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Article

Large-scale meta–genome-wide association study reveals common genetic factors linked to radiation-induced acute toxicities across cancer types

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Abstract

Background: This study was designed to identify common genetic susceptibility and shared genetic variants associated with acute radiation-induced toxicity across 4 cancer types (prostate, head and neck, breast, and lung).

Methods: A genome-wide association study meta-analysis was performed using 19 cohorts totaling 12 042 patients. Acute standardized total average toxicity ($STAT_{acute}$) was modelled using a generalized linear regression model for additive effect of genetic variants, adjusted for demographic and clinical covariates ($rSTAT_{acute}$). Linkage disequilibrium score regression estimated shared single-nucleotide variation (SNV—formerly SNP)–based heritability of $rSTAT_{acute}$ in all patients and for each cancer type.

Results: Shared SNV-based heritability of $STAT_{acute}$ among all cancer types was estimated at 10% (SE = 0.02) and was higher for prostate (17%, SE = 0.07), head and neck (27%, SE = 0.09), and breast (16%, SE = 0.09) cancers. We identified 130 suggestive associated SNVs with $rSTAT_{acute}$ ($5.0 \times 10^{-8} < P < 1.0 \times 10^{-5}$) across 25 genomic regions. rs142667902 showed the strongest association (effect allele A; effect size –0.17; $P = 1.7 \times 10^{-7}$), which is located near *DPPA4*, encoding a protein involved in pluripotency in stem cells, which are essential for repair of radiation-induced tissue injury. Gene-set enrichment analysis identified 'RNA splicing via endonucleolytic cleavage and ligation' ($P = 5.1 \times 10^{-6}$, $P = .079$ corrected) as the top gene set associated with $rSTAT_{acute}$ among all patients. In silico gene expression analysis showed that the genes associated with $rSTAT_{acute}$ were statistically significantly up-regulated in skin (not sun exposed P = .004 corrected; sun exposed $P = .026$ corrected).

Conclusions: There is shared SNV-based heritability for acute radiation-induced toxicity across and within individual cancer sites. Future meta–genome-wide association studies among large radiation therapy patient cohorts are worthwhile to identify the common causal variants for acute radiotoxicity across cancer types.

Approximately 50% of patients with cancer undergo radiation therapy (RT) (1). Some of these patients experience radiation-induced toxicities in nearby normal tissues that can occur during or shortly after RT (ie, acute radiation-induced toxicities) (2) or months to years later (ie, late radiation-induced toxicities). Although historical understanding of radiobiology separated tissues into classes defined as early and late responding, the contemporary view is that most if not all tissues have both an acute and late phase, but the biologic mechanisms underlying early and late injury may differ. Next to radiation dose distributions, individual variability in radiation-induced toxicity (3) occurs in part because of host factors, including comorbidities, body habitus, and underlying genetic susceptibility (4). Rare pathogenic variants in DNA damage genes (eg, *ATM*) result in monogenic syndromes with high risk of radiation-induced toxicities, but these do not explain the variation among most patients. Genome-wide association studies (GWASs) have identified common genetic susceptibility variants (eg, single-nucleotide variations [SNVs—formerly SNPs]) for late radiation-induced toxicities, but few studies have focused on acute radiation-induced toxicities (5-14).

Although some SNVs affect toxicity risk in a tissue-specific manner, others may be common across multiple tissues, with relative contributions differing for acute and late radiation-induced toxicity (15). Prior GWASs of late radiation-induced toxicities suggest that genetic susceptibility depends on molecular mechanisms specific to a given tissue type (5), but given that acute reactions across tissues depend on DNA damage and cell death, we hypothesized that there is shared susceptibility to acute radiation-induced toxicity across tissues. Therefore, we

aimed to carry out a GWAS meta-analysis of radiation-induced toxicities across tissues among 19 cohorts totaling 12 042 patients representing 4 cancers (prostate, head and neck, breast, and lung) from the Radiogenomic Consortium (https://epi.grants.cancer.gov/radiogenomics/). The large study population also enabled us to estimate for the first time SNV-based heritability (ie, the proportion of variation in acute radiation-induced toxicity explained by common genetic variants).

Methods

Study participants

The study design was a retrospective analysis of prospectively collected cohorts totaling 12 042 patients who presented with cancer of the prostate, head and neck, breast, or lung. Patients received RT (alone or as part of a combination regimen) with curative intent and were followed for the development of acute toxicity (Table 1; Supplementary Table 1, available online). Each study obtained ethical approval from local review boards, and all participants provided written informed consent. Supplementary Tables 2 through 5 (available online) summarize the characteristics of the cohorts; a description is provided in the Supplementary Methods (available online).

Assessment of acute radiation-induced toxicity

The primary endpoint was acute radiation-induced toxicity, adjusted for demographic and clinical covariates. The grading schema and assessment schedules are given in Supplementary Table 1 (available online). To achieve a composite score describing overall acute radiation-induced toxicity, we used the standardized total average toxicity (STAT) method described previously (16) and in the Supplementary Methods (available online). STAT is a scale- and grade-independent measure of radiation-induced toxicity developed to facilitate meta-analysis and data pooling.

Acute standardized total average toxicity ($STAT_{acute}$) was calculated using toxicity assessments collected within 90 days from the start of RT, which is a commonly used definition for acute radiation-induced toxicity in cooperative group trials (eg, Radiation Therapy Oncology Group). When more than 1 assessment was available within this time frame, the worst score was used. The individual toxicity endpoints used for calculating $STAT_{acute}$ within each cancer type (Supplementary Tables 6-9, available online) reflect the different organs at risk within the treatment field. Patients with high baseline toxicity such that the grade could not worsen were excluded (Supplementary Table 10, available online).

Genotyping, quality control, and imputation

Whole blood or buffy coat was stored at –80 °C; then, DNA was isolated using standard procedures (ie, silica membrane spin columns) and genotyped using genome-wide SNV arrays as part of a prior GWAS according to Supplementary Table 11 (available online). Germline DNA sequences are (near) constant across tissues; thus the SNVs present in blood cells will be identical to those in all cells of the normal tissues susceptible to acute radiation-induced toxicity. The following standard preimputation quality control filters were removed: SNVs with a low call rate and low maximum allele frequency (MAF) and that did not meet Hardy-Weinberg equilibrium as well as samples with discordant sex, higher-than-expected pairwise identity by descent, and non-European ancestry determined by principal components analysis or multidimensional scaling. SNVs were imputed on the Michigan Imputation Server using the Haplotype Reference Consortium, release 1.1 2016 reference panel or IMPUTE2 software and the 1000 Genome Phase 3 European reference panel. Postimputation filters removed SNVs with low MAF (<1%) or imputation quality (information metric < 0.3). Supplementary Table 11 (available online) summarizes the quality control steps and number of SNVs imputed for each cohort.

Covariables

Demographic and clinical factors are listed in Supplementary Tables 12 through 15 (available online). A backward stepwise selection procedure with a conservative $P = .2$ and $STAT_{acute}$ as the dependent variable was used to identify the most influential covariates within the breast, prostate, and lung cancer cohorts. In head and neck cancer cohorts, a predefined list of covariates (Supplementary Table 13, available online) was used. Residuals from the final adjusted multivariable linear model defined $rSTAT_{acute}$, our primary endpoint, in each cohort. $STAT_{acute}$, which is unadjusted for demographic and clinical covariates, was our secondary endpoint.

Table 1. Characteristics of study cohorts[a]

	Cohort	No.	Age, median (range), y	Female, %	$STAT_{acute}$, mean (SD)
Prostate	RAPPER-CHHiP	1487	68 (50-84)	N/A	0.000 (0.782)
n = 4213	RAPPER-RT01	219	66 (50-79)	N/A	0.000 (0.779)
	RADIOGEN-PrCa	647	72 (47-86)	N/A	0.000 (0.448)
	GenePARE	225	65 (47-82)	N/A	–0.007 (0.681)
	CCI-EBRT	151	68 (45-82)	N/A	0.000 (0.301)
	REQUITE-PrCa	1348	70 (46-88)	N/A	0.004 (0.494)
	UGhent-PrCa	136	65 (49-79)	N/A	0.001 (0.419)
Head and neck cancer	UMCG-HANS	1279	65 (19-93)	32.7	0.005 (0.781)
n = 4042	DAHANCA	1183	60 (27-90)	21.0	–0.003 (0.802)
	Ghent-HNC	273	60 (31-87)	16.5	–0.014 (0.821)
	RAPPER-HNC	187	61 (39-85)	7.6	0.078 (0.779)
	NIMRAD	270	73 (44-87)	21.0	–0.008 (0.719)
	Head and Neck 5000	672	60 (25-94)	22	0.098 (0.881)
	RADIOGEN-HNC	178	63 (35-92)	11.8	0.000 (0.588)
Breast	RAPPER-breast	907	59 (26-83)	100	0.006 (0.993)
n = 2966	REQUITE-breast	2059	58 (23-90)	100	0.007 (0.786)
Lung	RADIOGEN-Lung	154	65 (41-89)	13.6	–0.006 (0.546)
n = 821	REQUITE-Lung	467	70 (39-91)	31.0	0.002 (0.545)
	CONVERT	200	64 (29-82)	48.0	–0.015 (0.524)

[a] STAT = standardized total average toxicity; N/A = not applicable.

GWAS analysis

SNV associations with $rSTAT_{acute}$ and $STAT_{acute}$ were independently analyzed in each cohort by linear regression, assuming an additive model that includes the first 10 principal components to control for population stratification. SNVs were represented by the number of effect alleles or imputed genotype dosage. Statistical analyses were carried out using PLINK/1.90b3.44 (17) and SNPTEST (18), and GWAS results were checked using the GWASinspector (19) package in R (R Foundation for Statistical Computing, Vienna, Austria).

Meta-analysis

GWAS results were meta-analyzed across all cohorts and separately by cancer type by 2 independent centers using the inverse variance-weighted, fixed-effects method as implemented in PLINK/1.90b3.44 (17) and METAL, version 2011-03-25 (20). The Cochran Q test for heterogeneity was performed. SNVs were considered significant genome-wide if they had a $P < 5.0 \times 10^{-8}$ for association with outcome, heterogeneity $P > .05$, and were available in at least 50% of samples. All tests of statistical significance were 2-sided.

Linkage disequilibrium score regression, SNV-based heritability, and genetic correlation

Linkage disequilibrium (LD) score regression (21) used summary statistics (~1.1 million SNVs; χ^2 statistics from the GWAS meta-analysis) on the LD scores across the genome. An LD score regression intercept close to 1 suggests no confounding bias, whereas an inflated intercept (>1) may indicate population stratification, confounding, or model misspecifications. We filtered the included variants to the subset included in HapMap3 and excluded variants with duplicated rs-numbers, ambiguity, and MAF < 0.01. We used the default European LD score file based on the European 1000 genome reference panel. Cross-trait LD score regression estimated genetic correlation (22) for acute radiation-induced toxicities between the 4 cancer types in 1-by-1 comparisons. The slope of the regression estimated the genetic covariance between 2 radiation-induced toxicity endpoints.

Gene set and in silico tissue expression analysis

MAGMA (23) gene set association analysis was implemented in FUMA (24). The gene-wide *P* value was calculated by combining the *P* value of all SNVs inside genes after accounting for LD and outliers. We allowed for a window of 10 kilobase pairs upstream and downstream of each gene to capture SNVs in nearby regulatory regions. Next, we calculated competitive gene set *P* values on the gene-wide *P* value after accounting for gene size, gene set density, and LD between genes. We defined a gene set as statistically significant if its joint *P* value was below the threshold corresponding to a false discovery rate < 0.05.

In silico tissue expression analysis was based on the MAGMA gene property in FUMA. The normalized gene expressions (reads per kb per million) of 53 normal tissue types were obtained from Genotype-Tissue Expression, version 8 (25). To obtain differentially expressed gene sets for 53 tissue types, we used the normalized expression (zero mean of log2 (reads per kilobase pair per million + 1)). Two-sided *t* tests were performed per gene per tissue compared with all other tissues. Genes with Bonferroni $P < .05$ adjusted and absolute log-fold change ≥0.58 were defined as a differentially expressed gene set in a given tissue.

Results

Patient characteristics

Table 1 and Supplementary Tables 2 through 5 (available online) summarize the patient and clinical characteristics of the cohorts and the treatments received. Figure 1 shows the combined distribution of $STAT_{acute}$ and $rSTAT_{acute}$ scores for the 4 cancer types, and Supplementary Figure 1 shows the distributions for each participating cohort. Supplementary Tables 12 through 15 (available online) list the covariates used in statistical analyses to derive $rSTAT_{acute}$. Table 1 and Supplementary Figure 1 (available online) describe the distribution of $STAT_{acute}$ and $rSTAT_{acute}$ per cohort.

Meta-GWAS of acute radiation-induced toxicity

The additive effect of more than 6 million SNVs on $rSTAT_{acute}$ (n = 10 398) and $STAT_{acute}$ (n = 11 115) was estimated. The quantile-quantile plots showed no genomic inflation, suggesting that population structure was adequately controlled using 10 principal components (PCs) and included only individuals of European ancestry (Figure 2). No SNV reached genome-wide significance, but 130 SNVs with a $5.0 \times 10^{-8} < P < 1.0 \times 10^{-5}$ spanning 25 genomic regions had a suggestive association with $rSTAT_{acute}$. The strongest association, with an effect size of −0.174 ($P = 1.7 \times 10^{-7}$) per copy of the A allele was for rs142667902. The nearest gene to this SNV is *DPPA4* (Figure 3), which encodes a protein involved in the maintenance of pluripotency in stem cells. From association analysis with $STAT_{acute}$, the number of suggestive SNVs decreased to 66 across 27 genomic regions, with rs113548225 displaying the strongest association, at an effect size of 0.157 ($P = 2.2 \times 10^{-7}$) per copy of the A allele. Supplementary Tables 16 and 17 (available online) contain the suggestively associated SNVs and Supplementary Figure 2 (available online) displays Manhattan and quintile-quintile plots.

We found no genome-wide significant SNVs associated with $rSTAT_{acute}$ or $STAT_{acute}$ for the individual cancer sites. The suggestive findings are summarized in Supplementary Tables 18 through 25 (available online) and Supplementary Figures 2 and 3 (available online).

SNV-based heritability and genetic correlation

The LD score regression intercept close to 1 for all regression models (Table 2) confirmed negligible inflation attributable to relatedness and that observed associations were due to the polygenic architecture of ration-induced toxicities. SNV-based heritability (SE) estimates for $rSTAT_{acute}$ were 12% (0.07%) for prostate cancer, 16% (0.09%) for breast cancer, and 15% (0.09%) for head and neck cancer. The joint estimated SNV-based heritability (SE) for $rSTAT_{acute}$ was 7% (0.09%). SNV-based heritability (SE) for $STAT_{acute}$ was estimated as 17% (0.07%) for prostate cancer, 27% (0.09%) for head and neck cancer, and 16% (0.09%) for breast cancer. The joint SNV-based heritability (SE) for $STAT_{acute}$ was 10% (0.02%). SNV-based heritability estimates for $STAT_{acute}$ and $rSTAT_{acute}$ in lung cancer were imprecise because of small sample size (SE ≤ 0.40), precluding statistical inference. A 1-to-1 cross-cancer type joint analysis of both $rSTAT_{acute}$ and $STAT_{acute}$ showed no statistically significant genetic correlations between pairs of cancer types (Supplementary Table 26, available online).

Gene set analysis

The gene set *P* value was computed using the gene-based *P* value for 4728 curated gene sets (including canonical pathways) and 6166 gene ontology terms obtained from MsigDB, version 5.2. We

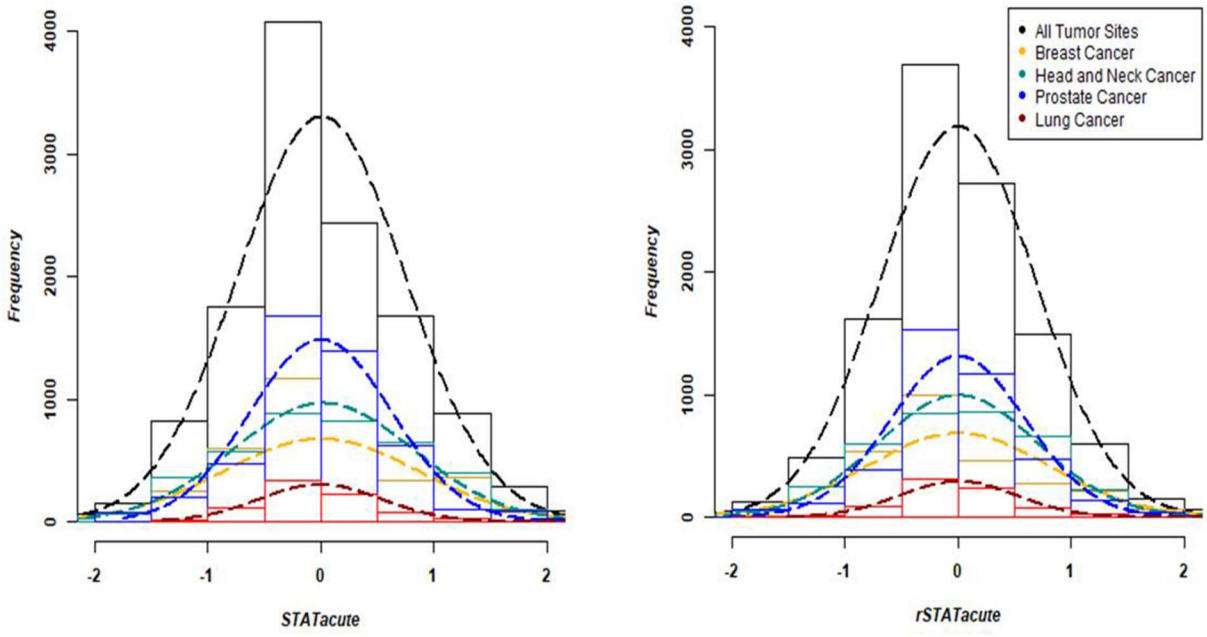

Figure 1. Histograms of $STAT_{acute}$ and $rSTAT_{acute}$ distribution per cancer type and curve of normal log distribution. STAT = standardized total average toxicity.

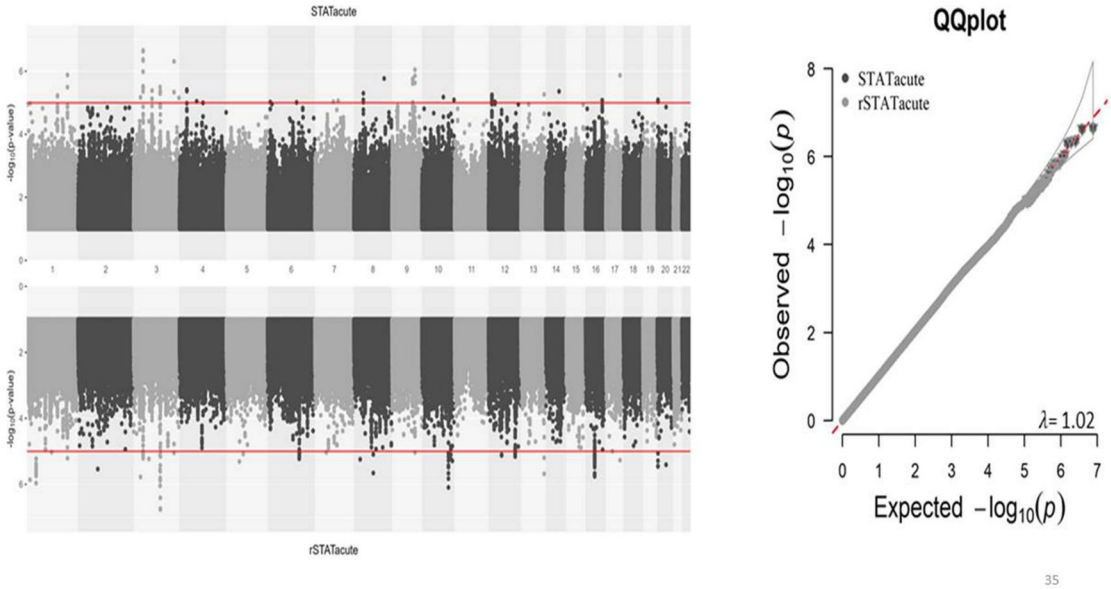

Figure 2. Manhattan (**left**) and Q-Q (**right**) plots of the overall meta-analysis for $STAT_{acute}$ and $rSTAT_{acute}$. Mirror. Manhattan plot: The GWAS for $STAT_{acute}$ and $rSTAT_{acute}$ are displayed in the top and bottom panels, respectively. The x-axis represents genomic locations, while the y-axis indicates $-\log_{10}$ *P* values for SNV associations with the outcome. Each SNV is a dot. Q-Q plot: Observed $-\log_{10}$ *P* values are on the y-axis, and expected $-\log_{10}$ *P* values are on the x-axis. Every SNV is represented as a **dot**, with the **red line** signifying the null hypothesis of no genuine association. Notable deviations from the expected *P* value distribution appear primarily at the tail, complemented by the λ coefficients, indicating effective control of population stratification. GWAS = genome-wide association study; Q-Q = quintile-quintile; SNV = single-nucleotide variant; STAT = standardized total average toxicity.

used Ensembl gene models for 19 079 genes and a Bonferroni-corrected *P* value threshold of 2.6×10^{-6}. MAGMA identified *protein glycosylation in Golgi* as statistically significantly associated with $rSTAT_{acute}$ in head and neck cancer ($P = 2.4 \times 10^{-6}$, $P = .037$ corrected). The next top-ranking pathway was *RNA splicing via endonucleolytic cleavage and ligation* ($P = 5.1 \times 10^{-6}$, $P = .079$ corrected, overall $rSTAT_{acute}$). Detailed results of the top 10 gene sets per cancer type are shown in Supplementary Tables 27 through 31 (available online).

In silico tissue expression analysis

The genes related to overall $rSTAT_{acute}$ reached statistically significant up-regulated expression in skin not sun exposed ($P = 7.2 \times 10^{-5}$, $P = .004$ corrected) and skin sun exposed

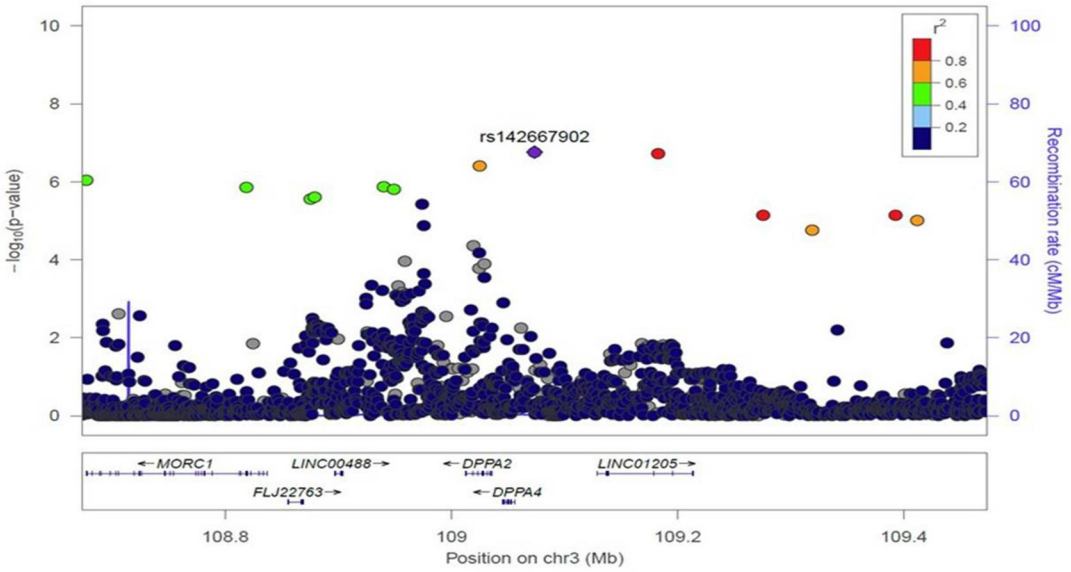

Figure 3. Locus zoom plot for the top locus associated with $rSTAT_{acute}$. The **purple diamond** shows the top single-nucleotide variant and variants in **red** are in linkage disequilibrium with the top single-nucleotide variant. The y-axis shows observed $-\log_{10} P$ values, and the x-axis shows the position across the genome, with genes mapped there. STAT = standardized total average toxicity

Table 2. Single-nucleotide variant–based heritability of $STAT_{acute}$ and $rSTAT_{acute}$ overall and per cancer type[a]

		Minimum, No.	Single-nucleotide variants, No.	λ Genomic control	Mean χ^2	Intercept (SE)	h2 (SE)
$STAT_{acute}$	Overall	7410	1 087 229	1.022	1.022	1.013 (0.006)	0.102 (0.026)
	Prostate cancer	2681	1 152 958	1.007	1.013	1.020 (0.006)	0.171 (0.069)
	Breast cancer	1890	1 067 908	1.004	1.009	1.007 (0.006)	0.161 (0.096)
	Head and neck cancer	2292	1 148 120	1.019	1.018	1.005 (0.005)	0.268 (0.088)
	Lung cancer	547	709 968	1.011	1.013	1.019 (0.007)	0.831 (0.399)
$rSTAT_{acute}$	Overall	6739	1 079 330	1.007	1.014	1.012 (0.006)	0.070 (0.028)
	Prostate cancer	2389	1 152 836	1.005	1.009	1.016 (0.006)	0.125 (0.076)
	Breast cancer	1786	1 067 908	1.007	1.009	1.012 (0.006)	0.158 (0.097)
	Head and neck cancer	2256	1 140 063	1.013	1.009	1.005 (0.006)	0.152 (0.091)
	Lung cancer	500	709 969	1.005	1.008	1.023 (0.007)	0.526 (0.424)

[a]h2 = single-nucleotide variation–based heritability; intercept = protects against bias from population stratification and cryptic relatedness; STAT = standardized total average toxicity.

($P = 4.8 \times 10^{-4}$, $P = .026$ corrected) tissues (Figure 4). No tissue reached a significant P value in the individual cancer types, but the genes associated with acute toxicity in patients with breast and lung cancer had maximum expression in their corresponding tissues (breast mammary and lung tissues); for those with head and neck cancer, esophagus mucosa ranked as the second-most expressed tissue (Supplementary Figure 4, available online).

Discussion

We identified 130 suggestive SNVs underlying shared genetic susceptibility to acute radiation-induced toxicity and showed that acute radiation-induced toxicity is likely to have a moderate SNV-based heritability of 10%. Higher heritability estimates within cancer types confirmed that the genetic susceptibility of acute radiation-induced toxicity is partially tissue type specific. Gene set analysis identified pathways not previously associated with acute radiation-induced toxicities that should be explored functionally and as potential targets for interventions to reduce radiation injury.

The top SNV associated with $rSTAT_{acute}$ was rs142667902, nearest to *DPPA4*, which encodes a nuclear factor involved in the maintenance of pluripotency in stem cells (26). The pathogenesis of acute radiation-induced toxicity involves the turnover and transit time for pluripotent stem cells to repopulate damaged tissue; thus, a role of *DPPA4* in radiation-induced toxicity is plausible. Gene set analysis identified *RNA splicing via endonucleolytic cleavage and ligation* associated with acute radiation-induced toxicity. Exposure to ionizing radiation can disrupt the coupling of RNA splicing with gene transcription involved in DNA repair, cell-cycle control, and apoptosis. This emerging trend sheds light on the complex cellular response to DNA damage (27). Interestingly, gene expression analysis estimated statistically significantly up-regulated expression in skin not sun exposed and sun exposed for genes related to overall $rSTAT_{acute}$. A simple interpretation is that skin is the shared organ at risk for all cancer types affected acutely by RT. In line with Fessé et al. (28), our results suggest that skin and damage to the skin resulting from sun exposure (nonionizing radiation) may be interesting to explore further for the understanding the mechanism involved in the response of tissues to DNA damage.

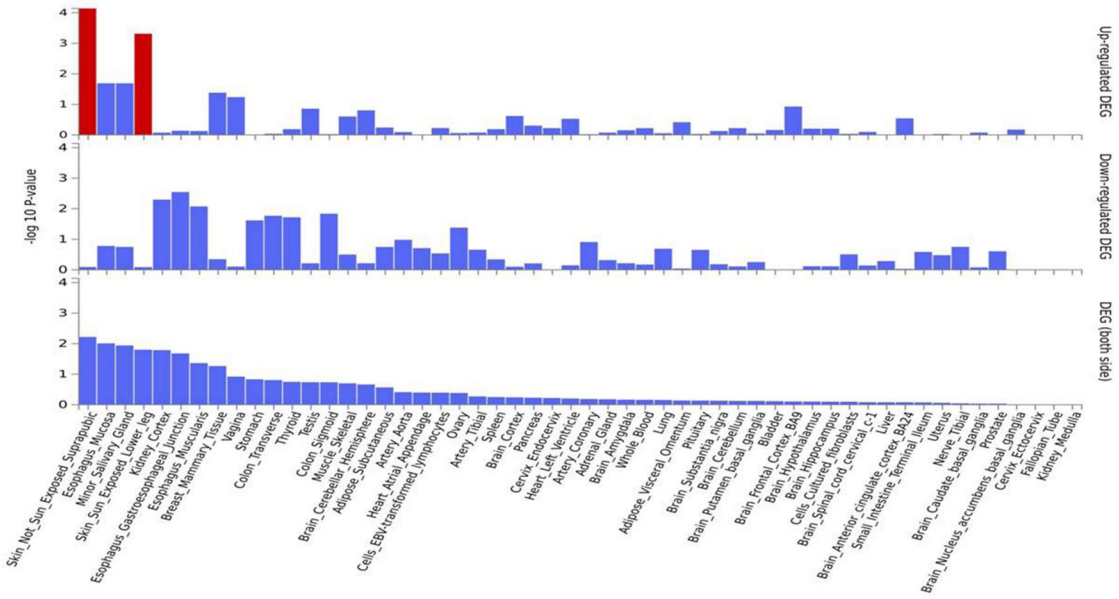

Figure 4. Tissue expression analysis in 53 tissue types for genes related to overall $rSTAT_{acute}$. Tissue expression analysis testing the positive relationship between all annotated genes using the full distribution of single-nucleotide variant *P* values and the average expression of genes per tissue type based on Genotype-Tissue Expression RNA sequencing data. DEG = differentially expressed gene set; STAT = standardized total average toxicity.

We found 95 suggestive SNVs located in 21 genomic regions associated with acute toxicity in patients with prostate cancer. The top SNV (rs72954279) was near to a pseudogene *OACYLP*. A transancestry meta-GWAS identified rs35283980 mapped to *OACYLP* associated with susceptibility to prostate cancer (29), though no studies have been published investigating a role in normal tissue radiation-induced toxicity. The top gene set in prostate cancer was "adrenergic receptor activity." Adrenergic receptors are found throughout the body in many cell types. The bladder is particularly rich in these receptors, which are functionally important regulators of the activities of muscles. Pharmacomechanical and molecular approaches have revealed roles for the β(3)-adrenoceptor in the urinary bladder and gastrointestinal tract smooth muscle, both organs susceptible to acute radiation-induced toxicity during prostate cancer treatment (30). Pullikuth et al. showed that adrenergic receptor signaling regulates tumor response to ionizing radiation (31), and our finding suggests that it would be worthwhile to investigate a role in normal tissue responses. Given that activity of the receptor would affect multiple tissue types, it is a good candidate for further study.

There were 83 suggestive SNVs in 21 genomic regions associated with acute radiation-induced toxicity in patients with head and neck cancer. The top SNV, rs137992872, mapped to *TCF20*, encoding a widely expressed transcriptional co-regulator. Our analyses suggested that *protein glycosylation in Golgi* is a potential mechanism involved in susceptibility to acute radiation-induced toxicity in head and neck cancer. Approximately half of all proteins undergo glycosylation, and this modification has a substantial impact on diverse cellular processes in all tissue types. Published studies linked up-regulation of glycosylation genes with radioresistance (32,33). Inhibition of glycosylation has also been shown to enhance sensitivity to cisplatin (a DNA damaging agent) in head and neck cancer cells (34). Toth et al (32) found that plasma protein glycosylation changes in response to partial body irradiation, and the effects last during follow-up.

Of 26 SNVs in 14 genomic regions possibly associated with acute radiation-induced toxicity among breast cancer cohorts, the top SNV was rs16882722, mapped near the tumor suppressor *UNC5D*, a netrin receptor involved in apoptosis (33). Moelans et al. observed an association between *DUSP26* and *UNC5D* loss and chemo-RT resistance, which predicted worse survival in patients with breast cancer (35). The top gene set associated with radiation-induced toxicity in breast cancer was *natural killer cell lectin-like receptor binding*. Natural killer cells are innate immune cells that can respond to inflammatory signals such as interferons and interleukins present at the site of normal tissue injury; they can potentiate vascular damage (36,37), and our findings suggest that it would be worthwhile to investigate their role in the pathogenesis of radiation-induced toxicity in breast cancer.

We found 30 SNVs in 10 genomic regions suggestive of an association with acute radiation-induced toxicity in patients with lung cancer. The nearest gene to the top SNV (rs1471101) was *MLLT3*. Ayako et al. found a joint effect of the *MLLT1* and *MLLT3* genes on the ATM-signaling pathway and a role in repressing genotoxic stresses because of DNA double-strand breaks and maintaining genome integrity (38). Furthermore, our analysis showed the highest expression of genes in lung tissue among all 53 tissues tested and that the top gene set in radiation-induced toxicity in lung cancer was *Debiasi apoptosis by reovirus infection dn*. Our findings suggest that comparing the mechanisms of reovirus-induced apoptosis with radiation-induced apoptosis could identify similarities in tissue damage pathogenesis.

Our observations highlight the complexity of radiation-induced toxicity and suggest new avenues to increase understanding of the pathogenesis of acute radiation-induced toxicity in a tissue-specific manner. The bioinformatic analyses can point to potential mechanisms but should be used for hypothesis generating and must be followed up with subsequent functional validation studies. Validation studies and subsequent functional characterization of radiation-induced toxicity–associated SNVs in cell lines and animal models will be important next steps to

understand the molecular mechanisms involved and, potentially, identify targetable pathways for intervention.

Our first estimate of shared SNV-based heritability of acute radiation-induced toxicity across 4 cancer types was 10%. The estimates were higher for prostate (17%), breast (16%), and head and neck (27%) cancers. These SNV-based heritability estimates are comparable with those for complex traits such as coronary artery disease (5%) (39), autism spectrum disorder (12%) (40), and schizophrenia (26%) (41). SNV-based heritability estimates depend on study size; thus, our estimates will improve with larger studies (42). Also, narrow-sense heritability used here misses heritability because of rare variants with large effects that are not tagged by common SNVs and to nonadditive genetic variation or epigenetic factors (43). Therefore, it is likely that the level of heritability of acute radiation-induced toxicity will be higher than that reported in our study.

No SNV achieved the stringent threshold for genome-wide significance, which is a challenge in GWAS (44). The rigorous correction for many statistical tests reduces false positives but may mask real associations. A second limitation is the lack of ancestral diversity in our cohorts because of limited statistical power to perform a multiethnic GWAS; this limits the generalizability of our findings to non-European and admixed populations. Future studies should be conducted on extended sample sizes, with particular effort devoted to building cohorts in non-European patient populations and more precisely defining phenotypes for radiation-induced toxicities. Although we examined common SNVs with a MAF greater than 1%, investigating the rare variants would provide significant insights.

Many common variants are potentially associated with acute radiation-induced toxicities across tumor sites, and it is worthwhile to carry out larger studies that have the statistical power to identify the causal variants. Our large meta-GWAS provides the first evidence for the heritability of common genetic variants associated with acute radiation-induced toxicity, which is higher within than across tissues. Further investigation to verify and expand our findings is merited to identify multiple genome-wide significant loci, with pooled clinically relevant effect sizes that can be used in clinical practice.

Data availability

This study was done using cohorts involved in the Radiogenomics Consortium. Summary statistics for GWAS results will be available to download from the GWAS Catalog.

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Conflicts of interest

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