicity	60 candidate genes in anthracycline metabolism and efflux	*POR*	No
Wang et al. (17)	287	COG-ALTE03N1	Any anthracycline/ variable dose & schedule	AHA guidelines	2,100 candidate genes on ITMAT/Broad CARe cardiovascular SNP array	*HAS3*	Yes
Visscher et al. (19)	344	Retrospective pediatric cohort	Doxorubicin & daunorubicin/ variable dose & schedule	CTCAE v3	Candidate 4,578 SNPs in drug pharmacokinetic and toxicity genes	*SLC22A7, SLC22A17*	Yes
Aminkeng et al. (11)	280	Retrospective pediatric cohort	Doxorubicin & daunorubicin/ variable dose & schedule	CTCAE v3	GWAS	*RARG*	Yes- two cohorts
Schneider et al. (current study)	102	ECOG-5103	Doxorubicin/uniform dose & schedule	Cardiologist adjudicated	GWAS	rs28714259 in intergenic region of chr 15	Yes- two cohorts

Abbreviations: ACC, American College of Cardiology; AHA, American Heart Association; COG, Children's Oncology Group; CTCAE, Common Terminology Criteria for Adverse Events; DSHNHL, German High-Grade Non-Hodgkin's Lymphoma Study Group; ECOG, Eastern Cooperative Oncology Group; LVFS, left ventricular fractional shortening; WHO, World Health Organization.

the NYHA criteria to define cardiac damage. BEATRICE also collected raw LVEF values to further refine cases and controls, whereas E1199 did not. Despite the heterogeneity of phenotype, the similar results across the three trials strengthen the clinical generalizability of this genetic predictor.

Our top SNP, *rs28714259*, lies in an intergenic region in chromosome 15 and had LD support from nearby SNPs (Fig. 3B). There is increasing evidence that intergenic regions play an important role in gene expression and regulation through long-range interactions (27). Chromatin immunoprecipitation data from the ENCODE project revealed that *rs28714259* is within the binding site for glucocorticoid receptor protein (28). Recent studies have demonstrated glucocorticoid signaling plays important roles in the structural and functional maturation of the fetal heart and in the maintenance of proper cardiac function in various animal models (29).

Future confirmatory studies might focus on even less severe phenotypes of cardiac damage, as prior data demonstrate that asymptomatic CHF has marked increase 10-year mortality (30). Rare toxicities, however, might be best explained by rare variants with large effect size, and these are not evaluated on current GWAS platforms. Thus, additional work evaluating the role of rare

variants and mechanistic work is warranted and ongoing. The ability to better understand which patients might be at higher risk for this serious toxicity is an important step toward personalizing therapy and providing better care for our patients.

Disclosure of Potential Conflicts of Interest

D.A. Cameron is a consultant/advisory board member for Roche. No potential conflicts of interest were disclosed by the other authors.

Disclaimer

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