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Whole-exome sequencing of 79 xenografts as a potential approach for the identification of genetic variants associated with sensitivity to cytotoxic anticancer drugs

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Abstract

Chemotherapy response remains unpredictable in most patients with cancer. In this study, we performed whole-exome sequencing of 79 cancer xenografts derived from human cancer tissues to identify genetic predictors of chemosensitivity to nine cytotoxic anticancer drugs. Xenografts were harvested from 12 organs with cancer and implanted into nude mice. The mice were exposed to one of nine cytotoxic anticancer drugs (5-fluorouracil, nimustine, adriamycin, cyclophosphamide, cisplatin, mitomycin C, methotrexate, vincristine, and vinblastine) to assess the correlation between chemosensitivity response and variant allele frequency. We found 162 candidate variants that were possibly associated with chemosensitivity to one or more of the nine anticancer drugs (P < 0.01). In a subgroup analysis of breast and gastric cancer xenografts, 78 and 67 variants, respectively, were possibly associated with chemosensitivity. This approach may help to contribute to the development of personalized treatments that may allow for the prescription of optimal chemotherapy regimens among patients with cancer.

Introduction

Cancer is a global health concern, with approximately 18.1 million new cases and 9.6 million deaths in 2018 [1]. Currently, most cancers are treated by surgery, radiation therapy, and/or chemotherapy [2,3]. Chemotherapy remains a gold standard for the treatment of blood cancers, such as leukemia and lymphoma [4–6], unresectable or metastatic cancers [7,8], and solid tumors, such as lung, breast and colorectal cancers [9–11]. Despite an improved understanding of cancer biology and the development of molecular targeted therapy and immunotherapy for select patients [12,13], chemotherapy still plays a primary role in cancer treatment regimes.

Competing interests: The authors have declared that no competing interests exist.

Indeed, chemotherapy regimens have improved considerably, now taking into consideration the organ of origin, histological appearance, and stage of progression. Yet, these improvements aside, chemotherapeutic efficacy still varies between individuals [14] and is often complicated by toxic reactions, including nausea, tiredness, diarrhea, and hair loss [14,15], causing physical and mental distress and decreased patient quality of life. As such, it is becoming increasingly important to identify effective treatments with fewer toxic side effects as a first-line therapy for each patient. Several recent studies have sought to establish diagnostic methods for predicting chemosensitivity to cytotoxic anticancer drugs before treatment is undertaken. However, clinically useful genetic markers have yet to be developed [16–20].

Patient-derived xenograft (PDX) models have been established for many types of tumors and have emerged as powerful tools for predicting drug efficacy and for understanding tumor characteristics. With PDX models, fresh human tissue is directly implanted into immunocompromised mice. These models retain the heterogeneity of the original patient tumors and thus allow for tests, predominantly to examine the efficiency of anticancer drugs [21].

Next-generation sequencing technologies have also been developed in recent years, exposing tumor genomic profiles and facilitating the detection of low frequency variants and other genetic mutations that could not otherwise be uncovered by conventional methods [22,23]. Indeed, several studies have reported associations between such genetic mutations in tumors and clinical outcomes [24–26]. Guided by these reports, we hypothesized that genetic variants within tumors, including low frequency, rare variants, may underpin patient responses to cytotoxic anticancer drugs, such as chemosensitivity and chemoresistance. To this end, we performed whole-exome sequencing of DNA samples taken from 79 human cancer xenografts prepared from 12 different organs. These xenografts were implanted in mice and treated with one of nine cytotoxic anticancer drugs. We assessed correlations between the chemosensitivities of the xenografts to nine anticancer drugs and the variant allele frequencies (VAFs) as a potential approach to identify variants that may be predictive of drug response.

Materials and methods

Animals and tumor xenograft model

Previously, a total of 79 human tumor tissues were obtained aseptically during surgery or at autopsy across 13 hospitals in Japan [27]. The samples included 12 breast cancers, 12 gastric cancers, 10 neuroblastomas, 10 non-small-cell lung cancers, 7 gliomas, 6 pancreatic cancers, 5 colon cancers, 5 choriocarcinomas, 4 small-cell lung cancers, 4 hematopoietic cancers, 3 ovarian cancers, and 1 osteosarcoma. These xenografts were separately transplanted into athymic BALB/c-nu/nu mice (Clea Japan, Inc, Tokyo, Japan) and maintained by serial subcutaneous transplantation of 2×2×2 mm fragments into the flank once a month, as described previously [27]. Microbiological monitoring of the tumor-bearing nude mice was performed for bacteria (e.g., Pasteurella pneumotropica and Mycoplasma pulmonis), viruses (e.g., mouse adenovirus and mouse hepatitis virus), and parasites (e.g., Giardia muris and Spironucleus muris) by culture, serological, or microscopic examinations [27]. Furthermore, histological examination and isozyme testing were carried out to assess for the risk of cross-contamination among the tumor lines, or cross-contamination between human tumor xenografts and a mouse tumor appearing at the inoculation site of the xenograft during passaging [27]. Tumor-bearing mice were euthanized with deep anesthesia followed by cervical dislocation. To minimize discomfort, euthanasia was performed quickly. Tumors were excised from the euthanized mice, and a piece of the tumor tissue was implanted into another mouse using a transplantation needle. All handling of mice was carried out in a gentle to minimize animal suffering and distress. The general conditions of the mice such as appetite and respiratory conditions were monitored

every 2 or 3 days after transplantation, and the size of the tumor was measured twice a week. Mice were housed in a controlled temperature of 23 ± 1 °C and relative humidity 50–70%, with ad libitum access to food and water. All animal experiments were performed in accordance with the guidelines of the Central Institute for Experimental Animals.

Anticancer drugs

The chemosensitivity tests on the xenograft model in this study were performed more than 20 years ago [27]. We chose nine cytotoxic anticancer drugs that could be classified into different categories based on their mechanism of action: 5-fluorouracil (5FU; Sigma-Aldrich; Merck KGaA, Darmstadt, Germany), nimustine (ACNU; Daiichi Sankyo Co., Ltd., Tokyo, Japan), adriamycin (ADR; Kyowa Hakko Bio Co., Ltd., Tokyo, Japan), cyclophosphamide (CPM; Shionogi & Co., Ltd., Osaka, Japan), cisplatin (DDP; Sigma-Aldrich), mitomycin (MMC; Kyowa Hakko Bio), methotrexate (MTX; Wyeth Lederle Japan, Ltd., Tokyo, Japan), vincristine (VCR; Shionogi & Co), and vinblastine (VLB; Shionogi & Co). These drugs have been used as a standard of care for cancer for over 20 years, and some of these drugs remain a standard of care. All of the drugs were dissolved in sterile 0.85% NaCl containing 1% mannitol (Wako Pure Chemical Industries, Ltd., Osaka, Japan).

Chemosensitivity analysis

A total of 7,900 mice were purchased from Japan CLEA Inc. (Tokyo, Japan) and used in this study. Each anticancer drug was administered individually at the maximum tolerated dose (MTD) to nude mice bearing human cancer xenografts (n = 6 mice per group), because this dose could clearly distinguish responders from non-responders for each drug. The MTD for each drug was as described previously [27]: 6.7 mg/kg MMC, 260 mg/kg CPM, 48 mg/kg ACNU, 10 mg/kg DDP, 12 mg/kg ADR, 1.6 mg/kg VCR, 11 mg/kg VLB, 19 mg/kg 5-FU, 15 mg/kg MTX. 5-FU and MTX were administered once a day for 5 days whereas all other drugs were administered once. The control groups did not receive any treatment (6 mice per xenograft). Each of the 79 xenografts was treated with nine drugs over the course of the experiment, with 6 mice bearing xenografts used to test each drug. Two to four drugs were tested as part of a single cohort for each xenograft; along with 6 control mice, this equated to 18 to 30 mice at one time. In addition, 4 spare mice for each drug were prepared. This meant that up to 50 mice were used for each cohort. Mice were sacrificed by cervical dislocation at 21 days after administration of the drug or when the tumor volume reached 250 mm³ (humane endpoint criteria). The next cohort of mice were then acquired, and the same protocols were followed for housing and treatment. For a single xenograft, it took about 1.5 months to test each of the 9 drugs. Given that there were 79 xenografts in total, this part of our experimental procedures was carried out over approximately 10 years.

Chemosensitivity was calculated as the relative tumor volume in the treated mice (T) compared with the control (C) using the mean values measured on day 14, as described previously (T/C [%]) [27]. Tumor volume (mm³) was calculated using the following formula: $0.5 \times$ major diameter \times minor diameter².

Ethics statement

All animal studies were approved by the Institutional Committee of Central Institute for Experimental Animals, and carried out as per published protocols [27]. The establishment of PDX models and chemosensitivity testing of these xenografts were performed between 1981 and 1991. These studies were performed before enforcement of the Ethical Guidelines for Human Genome/Gene Analysis Research in Japan. Therefore, acquisition of agreement of

patients for the use of their tumor was not obligated at the time. Furthermore, these xenografts are publicly available resources, and chemosensitivity data of them were published in 1996 [27]. Therefore approval from ethics committee is not necessary for this study.

Sample preparation and whole-exome sequencing

Tumor genomic DNA was extracted from 79 xenografts using the QIAmp DNA Mini kit (QIAGEN, Hilden, Germany), according to the manufacturer's protocol and as previously described [28]. Exome enrichment and library preparation were performed using Ion Ampi-Seq Exome RDY Kit PI v3, which targets >97% of consensus coding sequences (CCDS) with 5-bp padding around exons, and Ion Xpress Barcode Adapters (Thermo Fisher Scientific, Inc.). Pooled barcoded libraries were subsequently conjugated with sequencing beads by emulsion PCR and enriched using the Ion PI Hi-Q Chef kit and Ion Chef (Thermo Fisher Scientific, Inc.). Sequencing of templates was performed with 2 samples per Ion PI Chip V3 using the Ion Proton system (Thermo Fisher Scientific, Inc.), according to the manufacturer's protocols.

Variant calling

To avoid false-positive results, we removed reads derived from the mouse genome, as follows. Sequencing reads were aligned to the human genome build 19 (hg19) and two mouse genomes C57BL/6J (mm10, NCBI accession number: GCA 000001635.26) and BALB/c (GCA_001632525.1) using the Torrent Mapping Alignment Program (ver 3.0.1, Thermo Fisher Scientific, Inc.). Reads aligned to the mouse genomes with higher alignment score than to the human genome were considered to be contamination from the host mouse and were removed from subsequent analyses. The Torrent Variant Caller plugin (ver 5.10.1, Thermo Fisher Scientific, Inc.) was used to identify variants. The parameter file, optimized for somatic mutations with low stringency criteria, was obtained from the software vendor. Variants were annotated by ANNOVAR (ver. 2018-04-16) [29] using the following reference databases: RefSeq Gene (refGene); LJB non-synonymous variants annotation (dbnsfp35a); dbSNP version 150 (avsn150); the 1000 Genome Project (1000g2015aug_eas); Clinvar version 20190305 (clinvar_20190305); COSMIC Release v88 (cosmic88); segmental duplication region (genomicSuperDups); transcription factor binding site (tfbsConsSites); Human Genetic Variation Database version 2.3 [30]; 3.5K Japanese individuals allele frequency panel (3.5KJPNv2) [31]. Variants were filtered and excluded if they: (i) had a quality score < 30; (ii) had segmental duplication or repeat regions identified by Repeat Masker or Tandem Repeats Finder; (iii) were found in homopolymer regions or multi-allelic sites; (iv) were previously detected in 3.5KJPNv2 or Human Genetic Variation Database (HGVD).

Statistical analysis

The correlations between variant allele frequencies (VAFs) and drug sensitivities were assessed using Spearman correlation tests. Significance after Bonferroni correction for multiple testing was $P = 1.11 \times 10^{-6}$ (P < 0.05; 44,875 variants). Statistical tests were conducted using Microsoft Excel 2016 (Microsoft Corporation, Redmond, WA, USA).

Results

Identification of variants associated with chemosensitivity

Whole-exome sequencing was used to identify genetic variants associated with chemosensitivity to one or more of nine cytotoxic anticancer drugs (MMC, CPM, ACNU, DDP, ADR, VER, VLB, 5FU, and MTX). Drugs were administered to 79 PDX models of cancers prepared from

Drug	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction	of functional effect	P value	r_s^c
								SIFT	PolyPhen2		
5FU	4	170428839	-	NEK1	-/A	intron				1.76E-03	-0.355
	19	57840351	-	ZNF543	T/C	exon	Y507Y			2.20E-03	0.394
	20	44572085	-	PCIF1	G/C	intron				2.52E-03	0.393
	19	8386994	-	RPS28	-/C	intron				3.09E-03	-0.335
	7	89861889	-	STEAP2	-/T	exon	M475Ifs*51	NA	NA	3.83E-03	-0.328
	8	145059414	rs144026672	PARP10	G/A	exon	P264P			5.66E-03	0.345
	15	63433763	-	LACTB	-/A	exon	R469Kfs*8	NA	NA	6.56E-03	-0.309
	15	74468404	rs746868961	ISLR	A/G	exon	E402G	Tolerated	Benign	6.59E-03	0.313
	17	39190653	-	KRTAP1-3	C/A	exon	A141S	Tolerated	Probably damaging	6.73E-03	0.358
	19	37488298	-	ZNF568	-/G	exon	E505Gfs*6	NA	NA	6.96E-03	-0.307
	17	40556634	rs3833143	CAVIN1	-/GAGCCGAGA	3'UTR				7.10E-03	-0.306
	1	157566015	-	FCRL4	-/A	intron				8.19E-03	-0.301
	3	100039903	-	TBC1D23	T/G	intron				8.39E-03	0.304
	7	149477851	-	SSPO	A/G	intron				9.44E-03	0.320
	11	68772966	-	MRGPRF	A/G	exon	F271S	Tolerated	Benign	9.76E-03	0.318

Table 1. Variants associated with chemosensitivity to 5FU (P < 0.01), as identified among 79 xenografts.

^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug.

5FU, 5-fluorouracil; Ref., reference; fs, frameshift; NA, not available.

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12 different human tissues. Between 30,012 and 45,638 variants were detected for each xenograft, with a coverage of 62 to 249 (mean; 37,921 variants, with depth of 138×). Variants were filtered using an in-house program (see <u>Materials and methods</u>), leaving a total of 44,875 variants for correlation analysis. Chemosensitivity was calculated as T/C and the variants whose allele frequency was higher in xenografts with lower T/C as were defined as 'chemosensitive variants' and variants whose allele frequency were higher in xenografts with higher T/C as 'chemoresistant variants'.

Although no variants reached a significance level of $P < 1.11 \times 10^{-6}$ (see Materials and methods), we observed variants showing P < 0.01 (7.15 × 10⁻⁵ < $P < 9.97 \times 10^{-3}$; Tables 1–9). The variant (chr8:g.22960701 insC) with the highest significance (lowest *P* value) was associated with chemosensitivity to ADR, and was located on two overlapping genes: uncharacterized LOC254896 (*LOC254896*) and TNF receptor superfamily member 10c (*TNFRSF10C*) ($P = 7.15 \times 10^{-5}$, $r_s = 0.437$; Table 3, Fig 1). As presented in Fig 1, particular to this variant, xenografts with higher VAFs had poorer responses to ADR than those with lower VAFs. This may suggest that variant chr8:g.22960701 insC may be associated with resistance to ADR.

For the other eight drugs, the variants most strongly associated with chemosensitivity were as follows (Tables 1–9): NIMA-related kinase 1 (*NEK1*) showed strong associations with 5FU treatment ($P = 1.76 \times 10^{-3}$, $r_s = -0.355$, Table 1); coiled-coil domain containing 66 (*CCDC66*) with ACNU ($P = 5.04 \times 10^{-4}$, $r_s = 0.387$, Table 2); copine 7 (*CPNE7*) with CPM ($P = 9.17 \times 10^{-5}$, $r_s = 0.426$, Table 4); SEMA3F antisense RNA 1 (*SEMA3F-AS1*) with DDP ($P = 6.66 \times 10^{-4}$, $r_s = 0.389$, Table 5); PAS domain-containing serine/threonine kinase (*PASK*) with MMC ($P = 2.05 \times 10^{-3}$, $r_s = 0.397$, Table 6); leucyl-tRNA synthetase 1 (*LARS*) with MTX ($P = 2.11 \times 10^{-3}$, $r_s = -0.366$, Table 7); protein kinase C delta (*PRKCD*) with VCR ($P = 2.38 \times 10^{-3}$,

Drug	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid	Prediction o	f functional effect	P value	r_s^c
							change	SIFT	PolyPhen2		
ACNU	3	56650052	rs1553710792	CCDC66	-/CTT	exon	T571_S572insF	NA	NA	5.04E- 04	0.387
	7	64169017	rs199594424	ZNF107	-/GAA	exon	E816delinsGK	NA	NA	7.86E- 04	0.375
	12	46760562	rs201547018	SLC38A2	C/A	intron				1.17E- 03	-0.468
	11	60617832	-	CCDC86	-/A	3'UTR				1.81E- 03	-0.350
	7	23872045	rs199803936	STK31	GA/-	3'UTR				2.69E- 03	0.337
	8	96281481	rs142455613	C8orf37-AS1	-/GGGGACCTGGC	ncRNA_intron				3.06E- 03	0.335
	15	42305854	-	PLA2G4E	-/AGG	intron				3.67E- 03	-0.327
	9	136135237	rs34229678	ABO	AT/GC	exon				3.96E- 03	-0.325
	9	15510020	rs148022076	PSIP1	-/G	intron				4.00E- 03	-0.324
	19	44648728	rs376448556	ZNF234	GC/TT	5'UTR				4.81E- 03	-0.318
	19	15789257	rs34521056	CYP4F12	T/C	intron				5.59E- 03	0.313
	10	64967953	rs139722368	JMJD1C	AAACCT/-	exon	G939_L940del	NA	NA	6.55E- 03	-0.307
	12	5022038	-	KCNA1	-/A	3'UTR				7.42E- 03	0.303
	8	142231944	rs386730897	SLC45A4	GC/AG	intron				7.64E- 03	-0.302
	14	75643383	-	TMED10	T/G	upstream				7.74E- 03	-0.314
	16	81242149	rs386792900	PKD1L2	TTT/-	exon	N236del	NA	NA	8.13E- 03	-0.300
	2	11348365	rs1553298800	ROCK2	-/TAACT	intron				8.33E- 03	-0.305
	8	132052227	-	ADCY8	G/T	exon	A118D	Tolerated	Benign	8.39E- 03	0.315
	22	29655909	-	RHBDD3	C/T	3'UTR				8.62E- 03	-0.342
	3	50232000	-	GNAT1	T/C	exon	S259P	Deleterious	Probably damaging	9.26E- 03	-0.336
	2	226447080	rs1292467126	NYAP2	A/G	exon	K316R	Tolerated	Benign	9.33E- 03	-0.330
	2	119600548	-	EN1	G/C	exon	T382S	Deleterious	Benign	9.65E- 03	-0.340
	19	50771503	-	MYH14	-/G	exon	A931Gfs*46	NA	NA	9.97E- 03	-0.296

Table 2. Variants associated with chemosensitivity to ACNU (P < 0.01), as identified among 79 xenografts.

^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug.

ACNU, nimustine; Ref., reference; ncRNA, noncoding RNA; del, deletion; ins, insertion; fs, frameshift; NA, not available.

NR Image I	Jrug	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Predic	ction of	P value	rs ^c
NN 6 200000 ···· 1000000 ···· 2000000 ···· 2000000 ···· 2000000 ···· 2000000 ···· 2000000 ···· 2000000 ···· 2000000 ···· 2000000 ···· 2000000 ···· 2000000 ···· 20000000 ···· 20000000 ···· 2000000000000000000000000000000000000									SIFT	PolyPhen2		
12 674015 ····· <i>XIA12 CaAi- Imm</i> ··· 2.90- <th< td=""><td>NDR</td><td>∞</td><td>22960701</td><td>1</td><td>LOC254896, TNFRSF10C</td><td>-/C</td><td>ncRNA_exon, intron</td><td></td><td></td><td></td><td>7.15E- 05</td><td>0.437</td></th<>	NDR	∞	22960701	1	LOC254896, TNFRSF10C	-/C	ncRNA_exon, intron				7.15E- 05	0.437
20 3996.53 ··· TALA4 ··/A ··/A </td <td></td> <td>12</td> <td>56740015</td> <td>1</td> <td>STAT2</td> <td>GAA/-</td> <td>intron</td> <td></td> <td></td> <td></td> <td>2.59E- 03</td> <td>0.339</td>		12	56740015	1	STAT2	GAA/-	intron				2.59E- 03	0.339
28 691443 ··· FBJA ··/ ·/ 3.9% 3.3% 3.3		20	7980553	1	TMX4	-/A	intron				3.39E- 03	-0.330
7600446657580125 $KCTD7$ $KCTD7$ $IAGGA$ intron $IatronIa$		22	45914432	1	FBLN1	À/T	intron				3.59E- 03	0.341
0 1331223 (side) <i>MTRFL -JATATG</i> intron $ < < < < < < < < < < < < < < < < < << << << << << << << <<< <<< <<< <<< <<<<<>< <<<<<>< <<<<<<<<>< <<<<<<<<<<><<<<<<<<<<<<<<<<<<>><<<<<<<$		~	66103436	rs57580125	KCTD7	-/AGGA	intron				4.01E- 03	-0.326
10 0.2363 0.718883 $EH3G$ $TGCCL$ inton 16 4.16 4.16 4.16 4.16 4.16 4.16 4.16 4.16 4.16 4.16 4.16 4.16 4.16 4.06 4.16 4.05 4.05 5.017 5.017 5.017 5.017 5.017 5.016		9	153312232	rs149540839	MTRF1L	-/ATATG	intron				4.14E- 03	0.325
15 667391 \cdots $CA12$ -16 -16 $5'UTR$ \cdots $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,685$ $16,815$ $10,815$ <		19	10226353	rs71188883	EIF3G	- 'JGCC/-	intron				4.41E- 03	-0.321
2 22647108 N1242 N1AP2 M/G <th< td=""><td></td><td>15</td><td>63673951</td><td>1</td><td>CA12</td><td>Ð/-</td><td>5'UTR</td><td></td><td></td><td></td><td>4.68E- 03</td><td>0.323</td></th<>		15	63673951	1	CA12	Ð/-	5'UTR				4.68E- 03	0.323
35015710 \cdot SEMA3F-AS1 h/C h/C $nCNA_intron$ $nCNA_intron$ $rrr<$		2	226447080	rs1292467126	NYAP2	A/G	exon	K316R	Tolerated	Benign	4.95E- 03	-0.355
314845934 \cdot $AGTRI$ $-IT$ $II33D16^{7}34$ NA $S_{10}0^{-}$ $S_{10}10^{-}$ S		ŝ	50155710	1	SEMA3F-ASI	A/C	ncRNA_intron				4.98E- 03	0.325
15 78581889 \cdot WDR61 $-^{/T}$ intron 5.4He 5.4He 0.31 16 113639 \cdot <i>RHBDF1</i> σ/A $exon$ S136S $r<$		3	148459394	1	AGTR1		exon	1193Dfs*34	NA	NA	5.10E- 03	-0.316
16 113639 ·· <i>KHBDF1</i> G/A $S/64^{-}$ $S.664^{-}$		15	78581889	1	WDR61	л/-	intron				5.44E- 03	0.314
19 44648728 rs37644856 ZNF234 GC/T4 GC/T4 5'UTR 5'UTR 6'52E 0.30 6'52E 0.308 0'3 <th< td=""><td></td><td>16</td><td>113639</td><td>1</td><td>RHBDF1</td><td>G/A</td><td>exon</td><td>S136S</td><td></td><td></td><td>5.66E- 03</td><td>0.386</td></th<>		16	113639	1	RHBDF1	G/A	exon	S136S			5.66E- 03	0.386
2 1134836 ts15329800 ROCK2 -/TAACT intron 6.99E 0.31E 17 4728467 rs1830594 GNGT2 -/A intron not 0.3 0.301 17 4728467 rs3830594 GNGT2 -/A intron A251delins NA NA 7.89E 0.301 2 7464267 rs768089535 C20781 -/GCGGAGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGG		19	44648728	rs376448556	ZNF234	GC/TT	5'UTR				6.52E- 03	-0.308
		7	11348365	rs1553298800	ROCK2	-/TAACT	intron				6.99E- 03	-0.311
2 7464267 rs768089535 C2orf81 -/GCGGAGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGGG		17	47284675	rs3830594	GNG72	-/A	intron				7.79E- 03	0.301
6 24450169 rs5874981 GPLD1 -/CCT intron 9.61E- 0.37 20 238437 - DEFB132 GGTCTT/- exon V7_L8del NA 9.50E- 0.296		2	74642267	rs768089535	C2orf81	-/GCGGAGGGGGGGGGGGGGCGCCCC	exon	A251delins GAAPPAPPP	NA	NA	7.89E- 03	0.301
20 238437 - DEFB132 GGTCTT/- exon V7_L8del NA 9.50E- -0.296		9	24450169	rs5874981	GPLD1	-/CCT	intron				8.61E- 03	0.297
		20	238437	-	DEFB132	GGTCTT/-	exon	V7_L8del	NA	NA	9.50E- 03	-0.296

Table 3. Variants associated with chemosensitivity to ADR (P < 0.01), as identified among 79 xenografis.

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^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug.

ADR, adriamycin; Ref., reference; ncRNA, noncoding RNA; del, deletion; ins, insertion; fs, frameshift; NA, not available.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

Allele Ref./Variant Location Amino ac
-/C intron
CCAGCCT/- intron
C/A exon
-/CGCCTACCTTGCC AGACCCTGGGCA intron
-/A intron
CC/AA intron
-/GACGGCTCAGCCAG CCTGTGGCATGG intron
-/CCT intror
-/CCTCGCGCTGTCTT intro:
C/A intro
-/A intro
-/CTGGATACCACAAATC intro
-/CT intr
A/G exo
G/T exc
-/GGCCCCTGCCC
G/T ex
G/T 5'U
TC/AA intr
GC/AA intr
-/C intr
TG/CA exc
C/A exc
AA/TT intr
-/T intr
C/T exc
-/G
T/G 5'U
-/AAAT intr

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^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug. CPM, cyclophosphamide; Ref., reference; NA, not available.

)rug	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction	of functional effect	P value	rs ^c
								SIFT	PolyPhen2		
DPP	ŝ	50155710	1	SEMA3F-ASI	A/C	ncRNA_intron				6.66E-04	0.389
	21	45843709	rs765207853	TRPM2-AS	AGG/-	ncRNA_intron				1.44E-03	-0.357
	19	40541037	1	ZNF780B	L/-	exon	S577Kfs*9	NA	NA	1.58E-03	0.354
	~	64169017	rs199594424	ZNF107	-/GAA	exon	E816delinsGK	NA	NA	1.73E-03	0.351
	~	23854839	rs5882915	STK31	-/A	intron				1.77E-03	0.351
	~	123190494	rs4147636	ND UFA5	-/CTGGATACC ACAAATC	intron				3.23E-03	-0.347
	17	7225146	1	NEURL4	5/-	intron				4.74E-03	-0.319
	19	2980268	,	TLE6	-/A	intron				5.03E-03	-0.317
	3	47453783	1	PTPN23	G/T	exon	G1271C	Deleterious	Probably damaging	5.55E-03	0.324
	15	78581889	,	WDR61	L/-	intron				5.68E-03	0.312
	15	73994678	rs1271868805	CD276	T/C	exon	P54P			6.42E-03	-0.351
	S	147695284	rs3217238	LOC102546294	-/TCA	ncRNA_intron				6.63E-03	-0.307
	9	350941	rs11408655	DUSP22	-/A	3'UTR				6.69E-03	0.307
	13	50092133	,	PHF11, SETDB2-PHF11	GTA/-	intron, intron				7.21E-03	0.319
	9	36759740	,	CPNE5	-/A	intron				7.38E-03	-0.303
	~	124749609	,	ANXA13	L/-	5'UTR				7.70E-03	-0.302
	19	21300592	,	ZNF714	C/A	exon	G374G			7.89E-03	-0.355
	22	20784908	rs35574298	SCARF2	TT/GA	intron				8.26E-03	0.299
	9	28331127	rs371085669	ZKSCAN3	AA/GC	exon	K52A	NA	NA	9.56E-03	0.294
	-	85787116	rs1172711726	DDAHI	-/C	3'UTR				9.92E-03	-0.292
	~	111846724	rs773775063	ZNF277	C/A	5'UTR				9.97E-03	0.372
			:		_						

Table 5. Variants associated with chemosensitivity to DDP (P < 0.01), as identified among 79 xenografts.

^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug. DDP, cisplatin; Ref., reference; ncRNA, noncoding RNA; del, deletion; ins, insertion; fs, frameshift; NA, not available.

Drug	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Predi functio	ction of nal effect	P value	r_s^c
								SIFT	PolyPhen2		
MMC	2	242078024	-	PASK	C/A	intron				2.05E-03	0.397
	8	68993013	rs368406603	PREX2	AT/GC	exon	F605F			3.11E-03	-0.329
	21	47910655	-	DIP2A	-/G	intron				3.21E-03	0.328
	9	71098986	rs377702519	PGM5	CA/TG	intron				4.84E-03	0.314
	12	58335540	-	ATP23	A/T	exon	Q19L	Tolerated	Benign	6.58E-03	0.322
	1	22852713	-	ZBTB40	-/C	exon	V1071Gfs*34	NA	NA	7.09E-03	0.301
	3	56593542	-	CCDC66	-/A	intron				7.59E-03	-0.298
	18	47320561	rs35615995	ACAA2	-/TAAA	intron				7.93E-03	-0.299
	5	79368050	-	CTD-2201118.1	GAA/-	ncRNA_intron				8.47E-03	-0.294
	6	52138176	-	МСМ3	-/A	intron				8.65E-03	0.294
	15	78581889	-	WDR61	-/T	intron				8.71E-03	0.293

Table 6. Variants associated with chemosensitivity to MMC (P < 0.01), as identified among 79 xenografts.

^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug.

MMC, mitomycin C; Ref., reference; ncRNA, noncoding RNA; fs, frameshift; NA, not available.

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 r_s = -0.343, Table 8); and arachidonate 5-lipoxygenase (*ALOX5*) with VLB (P = 1.65 ×10⁻³, r_s = 0.355, Table 9).

Genetic variants associated with multi-drug sensitivity

There were 162 variants possibly associated with chemosensitivity to more than one of the nine anticancer drugs (P < 0.01, Tables 1–9). rs1292467126 (chr2:g.226447080 A>G) in exon 4 of neuronal tyrosine-phosphorylated phosphoinositide-3-kinase adaptor 2 (*NYAP2*) was the most commonly associated variant, with chemosensitivity to four anti-cancer drugs: CPM ($P = 3.98 \times 10^{-3}$, $r_s = -0.361$; Table 4), ADR ($P = 4.95 \times 10^{-3}$, $r_s = -0.355$; Table 3), VCR ($P = 6.22 \times 10^{-3}$, $r_s = -0.347$; Table 8), and ACNU ($P = 9.33 \times 10^{-3}$, $r_s = -0.330$; Table 2). Xeno-grafts with higher VAFs of rs1292467126 had better responses to the four drugs, as shown in Table 10 and Fig 2. Furthermore, three variants were associated with three drugs and 13 variants with two drugs (Table 10). For example, rs773775063 (chr7:g.111846724 C>A) in the 5'UTR of zinc finger protein 277 (*ZNF277*) was associated with resistance to VLB ($P = 6.54 \times 10^{-3}$, $r_s = 0.395$), MTX ($P = 9.87 \times 10^{-3}$, $r_s = 0.398$), and DDP ($P = 9.97 \times 10^{-3}$, $r_s = 0.372$) (Table 10).

Subgroup analysis

We further performed a subgroup analysis based on cancer type to identify tissue-specific chemosensitivity-related variants. Subgroups of breast and gastric cancers were analyzed because more than 10 xenografts of these cancer types were available. In breast and gastric cancer xenografts, 78 and 67 variants, respectively, were possibly associated with chemosensitivity to one or more drugs, with *P* values < 0.01 (Tables 11 and 12). rs386792906 (chr16:g.81253642 AG>TC) in polycystin 1 like 2 (*PKD1L2*), which was associated with resistance to MTX, showed the strongest association of the nine tested anti-cancer drugs among the breast cancer

value r_s^c	1E-03 -0.366	(1E-03 0.365	2E-03 0.368		6E-03 -0.399	6E-03 -0.399 6E-03 0.350	6E-03 -0.399 6E-03 0.350 3E-03 0.349	6E-03 -0.399 6E-03 0.350 6E-03 0.349 3E-03 0.349 2E-03 0.346	6E-03 -0.399 6E-03 0.350 6E-03 0.349 3E-03 0.349 2E-03 0.346 9E-03 0.400	6E-03 -0.399 6E-03 0.350 3E-03 0.349 2E-03 0.346 2E-03 0.346 9E-03 0.346 5E-03 0.346	6E-03 -0.399 6E-03 0.350 6E-03 0.349 3E-03 0.349 2E-03 0.346 9E-03 0.346 6E-03 0.349 06-03 0.349 07-03 0.346 06-03 0.346 06-03 0.346 06-03 0.346 06-03 -0.334 06-03 -0.334	6E-03 -0.399 6E-03 0.350 6E-03 0.349 3E-03 0.349 2E-03 0.346 9E-03 0.400 5E-03 0.346 0E-03 0.346 05-03 0.364	6E-03 -0.399 6E-03 0.350 6E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.334 9E-03 0.334 9E-03 0.334 9E-03 0.364 5E-03 0.364 5E-03 0.364	6E-03 -0.399 6E-03 0.350 3E-03 0.349 3E-03 0.346 2E-03 0.346 9E-03 0.334 9E-03 0.334 9E-03 0.334 9E-03 0.364 9E-03 0.364 5E-03 0.364	6E-03 -0.399 6E-03 0.350 3E-03 0.349 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.340 9E-03 0.400 9E-03 0.400 9E-03 0.346 9E-03 0.304 9E-03 0.346 9E-03 0.334 9E-03 0.324 9E-03 0.324 9E-03 0.324	6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.330 9E-03 0.331 9E-03 0.332 9E-03 0.332 9E-03 0.364 2E-03 0.324 2E-03 0.324 2E-03 0.324 2E-03 0.324 2E-03 0.324 2E-03 0.324 2E-03 0.324	6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 9E-03 0.346 5E-03 0.364 1E-03 0.324 2E-03 0.322 2E-03 0.322 7E-03 0.322 7E-03 0.322 75E-03 0.322	6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.364 9E-03 0.320 5E-03 0.322 2E-03 0.322 7E-03 0.322 6E-03 0.322 6E-03 0.322 6E-03 0.322 6E-03 0.323 6E-03 0.323	6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.3034 9E-03 0.3046 9E-03 0.3046 9E-03 0.3024 9E-03 0.302 9E-03 0.324 9E-03 0.324 9E-03 0.324 9E-03 0.324 9B-03 0.327 3E-03 0.327	6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.340 9E-03 0.346 9E-03 0.340 9E-03 0.304 9E-03 0.304 9E-03 0.304 9E-03 0.302 9E-03 0.324 9E-03 0.324 92 0.323 932 9.324 932 9.327 932 9.327 917 9.323 8E-03 0.317 8E-03 0.327 8E-03 0.327 8E-03 0.317 8E-03 0.317 8E-03 0.317	6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.364 6E-03 0.324 9E-03 0.324 9D 0.323 9D 0.323 9D 0.324 9D 0.322 9D 0.322 9D 0.323 9D 0.324 9D 0.324 9D 0.322 9D 0.323 9D 0.323 9D 0.323 8E-03 0.327 8E-03 0.327 8E-03 0.327 8E-03 0.327 9D 0.323 8E-03 0.327 8E-03 0.327 8E-03 0.327	6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 9E-03 0.364 9E-03 0.320 5E-03 0.364 5E-03 0.324 1E-03 0.324 2E-03 0.327 2E-03 0.320 4E-03 0.327 5E-03 0.322 5E-03 0.377 5E-03 0.377 5E-03 0.377 5E-03 0.377 5E-03 0.377 5E-03 </th <th>6E-03 -0.399 6E-03 0.350 3E-03 0.350 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.304 9E-03 0.304 9E-03 0.304 9E-03 0.304 9E-03 0.304 9E-03 0.324 9E-03 0.324 5E-03 0.324 1E-03 0.324 1E-03 0.324 2E-03 0.324 2E-03 0.327 4E-03 0.327 5E-03 0.320 4E-03 0.327 5E-03 0.317 5E-03 0.317 5E-03 0.363 3E-03 0.317 5E-03 0.317 5E-03 0.317 5E-03 0.317 5E-03 0.314 5E-03 0.314 5E-03<!--</th--><th>6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.340 9E-03 0.346 9E-03 0.3046 9E-03 0.302 9E-03 0.302 9E-03 0.324 9E-03 0.320 5E-03 0.324 1E-03 0.324 1E-03 0.324 2E-03 0.327 4E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.373 8E-03 0.317 6E-03 0.317 6E-03 0.312 6E-03 0.312 7E-03 0.312 7E-03 0.312 7E-03<</th></th>	6E-03 -0.399 6E-03 0.350 3E-03 0.350 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.304 9E-03 0.304 9E-03 0.304 9E-03 0.304 9E-03 0.304 9E-03 0.324 9E-03 0.324 5E-03 0.324 1E-03 0.324 1E-03 0.324 2E-03 0.324 2E-03 0.327 4E-03 0.327 5E-03 0.320 4E-03 0.327 5E-03 0.317 5E-03 0.317 5E-03 0.363 3E-03 0.317 5E-03 0.317 5E-03 0.317 5E-03 0.317 5E-03 0.314 5E-03 0.314 5E-03 </th <th>6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.340 9E-03 0.346 9E-03 0.3046 9E-03 0.302 9E-03 0.302 9E-03 0.324 9E-03 0.320 5E-03 0.324 1E-03 0.324 1E-03 0.324 2E-03 0.327 4E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.373 8E-03 0.317 6E-03 0.317 6E-03 0.312 6E-03 0.312 7E-03 0.312 7E-03 0.312 7E-03<</th>	6E-03 -0.399 6E-03 0.350 3E-03 0.346 3E-03 0.346 2E-03 0.346 9E-03 0.346 9E-03 0.346 9E-03 0.340 9E-03 0.346 9E-03 0.3046 9E-03 0.302 9E-03 0.302 9E-03 0.324 9E-03 0.320 5E-03 0.324 1E-03 0.324 1E-03 0.324 2E-03 0.327 4E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.327 6E-03 0.373 8E-03 0.317 6E-03 0.317 6E-03 0.312 6E-03 0.312 7E-03 0.312 7E-03 0.312 7E-03<
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SIFT	NA		NA	Deleterious			NA	NA NA	NA NA NA Deleterious	NA NA NA Deleterious	NA NA Deleterious	NA NA Deleterious	NA NA Deleterious	NA NA Deleterious	NA NA Deleterious	NA NA Deleterious	NA NA Deleterious NA	NA NA Deleterious NA NA	NA NA Deleterious NA NA	NA NA Deleterious NA NA NA	NA NA Deleterious NA NA NA NA NA Tolerated	NA NA Deleterious NA NA NA NA NA Tolerated	NA NA Deleterious NA NA NA NA Tolerated NA	NA NA Deleterious NA NA NA NA NA Tolerated NA
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9	MTX																							

Table 7. Variants associated with chemosensitivity to MTX (P < 0.01), as identified among 79 xenografts.

^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug. MTX, methotrexate; Ref., reference; ncRNA, noncoding RNA; fs, frameshift; NA, not available.

Drug	Chr	Position ^a	$rsID^b$	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction 6	of functional effect	P value	rs ^c
								SIFT	PolyPhen2		
VCR	3	53220765	rs3830265	PRKCD	TCAGAGCC/-	intron				2.38E-03	-0.343
	4	151187012	rs150278643	LRBA	-/GAGAT	intron				2.81E-03	0.338
	22	45182326	rs67401095	ARHGAP8, PRR5-ARHGAP8	CTT/-	intron, intron				3.08E-03	-0.335
	10	73115941	rs34040486	SLC29A3	TG/CA	exon	V239I	NA	NA	3.12E-03	-0.335
	1	248367014	-	OR2M3	TG/CA	exon	A216T	NA	NA	3.47E-03	-0.331
	14	77751922	1	POMT2	-/Τ	exon	F463Ifs*77	NA	NA	3.87E-03	0.328
	4	184619035	rs200831837	TRAPPC11	C/T	intron				4.46E-03	0.323
	18	40695532		RIT2	T/C	5'UTR				5.29E-03	-0.392
	19	21300592	1	ZNF714	C/A	exon	G374G			5.34E-03	-0.374
	3	24006476		NR1D2	-/A	exon	L311Ifs*2	NA	NA	5.52E-03	-0.315
	15	43028509	-	CDANI	-/C	exon	T189Yfs*45	NA	NA	6.03E-03	0.312
	19	21606610	1	ZNF493	C/A	exon	G255G			6.22E-03	-0.361
	2	226447080	rs1292467126	NYAP2	A/G	exon	K316R	Tolerated	Benign	6.22E-03	-0.347
	19	21240167		ZNF430	C/A	exon	G350G			6.31E-03	-0.349
	3	124646709	rs869290005	MUC13	-/AAG	exon	T60_P61insL	NA	NA	6.35E-03	-0.310
	19	36355594	1	KIRREL2	-/G	exon	K541Efs*50	NA	NA	6.47E-03	0.310
	16	22161135		VWA3A	-/C	exon	F1005Lfs*16	NA	NA	7.42E-03	-0.305
	20	948071	1	RSPO4	T/C	intron				8.00E-03	-0.357
	12	75816816	rs59277111	GLIPR1L2	-/CAA	exon	D239_K240insQ	NA	NA	8.66E-03	-0.299
	17	47888851	1	KAT7	-/G	exon	Q88Tfs*4	NA	NA	8.88E-03	-0.298
	10	78084100	rs372941859	LRMDA	GG/CC	intron				9.29E-03	-0.297
	19	20002842		ZNF253	C/A	exon	G186G			9.46E-03	-0.347
	3	50232000	1	GNATI	T/C	exon	S259P	Deleterious	Probably damaging	9.58E-03	-0.337
	8	74335015	rs60338415	STAU2-ASI	-/AGAAAGAC	ncRNA_intron				9.77E-03	0.297
	20	57430029	ı	GNAS	C/A	exon	P508T	Tolerated	Benign	9.94E-03	-0.325
	1	67423998	rs142198730	MIERI	-/TTCTC	intron				9.95E-03	-0.302
- -	Ę		:								

Table 8. Variants associated with chemosensitivity to VCR (P < 0.01), as identified among 79 xenografts.

^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug.

VCR, vincristine; Ref., reference; ncRNA, noncoding RNA; ins, insertion; fs, frameshift; NA, not available.

Drug	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Pre funct	diction of ional effect	P value	r _s ^c
								SIFT	PolyPhen2		
VLB	10	45869697	-	ALOX5	-/C	5'UTR				1.65E-03	0.355
	1	169679473	rs4987281	SELL	-/CT	intron				1.96E-03	0.350
	3	169540395	-	LRRIQ4	-/C	exon	C231Vfs*3	NA	NA	2.82E-03	-0.340
	9	132652688	-	FNBP1	-/GAC	3'UTR				4.06E-03	0.326
	4	184619035	rs200831837	TRAPPC11	C/T	intron				4.16E-03	0.325
	8	96281481	rs142455613	C8orf37-AS1	-/GGGGACCTGGC	ncRNA_intron				4.21E-03	0.327
	4	6293234	-	WFS1	-/G	intron				5.35E-03	0.316
	7	1528998	-	INTS1	AT/CA	exon	M767W	NA	NA	6.08E-03	0.312
	22	50659594	-	TUBGCP6	TC/CT	exon	E1065R	NA	NA	6.52E-03	0.309
	7	111846724	rs773775063	ZNF277	C/A	5'UTR				6.54E-03	0.395
	6	74123314	rs35252896	DDX43	-/GCT	intron				8.14E-03	-0.301
	7	12391269	rs11454536	VWDE	-/A	exon	K1158fs*0	NA	NA	8.80E-03	0.299
	13	24869045	-	SPATA13	-/C	intron				8.97E-03	0.298
	19	21300592	-	ZNF714	C/A	exon	G374G			9.32E-03	-0.351
	7	111846719	-	ZNF277	C/A	5'UTR				9.88E-03	0.369
	2	160075887	rs3214491	TANC1	-/C	intron				9.90E-03	-0.294

Table 9. Variants associated with chemosensitivity to VLB (P < 0.01), as identified among 79 xenografts.

^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug.

VLB, vinblastine; Ref., reference; ncRNA, noncoding RNA; fs, frameshift; NA, not available.

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subgroup ($P = 1.52 \times 10^{-5}$, $r_s = 0.991$, Table 11). rs73302038 (chr17:g.21215682 G>A) in mitogen-activated protein kinase kinase 3 (*MAP2K3*), which was associated with resistance to VCR, showed the strongest association for the gastric cancer subgroup ($P = 8.32 \times 10^{-6}$, $r_s = 0.935$, Table 12). However, of the variants with P < 0.01 in the subgroup analyses, only three (chr15:g. 22960698 insG in MTX, rs59277111 in VCR, and rs3217238 in DDP) were significant at P < 0.01 in the whole-group analysis (Tables 11 and 12).

Discussion

Precision medicine demands the development of biomarkers to detect patient chemosensitivity to anti-cancer drugs. Here, we sought to identify clinically useful genetic markers for chemosensitivity to one or more of nine cytotoxic anticancer drugs by whole-exome sequencing for 79 xenografts. Although none of the genetic variants achieved a significance level after Bonferroni correction for multiple testing ($P = 1.11 \times 10^{-6}$), numerous variants showed possible associations with chemosensitivity to each of the nine tested drugs. Moreover, the subgroup analysis indicated chemosensitivity markers specific for breast and gastric cancers. We propose that our method could contribute to the development and optimization of personalized chemotherapy regimens among patients with cancer.

In the whole-exome sequencing analysis of 79 xenografts, we found that, the variant chr8: g.22960701insC, located in *TNFRSF10C* and *LOC254896*, had the most significant (i.e., lowest) *P* value for its associated chemosensitivity to ADR ($P = 7.15 \times 10^{-5}$, $r_s = 0.437$, <u>Table 3</u>, <u>Fig 1</u>). Although the function of *LOC254896* remains to be clarified, the down-regulated expression



Fig 1. Correlation between variant chr8:g.22960701insC and chemosensitivity to ADR. Chemosensitivity to ADR is represented by relative tumor volume of treated mice (T) with respect to that of the control mice (C). Xenografts with a higher VAF exhibited a poorer response to ADR than those with a lower VAF.

https://doi.org/10.1371/journal.pone.0239614.g001

[32,33] and hypermethylation [34,35] of *TNFRSF10C* in colorectal, prostate, and breast cancers has been reported previously. Additionally, in vitro experiments have suggested that an upre-gulation in *TNFRSF10C* in response to ADR treatment may induce resistance to ADR [36]. TNFRSF10C is reported to protect cells from TRAIL-induced apoptosis [37], and thus may be associated with resistance to ADR through these pathways.

A variant (chr15:g.63673951 insG) located in the 5'UTR of carbonic anhydrase 12 (*CA12*) was also associated with resistance to ADR (<u>Table 3</u>). CA12 is a membrane carbonic anhydrase and plays important roles in several physiological functions, such as acid-base balance and calcification [38]. A recent in vitro study showed CA12 overexpression in chemoresistant colon cancer cells expressing the drug efflux transporter P-glycoprotein (Pgp). Moreover, ADR chemosensitivity in tumors overexpressing both CA12 and Pgp can be increased using CA12 inhibitors [39]. Therefore, the chr15:g.63673951 insG variant may increase resistance to ADR by altering CA12 expression; further functional analyses would be required to verify this hypothesis.

Moreover, a variant (chr2:g.189916175 T>G) associated with resistance to MTX was located in exon 42 of collagen type V alpha 2 chain (*COL5A2*) (Table 7). COL5A2 is upregulated in colorectal and breast cancers [40,41] and is associated with poor clinical outcome and poor survival rates in bladder cancer [42]. Studies have suggested that collagen expression increases tumor drug resistance by inhibiting drug penetration into the cancer tissue and increasing cellular resistance to apoptosis [43]. As shown in Table 10, we identified genetic variants that could be associated with multi-drug resistance or sensitivity. Some of these genes may be involved in the proliferation and invasion of tumor cells; for example, *NYAP2* is reported to activate PI3K, Akt and Rac1, and mediates remodeling of the actin cytoskeleton

Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction ef	of functional fect	P value	r_s^c	Drug
							SIFT	PolyPhen2			
2	226447080	rs1292467126	NYAP2	A/G	exon	K316R	Tolerated	Benign	3.98E- 03	-0.361	СРМ
									4.95E- 03	-0.355	ADR
									6.22E- 03	-0.347	VCR
									9.33E- 03	-0.330	ACNU
19	21300592	-	ZNF714	C/A	exon	G374G			5.34E- 03	-0.374	VCR
									7.89E- 03	-0.355	DDP
									9.32E- 03	-0.351	VLB
15	78581889	-	WDR61	-/T	intron				5.44E- 03	0.314	ADR
									5.68E- 03	0.312	DDP
									8.71E- 03	0.293	MMC
7	111846724	rs773775063	ZNF277	C/A	5'UTR				6.54E- 03	0.395	VLB
									9.87E- 03	0.398	MTX
									9.97E- 03	0.372	DDP
3	50155710	-	SEMA3F-AS1	A/C	ncRNA_intron				6.66E- 04	0.389	DDP
									4.98E- 03	0.325	ADR
7	64169017	rs199594424	ZNF107	-/GAA	exon	E816delinsGK	NA	NA	7.86E- 04	0.375	ACNU
									1.73E- 03	0.351	DDP
7	23854839	rs5882915	STK31	-/A	intron				1.77E- 03	0.351	DDP
									2.17E- 03	0.340	СРМ
6	24450169	rs5874981	GPLD1	-/CCT	intron				2.43E- 03	0.337	СРМ
									8.61E- 03	0.297	ADR
8	96281481	rs142455613	C8orf37-AS1	-/GGGGACCTGGC	ncRNA_intron				3.06E- 03	0.335	ACNU
									4.21E- 03	0.327	VLB
7	123190494	rs4147636	NDUFA5	-/CTGGATACCACAAATC	intron				3.23E- 03	-0.347	DDP
									3.97E- 03	-0.335	СРМ

Table 10. Variants commonly associated with chemosensitivity to two or more anticancer drugs (P < 0.01).

(Continued)

Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction ef	of functional fect	P value	r_s^c	Drug
							SIFT	PolyPhen2			
1	248367014	-	OR2M3	TG/CA	exon	A216T	NA	NA	3.47E- 03	-0.331	VCR
									5.89E- 03	-0.307	СРМ
7	12391269	rs11454536	VWDE	-/A	exon	K1158fs*0	NA	NA	3.82E- 03	0.346	MTX
									8.80E- 03	0.299	VLB
4	184619035	rs200831837	TRAPPC11	C/T	intron				4.16E- 03	0.325	VLB
									4.46E- 03	0.323	VCR
19	44648728	rs376448556	ZNF234	GC/TT	5'UTR				4.81E- 03	-0.318	ACNU
									6.52E- 03	-0.308	ADR
2	11348365	rs1553298800	ROCK2	-/TAACT	intron				6.99E- 03	-0.311	ADR
									8.33E- 03	-0.305	ACNU
17	47284675	rs3830594	GNGT2	-/A	intron				7.79E- 03	0.301	ADR
									9.65E- 03	0.312	MTX
3	50232000	-	GNAT1	T/C	exon	S259P	Deleterious	Probably damaging	9.26E- 03	-0.336	ACNU
									9.58E- 03	-0.337	VCR

Table 10. (Continued)

^a Based on GRCh37 genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

^c Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug.

ACNU, nimustine; ADR, adriamycin; CPM, cyclophosphamide; DDP, cisplatin; MMC, mitomycin C; MTX, methotrexate; VCR, vincristine; VLB, vinblastine; ncRNA, noncoding RNA; del, deletion; ins, insertion; fs, frameshift; NA, not available.

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[44], whereas *ZNF277* regulates cell migration and invasion through phosphatase and tensin homolog (*PTEN*) [45].

In the subgroup analysis using breast and gastric cancer xenografts, we identified possible tissue-specific biomarkers in the response to anticancer drugs; however, most of these variants showed weak or no association in the whole-group analysis. These results suggest a degree of tissue specificity in sensitivity to cytotoxic anticancer drugs. rs386792906 (chr16:g.81253642 AG>TC), which showed the strongest association with MTX chemosensitivity in breast cancer xenografts, was located in intron 1 of *PKD1L2*. PKD1L2 is a member of the polycystin protein family, and may function as a component of cationic channel pores [46]. According to a previous study using The Cancer Genome Atlas (TCGA) dataset, overexpression of *PKD1L2* mRNA is associated with improved prognosis in patients with breast cancer [47]. Although the functional association between PKD1L2 and MTX is unknown, this variant may be a useful



Fig 2. Correlation between variant rs1292467126 and chemosensitivity to CPM (A), ADR (B), VCR (C), and ACNU (D). Chemosensitivity to each drug is represented by relative tumor volume of treated mice (T) with respect to that of the control mice (C). rs1292467126 was commonly associated with increased sensitivity to four of the nine tested anticancer drugs.

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marker for predicting sensitivity to MTX, and may act as an indicator of prognosis for breast cancer in the clinical setting.

We investigated the functional consequences of the associations between the top variants and the response to chemotherapy by interrogating the expression quantitative trait loci (eQTL) information in the Genotype-Tissue Expression (GTEx) database [48]. rs3830265, which showed the strongest association with sensitivity to VCR, was associated with the expression of *PRKCD* in the skin ($P = 7.3 \times 10^{-7}$) and esophagus ($P = 1.7 \times 10^{-5}$). Moreover, rs3842515, which showed the strongest association with sensitivity to ACNU in breast cancer xenograft, displayed a cis-regulatory effect on *CCDC82* expression in several tissues, including esophagus, thyroid, skin, and nerve ($P_{min} = 2.3 \times 10^{-13}$). However, the functional associations between these genes (*PRKCD* and *CCDC82*) and sensitivities to the aforementioned drugs or mechanisms of drug metabolism remain unknown and require further investigation.

There were several strengths and limitations in our study. The main strength of our study is that we sought to identify tissue-agnostic predictive markers for chemosensitivity to nine cyto-toxic anticancer drugs. As we have entered a new era of precision medicine, tissue-agnostic cancer therapy will continue to grow and expand treatment options for patients with cancer [49]. In addition to our tissue-agnostic approach, we also performed subgroup analyses of breast and gastric cancers as a deeper understanding of the genomic profiles of specific tumor types is also important. There were several limitations in our study. First, the total number of xenografts and the total number of each tumor type are small, and there were differences in the numbers of tumor types. Therefore, our study is likely to be underpowered to detect

rafts	rs ^c	0.084	-0.048	0.258	-0.257	0.223	0.227	-0.144	-0.017	0.004	0.116	-0.128	-0.065	-0.003	-0.13	0.046	-0.077	-0.224	0.085	0.019	0.039	
vll xenog	<i>P</i> value	4.69E- 01	6.96E- 01	2.43E- 02	2.47E- 02	5.31E- 02	4.83E- 02	2.15E- 01	8.83E- 01	9.76E- 01	3.18E- 01	2.72E- 01	5.72E- 01	9.80E- 01	2.58E- 01	6.94E- 01	5.08E- 01	4.99E- 02	4.60E- 01	8.67E- 01	7.38E- 01	
V	z	76	70	76	76	76	76	76	75	68	76	76	77	77	77	77	77	77	77	77	77	
ncer	rs ^c	0.891	-0.907	0.814	-0.798	0.798	0.791	-0.790	-0.782	-0.844	0.773	-0.768	-0.861	-0.844	-0.821	-0.820	0.777	-0.766	0.758	0.747	-0.745	
reast car	P value	5.42E- 04	7.34E- 04	4.16E- 03	5.69E- 03	5.69E- 03	6.38E- 03	6.51E- 03	7.55E- 03	8.35E- 03	8.75E- 03	9.47E- 03	6.67E- 04	1.08E- 03	1.94E- 03	2.00E- 03	4.91E- 03	5.99E- 03	6.86E- 03	8.29E- 03	8.57E- 03	
B	z	10	6	10	10	10	10	10	10	∞	10	10	1	Ξ	Ξ	Ξ	Ξ	Ξ	Ξ	Ξ	Ξ	
of functional fect	PolyPhen2				NA			NA						NA		NA				NA	NA	
Prediction c	SIFT				NA			NA						NA		NA				NA	NA	
Amino acid change					E250Gfs*6			E200delinsGK, E265delinsGK						L2436Afs*4		X808delinsX				Y268H	Q243Pfs*9	
Location		intron	intron	3'UTR	exon	intron	intron	exon, exon	intron	intron	intron	intron	5'UTR	exon	intron	exon	intron	intron	intron	exon	exon	
Allele Ref./Variant		-/TCTG	-/GCTGGGGC CTGGAGC	GT/AC	-/C	-/A	-/G	-/GAA	-/ATTA	-/GACGGCTCAGCCA GCCTGTGGCATGG	GG/AA	-/A	-/G	-/G	GTTT/-	-/A	GG/CC	-/A	-/TCC	AT/TC	-/C	
Gene		MTR	MYO7A	NOP10	HPSE	SLC35F3	DIP2A	SH3BGR, WRB-SH3BGR	ESFI	OBSCN	KCNABI	COL13A1	JRKL	ITPR3	SYNEI	ABCF1	LRMDA	TLE6	RPL7L1	UGT2B7	MAN2B2	
rsID ^b		rs35668201	rs35298297	rs386782889	1	1	rs74854320	1	rs28372964	rs71180792	rs34680920	rs74503792	rs3842515	1	1	1	rs372941859	1	1	rs386675647	1	
Position ^a		236978993	76901624	34634138	84230617	234458667	47985555	40883673	13763897	228529430	156175167	71648214	96123735	33659469	152461050	30558478	78084100	2980268	42853640	69964337	6594943	
Chr		-	=	15	4	-	21	21	20	-	ε	10	11	9	9	9	10	19	9	4	4	
Drug		5FU	<u> </u>	<u> </u>	<u> </u>	1	1	I	<u> </u>	<u> </u>	<u> </u>	1	ACNU	<u> </u>	<u> </u>	<u> </u>	<u> </u>	1	<u> </u>	1	1	

Drug	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction o effi	f functional ect	Bı	reast can	Icer	A	ll xenog	grafts
								SIFT	PolyPhen2	z	<i>P</i> value	rs ^c	z	<i>P</i> value	rs ^c
ADR	14	69791440	1	GALNT16	C/A	exon	P123T	Deleterious	Probably damaging	10	6.43E- 04	0.886	60	9.04E- 01	-0.016
	11	56468448	rs1554964167	OR9G1, OR9G9	AA/GT	exon, exon	I196F, I196F	NA	NA	=	2.88E- 03	-0.804	77	4.33E- 02	-0.231
	12	6886294	1	LAG3	A/G	intron				~	3.18E- 03	0.889	51	3.70E- 02	0.293
	16	4254408	rs1555454446	SRL	-/AGATACAGCCC CGGCCTCCA	intron				Ξ	3.63E- 03	-0.792	76	7.37E- 01	0.039
	14	75745752	ı	FOS	G/C	exon	A23P	Tolerated	Probably damaging	~	4.80E- 03	0.907	67	5.00E- 01	0.084
	~	100361392	rs3215395	ZAN	-/C	intron				11	5.83E- 03	0.767	77	4.18E- 01	0.094
	ŝ	1093610	rs56276350	SLC12A7	-/GGGCGGGGGACT	intron				10	5.93E- 03	0.795	73	5.48E- 01	0.071
	10	73571582	rs59718926	CDH23	-/CT	intron				Ξ	7.44E- 03	-0.753	77	5.28E- 01	-0.073
	16	138773	rs57321480	NPRL3	Ð/-	exon				Ξ	7.82E- 03	0.750	77	9.47E- 01	0.008
	22	50927448	rs55651311	XOIW	-/GTCCCTCCT	intron				10	9.83E- 03	-0.766	71	6.24E- 01	-0.059
CPM	9	42853640	I	RPL7L1	-/TCC	intron				12	3.32E- 03	0.771	79	1.56E- 01	0.161
	10	78084100	rs372941859	LRMDA	GG/CC	intron				12	3.43E- 03	0.769	79	8.48E- 01	0.022
	ŝ	49148887	ı	61dSD	-/C	intron				12	4.10E- 03	0.760	79	2.86E- 01	0.122
	~	100229467	ı	TFR2	G/T	exon	A185A			∞	5.15E- 03	0.868	42	1.03E- 01	0.255
		234458667	ı	SLC35F3	-/A	intron				12	8.52E- 03	0.718	79	4.80E- 02	0.223
	-	164781111	rs869176116	PBXI	-/ATATAAG	intron				12	9.83E- 03	0.709	75	3.10E- 02	0.249
	11	66333595		CTSF	-/T	exon	E256Rfs*13	NA	NA	12	9.98E- 03	-0.708	79	1.85E- 02	-0.264
														(Co	ntinued)

interplate interpl	Ū	hr Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction of effe	f functional ct	m	reast car	ıcer	W	l xenogr	afts
JYA6356 ··· <i>IAAGC GaT Gato Gato Gato G Gat G Gat G Gat Gat</i>								SIFT	PolyPhen2	z	P value	rs ^c	z	<i>P</i> value	rs ^c
4353640 ········ <i>R R L L</i> ······· <i>L L</i>		137682568	1	FAM53C	G/T	exon	G367C	Deleterious	Benign	9	7.14E- 04	0.978	46	7.39E- 01	0.051
78864100 Ca724489 LMMA Cackc Into Into 233 7 734 734		42853640	1	RPL7L1	-/TCC	intron				Ξ	1.14E- 03	0.842	5	2.29E- 01	0.139
0604374 05366756/T 07213F M/T 0406 NA 013 056 073 7 3 05 0 0 07868765 RATNU -77CC exon C942.1 NA 11 5 17 17 17 17 17 17 17 17 10 0786876 RATNU -77CC exon C942.14346 NA 11 5 17 17 17 17 10 18387801 -0.0571 -0.0571 exon PAXJ NA 11 10 10 11	0	78084100	rs372941859	LRMDA	GG/CC	intron				Ξ	1.32E- 03	0.837	12	7.23E- 01	0.041
678.882 0388.8806 RTTV -7 CC exon 22.2 D245ins NA		69964337	rs386675647	UGT2B7	AT/TC	exon	Y268H	NA	NA	Ξ	3.68E- 03	0.792	12	3.46E- 01	0.109
UBCR7101 UCGT1 JA exon V320G162 20 NA NA I1 S716 C16 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01 7 5.01	~	67863852	rs386388096	RTTN	-/TCC	exon	G242_D243insE	NA	NA	Ξ	5.30E- 03	-0.773	5	9.17E- 01	-0.012
134353rescripted $CDRT4$ $-CTT$ exon $E9$ -VI0ind NA $[056]$ 056 076 7 206 0.17 134023 $-ETFI-HOLCNS$ $DPYX4$ $CCSAGGGC$ intron $EFIFI-HOLCNS$ $CCSAGGGC$ $IntronCC_{-0}12202$		128878011	1	UGGTI	-/A	exon	V320Gfs*20	NA	NA	Ξ	5.71E- 03	0.769	12	3.69E- 01	0.104
1401202 \cdot DPYSIACCGAGGG/-IntrolIntrol \cdot <td></td> <td>15343525</td> <td>rs66754946</td> <td>CDRT4</td> <td>-/CTT</td> <td>exon</td> <td>E9_V10insK</td> <td>NA</td> <td>NA</td> <td>Ξ</td> <td>6.05E- 03</td> <td>-0.765</td> <td>12</td> <td>2.70E- 01</td> <td>-0.127</td>		15343525	rs66754946	CDRT4	-/CTT	exon	E9_V10insK	NA	NA	Ξ	6.05E- 03	-0.765	12	2.70E- 01	-0.127
03635 ··· EFLIFJ.BLOCK3 GAGGTATTV- ReNA_intro ReNA_intro 2	0	134012502	1	DPYSL4	CCGAGGGG/-	intron				Ξ	7.02E- 03	0.757	5	9.36E- 01	0.00
238464001 DBSCN AGTT exon R2321 NA I 806 7 7 838 0 238464001 DBSCN CA/TT exon E332_53373 NA I 806 7 7 801 23847536 rs38640016 DBSCN CA/TT exon E337_53373 NA I 806 7 7 80 7 80 7 80 7 80 7 80 7 80 7 80 7 80 7 80 7 80 7 80 7 80 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 90 7 10 101		8073625	1	EEF1E1-BLOC1S5	AGAGTAGTTT/-	ncRNA_intron				Ξ	7.65E- 03	-0.752	12	2.92E- 01	0.122
2844566 resolution Galvar exon E332_S3373 NA 13441417		228469903	rs386640014	OBSCN	AG/TT	exon	R2823L	NA	NA	Ξ	8.60E- 03	0.744	12	3.38E- 01	0.024
1247306 · EC72 · AAT inton		228476366	rs386640016	OBSCN	GA/TT	exon	E3372_S3373 delinsDC	NA	NA	11	8.72E- 03	0.744	27	3.83E- 01	0.101
96123735 r5344215 JRKL -/G 5'UTR 5'UTR -/G 0,3 7 5,06-b 0,3 7 5,06-b 0,3 7 5,06-b 0,3 1 11344411 OAS2 -/C intro int	~	172473062	'	ECT2	-/AT	intron				11	9.22E- 03	0.740	22	5.68E- 01	0.066
11344417 ·· <	-	96123735	rs3842515	JRKL	Ð/-	5'UTR				11	9.43E- 03	-0.739	77	5.06E- 01	0.077
36587848 rs69113251 APOL4 -/CT exon exon r 1 3.66E 0.857 7 2.82E 0.187 7 0 0 772841 - - -/C intron -/C intron 2.91E 0.785 79 8.15E 0.01 772841 - CCDC78 -/C intron intron 12 2.49E 0.785 79 8.15E 0.02 26906340 - C UH9 T/G intron R285R 12 2.91E 0.10 0.10 38318262 - MICALLI C/A won R285R 10 8.95E 0.85 6 0.16 0.12 38318262 - MICALLI C/A won R285R 10 8.95E 0.805 6 9.16 0.10 38318262 - MICALLI C/A MICALLI MICALLI 10 10 10 10 10 10	2	113444417	'	OAS2	-/C	intron				12	2.85E- 04	-0.864	79	3.18E- 02	-0.242
772841 CCDC78 -/C intron intro	2	36587848	rs869115251	APOL4	-/CT	exon				12	3.66E- 04	0.857	79	2.82E- 01	0.123
26906240 - CDH9 T/G intron intro intron intro <	9	772841	'	CCDC78	-/C	intron				12	2.49E- 03	-0.785	79	8.15E- 01	0.027
38318262 - MICALL1 C/A exon R285R - 9 6.15E - 0.825 68 9.52E- 0.008 44274143 - MARS2 GAA/- intron intron 12 6.99E- 0.730 79 1.56E- 0.16 44274143 - MARS2 GAA/- intron 1440N 70 12 6.99E- 0.730 79 161 0.16 203816588 - ZC3H11A T/A exon 1440N Tolerated Benign 7 9.74E- 0.876 53 881E- 0.237		26906240	'	CDH9	T/G	intron				10	4.95E- 03	-0.805	76	2.91E- 01	-0.123
44274143 - AARS2 GAA/- intron intron 12 6.9E- 0.730 79 1.5E- 0.161 203816588 - ZC3H11A T/A exon 1440N Tolerated Benign 7 9.74E- 0.876 53 8.81E- 0.237	2	38318262	'	MICALL1	C/A	exon	R285R			6	6.15E- 03	-0.825	68	9.52E- 01	-0.008
203816588 - <i>ZC3H11A</i> T/A exon 1440N Tolerated Benign 7 9.74E- 0.876 53 8.81E- 0.237 0.37		44274143	1	AARS2	GAA/-	intron				12	6.99E- 03	0.730	79	1.56E- 01	0.161
		203816588	'	ZC3H11A	T/A	exon	I440N	Tolerated	Benign	~	9.74E- 03	0.876	23	3.81E- 02	-0.237

ufts	rs ^c	0.02	0.092	0.301	0.0	0.18	0.069	0.334	0.066	0.071	(pənu
xenogr:	<i>P</i> alue	.70E- 01	.54E- 01	.26E- 02	.67E 01	.43E 01	.76E 01	.35E 03	.92E 01	.63E- 01	(Contr
All	Z	58 8.	58 4.	58 1.	58 4.	58 1.	58 5.	58 5.	5 5.	58 5.	
•.		9 166	963 (954 (945 (934 (915 (905 6	395 (391 (
ancer	ŗ	0.9	0.0	0.9	-0.9	-0.0	0.9	-0.0	0.8	0.8	
Breast c	<i>P</i> value	1.52E- 05	4.97E- 04	8.67E- 04	1.30E- 03	2.06E- 03	3.92E- 03	5.13E- 03	6.53E- 03	7.11E- 03	
	z	~	~	~	~	~	~	~	~	~	
of functional ect	PolyPhen2		NA						NA		
Prediction off	SIFT		NA						NA		
Amino acid change			F318L						Q157K, Q498K		
Location		intron	exon	ncRNA_intron	ncRNA_exon	intron	5'UTR, 5'UTR	intron	exon, exon	intron	
Allele Ref./Variant		AG/TC	GT/AC	AGG/-	-/TCTCTGATA TGCCATCC	TT/GA	-/GGGCGTGC AGGACGC	-/C	TC/GA	-/GTT	
Gene		PKD1L2	OR51M1	TRPM2-AS	LINC01621	SCARF2	ACADVL, DLG4	CYFIP1	RPP21, TRIM39-RPP21	NAA35	
rsID ^b		rs386792906	rs369353765	rs765207853	ı	rs35574298	I	I	rs35287137	rs368374310	
Position ^a		81253642	5411579	45843709	80513567	20784908	7123256	22960698	30314566	88631383	
Chr		16	11	21	9	22	17	15	9	6	
Drug		XTM									

Drug	Chr	Position ^a	$rsID^b$	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction o eff	f functional ect	B	treast car	ıcer	A	ll xenogi	rafts
								SIFT	PolyPhen2	z	P value	rs ^c	z	P value	rs ^c
VCR	21	30714976	1	BACH1	C/G	exon	A678G	Tolerated	Benign	10	3.71E- 05	0.945	72	9.04E- 01	0.015
	12	75816816	rs59277111	GLIPR1L2	-/CAA	exon	D239_K240insQ	NA	NA	10	6.11E- 04	-0.888	76	8.66E- 03	-0.299
	10	97397087	1	ALDH18A1	A/C	exon	V26G	Deleterious	Possibly damaging	9	7.47E- 04	0.978	51	7.94E- 01	-0.037
	19	56001803	rs5828624	SSC5D	-/CCAAGCAA	intron				6	9.35E- 04	-0.900	72	6.99E- 01	-0.046
	2	47277207	rs71416119	TTC7A	CA/AG	intron				10	1.44E- 03	0.859	76	3.70E- 01	0.104
	15	78581889	1	WDR61	-/T	intron				10	2.46E- 03	0.838	76	1.19E- 02	0.287
	3	44803116	rs3082548	KIAA1143	AAGACAG/-	5'UTR				10	3.69E- 03	-0.820	76	2.53E- 01	-0.133
	8	145692652	ı	KIFC2	A/G	exon	S133G	Tolerated	Benign	~	4.86E- 03	0.907	48	4.39E- 01	0.114
	ъ	1246263	1	SLC6A18	-/ecccc	3'UTR				10	4.99E- 03	0.805	76	4.36E- 01	0.091
	12	29908581	rs3830194	TMTC1	-/TTGTT	intron				10	6.95E- 03	-0.787	75	3.31E- 01	-0.114
	7	100361392	rs3215395	ZAN	-/C	intron				10	7.45E- 03	0.783	76	4.66E- 01	0.085
	9	32713619	rs146449814	HLA-DQA2	C/A	exon	P128H	Deleterious	Probably damaging	6	8.51E- 03	0.807	62	6.81E- 01	0.053
	8	134292515	1	NDRG1	9/-	intron				10	8.87E- 03	-0.772	76	8.60E- 01	0.021
	1	203816588	1	ZC3H11A	T/A	exon	I440N	Tolerated	Benign	ъ	9.01E- 03	0.962	50	2.41E- 01	0.169
	6	138523408	rs34000956	GL T6D1	-/T	intron				10	9.25E- 03	-0.770	76	6.23E- 01	0.057
														(Con	tinued)

Drug	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction o effe	f functional ect	В	reast can	cer	ЧI	xenogra	ufts
								SIFT	PolyPhen2	z	<i>P</i> value	r_{s}^{c}]	Z	P value	rs ^c
VLB	6	33037639	rs386699859	HLA-DPA1	GC/AT	exon	A42M	NA	NA	10	6.37E- 04	0.886	6 1	.96E- 01	0.150
	9	32948287	1	BRD2	TT/GC	intron				10	6.49E- 04	0.886	20	.54E- 01	0.052
	15	89864317	rs2307433	570d	-/CTAC	intron				10	3.72E-	0.819 7	2	01E- 01	0.148
	11	94322352	rs386756343	PIWIL4	AG/TA	exon	Q327L	NA	NA	10	4.26E-	0.813 7	6 4	L.89E- 01	0.081
	20	62492851	1	ABHD16B	G/C	5'UTR				×	4.66E- 03	0.873	55	1.77E- 01	0.020
	12	16055927	rs71042275	STRAP	T/-	3'UTR				10	5.38E-	0.801	6 3	62E- 01	0.106
	2	211421454	1	CPS1	-/CTT	exon	I5_K6insL	NA	NA	10	6.08E- 03	0.794 7	7 67	.54E- 01	0.036
	1	9324725	1	Н6РD	C/A	exon	P736T	Deleterious	Probably damaging	5	8.08E- 03	0.964	15	.12E- 01	0.057
	21	47754410	rs57603484	PCNT	A/G	exon	S5G	Tolerated	Possibly damaging	6	9.17E- 03	0.803	75 1	.77E- 01	0.158
	15	89864318	,	POLG	-/TACC	intron				10	9.98E-	0.765	6 4	l.25E- 01	0.093
Based	un GR	Ch 37 genome	e accembly												

based on URCh3/ genome assembly.

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

5FU, 5-fluorouracil; ACNU, nimustine; ADR, adriamycin; CPM, cyclophosphamide; DDP, cisplatin; MMC, mitomycin C; MTX, methotrexate; VCR, vincristine; VLB, vinblastine; ncRNA, ⁵ Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug. noncoding RNA; del, deletion; ins, insertion; fs, frameshift.

https://doi.org/10.1371/journal.pone.0239614.t011

All xenografts	$ P value r_s^c$	9 7.29E- 0.235 02	6 6.63E- 0.051	10	5 5.29E0.074 01	5 5.29E0.074 01 6 3.56E0.107	5 5.29E -0.074 6 3.56E -0.107 6 1.54E -0.165 6 1.54E -0.165	5 5.29E -0.074 6 3.56E -0.107 6 0.1 -0.165 6 1.54E -0.165 0.1 0.1 -0.165 6 3.34E 0.244 7 0.2 0.244	5 5.29E -0.074 6 3.56E -0.107 7 01 01 6 1.54E -0.165 7 7.85E -0.032	5 5.29E -0.074 6 3.56E -0.107 7 01 -0.107 6 1.54E -0.165 7 3.34E 0.244 7 7.85E -0.032 7 2.98E -0.120 7 2.98E -0.120	5 5.29E -0.074 6 3.56E -0.107 7 0.1 0.1 7 2.34E 0.165 7 7.85E -0.032 7 2.98E -0.033 7 2.98E -0.120 7 4.75E -0.033 7 0.1 0.1 7 0.1 0.033 7 0.1 0.033	5 5.29E -0.074 6 3.56E -0.107 7 0.1 -0.165 7 1.54E -0.165 7 2.34E -0.244 7 2.85E -0.032 7 4.75E -0.032 7 4.75E -0.033 7 4.03E -0.093 7 4.03E -0.093	5 5.29E -0.074 6 3.56E -0.107 7 01 -0.107 6 1.54E -0.165 7 3.34E 0.244 7 7.85E -0.032 7 2.98E -0.120 7 4.75E -0.033 7 4.75E -0.083 7 4.75E -0.083 7 4.03E -0.097 7 4.75E -0.083 7 4.47E -0.088 7 4.47E -0.088	5 5.29E -0.074 6 3.56E -0.107 7 01 -0.107 8 3.56E -0.107 9 01 -0.165 6 1.54E -0.165 7 3.34E 0.244 02 01 -0.032 7 7.85E -0.032 01 01 -0.083 7 4.75E -0.083 01 01 -0.097 7 4.75E -0.083 01 01 -0.088 7 4.75E -0.083 01 01 -0.097 7 4.75E -0.083 6 8.74E -0.018 01 01 -0.018 01 01 -0.018 01 01 -0.018	5 5.29E -0.074 6 3.56E -0.107 7 01 -0.107 6 1.54E -0.165 7 3.34E 0.244 02 01 -0.032 7 7.85E -0.032 7 2.98E -0.033 7 4.75E -0.083 01 01 -0.083 7 4.75E -0.083 01 01 -0.097 7 4.47E -0.083 01 01 -0.033 6 8.74E -0.018 01 01 -0.033 6 01 -0.033 7 4.10E 0.233 7 4.10E 0.233	5.29E -0.074 6 3.56E -0.107 7 01 -0.107 6 1.54E -0.165 7 3.34E 0.244 02 01 -0.032 7 2.9E -0.032 7 2.9E -0.032 7 01 -0.032 7 4.75E -0.033 7 4.75E -0.033 7 4.75E -0.033 7 4.47E -0.033 6 8.74E -0.038 01 01 -0.233 7 4.10E 0.233 7 5.13E 0.233 7 5.13E 0.233	5 5.29E -0.074 6 3.56E -0.107 6 1.54E -0.107 6 1.54E -0.107 6 3.36E -0.107 7 3.56E -0.107 7 3.34E 0.244 01 0.2 0.032 7 2.98E -0.032 01 0.1 -0.083 7 4.75E -0.083 01 0.1 -0.097 7 4.47E -0.083 01 0.1 -0.033 6 8.74E -0.038 01 0.1 -0.233 7 4.10E 0.233 7 5.13E 0.233 8 6.80E 0.051	5 5.29E -0.074 6 3.56E -0.107 7 01 -0.107 6 1.54E -0.165 7 3.34E 0.244 02 0.01 -0.032 7 7.85E -0.032 01 0.2 -0.032 7 2.98E -0.120 01 0.1 -0.032 7 4.75E -0.033 7 4.03E -0.097 01 01 -0.033 6 8.74E -0.088 01 01 -0.233 7 4.10E 0.233 8 6.80E 0.051 01 0.2 0.051 7 9.81E 0.003
ncer	r _s ^c N	-0.833 59	-0.753 70	-	-0.771 7	0.741 7	-0.771 7/ 0.741 7/ -0.735 7/	-0.771 7 0.741 7 -0.735 7 0.713 7	-0.771 77 0.741 77 -0.735 77 0.713 77 0.713 77 0.713 77	-0.771 77 0.741 77 -0.735 77 0.713 77 0.713 77 0.713 77 -0.755 77	-0.771 77 0.741 77 -0.735 77 0.713 70 0.713 70 0.713 77 -0.755 77 -0.746 77	-0.771 77 0.741 77 -0.735 77 -0.735 77 0.713 77 0.713 77 -0.755 77 -0.734 77	-0.771 77 -0.741 77 -0.735 77 -0.735 77 -0.788 77 -0.755 77 -0.755 77 -0.734 77 -0.733 77	-0.771 7 -0.741 7 -0.735 7 -0.735 7 0.713 7 0.713 7 -0.735 7 -0.746 7 -0.734 7 -0.733 7 -0.733 7 -0.733 7	-0.771 7 -0.741 7 -0.735 7 -0.735 7 -0.755 7 -0.746 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.713 7 -0.713 7 -0.713 7	-0.771 7 -0.771 7 -0.735 7 -0.735 7 -0.713 7 -0.735 7 -0.7346 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.713 7 -0.713 7 -0.713 7 -0.710 7	-0.771 7 -0.771 7 -0.735 7 -0.735 7 -0.713 7 -0.735 7 -0.734 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.733 7 -0.713 7 -0.713 7 -0.713 7 -0.713 7 -0.713 7	-0.771 7 -0.771 7 -0.735 7 -0.735 7 -0.746 7 -0.733 7 -0.713 7 -0.713 7 -0.754 7
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effect	PolyPher	ls Possibly damagin						NA	NA NA	NA NA	NA NA NA	NA N	V V V V	V V V V V V V V V V V V V V V V V V V	V V V V V V V V V V V V V V V V V V V	V V V V V V V V V V V V V V V V V V V	NA NA NA Benign	NA NA NA Benign
Ű	SIFT	Deleterious						NA	NA NA	NA NA	NA NA NA	VA VA VA	VA VA VA	VA V	NA AN A	NA AN A	NA NA NA NA Tolerated Tolerated	NA NA NA NA NA
change		Q309K						A42M	A42M Q907Pfs*23	A42M Q907Pfs*23	A42M A42M Q907Pfs*23 E271Gfs*19	A42M A42M Q907Pfs*23 E271Gfs*19	A42M A42M Q907Pfs*23 E271Gfs*19	A42M A42M Q907Pfs*23 E271Gfs*19	A42M A42M Q907Pfs*23 E271Gfs*19	A42M A42M Q907Pfs*23 E271Gfs*19	A42M A42M Q907Pfs*23 E271Gfs*19 E271Gfs*19	A42M A42M Q907Pfs*23 E271Gfs*19 E271Gfs*19
LOCALIOII		exon	intron		intron	intron intron	intron intron intron	intron intron intron exon	intron intron intron exon exon	intron intron intron exon exon exon	intron intron intron exon exon exon intron	intron intron intron exon exon intron exon	intron intron intron exon exon exon intron intron intron	intron intron intron exon exon intron intron intron intron	intron intron exon exon exon intron intron intron intron	intron intron exon exon exon intron intron intron intron intron intron intron	intron intron intron exon exon exon intron intron intron intron exon exon exon exon exon exon exon ex	intron intron intron exon exon intron exon intron intron intron exon exon exon exon intron exon
Allele Ket./Variant		C/A	-/C		-/ATATG	-/ATATG G/A	-/ATATG G/A CA/TG	-/ATATG G/A CA/TG GC/AT	-/ATATG G/A CA/TG GC/AT -/G	-/ATATG G/A CA/TG GC/AT -/G	-/ATATG G/A G/AT GC/AT -/G -/G	-/ATATG G/A G/AT CA/TG GC/AT -/G -/G -/T	-/ATATG G/A G/AT CA/TG GC/AT -/G -/G -/G -/T	-/ATATG G/A G/A CA/TG GC/AT -/G -/G -/G -/T -/T -/T -/T	-/ATATG G/A G/AT CA/TG GC/AT -/G -/G -/G -/G -/T A -/T A -/TT AATCAGCC	-/ATATG G/A G/AT GC/AT -/G -/G -/G -/G -/T -/T -/T -/T -/T -/TTAAT -/ATCAGCC -/A	-/ATATG G/A G/AT CA/TG CA/TG GC/AT -/G -/G -/G -/G -/A -/T -/T CA -/A -/T -/T CA -/A -/A -/A -/A -/A -/A -/A -/A -/A -/	-/ATATG G/A G/AT CA/TG GC/AT -/G -/G -/G -/G -/G -/A A T -/T CA CA CA CA CA CA CA CA CA CA CA CA CA
		0 ISLNI	CENPJ		MTRF1L	MTRF1L MAP2K3	MTRFIL MAP2K3 PSD4	MTRFIL MAP2K3 PSD4 HLA-DPA1	MTRFIL MAP2K3 PSD4 HLA-DPAI AARS	MTRFIL MAP2K3 PSD4 HLA-DPAI AARS JAK3	MTRFIL MAP2K3 PSD4 HLA-DPAI AARS JAK3 JAK3 CHGA	MTRFIL MAP2K3 PSD4 HLA-DPAI AARS JAK3 JAK3 CHGA DUSP26	MTRFIL MAP2K3 PSD4 PSD4 HLA-DPAI AARS JAK3 JAK3 CHGA CHGA DUSP26 KIF1B	MTRFIL MAP2K3 PSD4 PSD4 HLA-DPAI AARS JAK3 JAK3 CHGA DUSP26 KIF1B KIF1B	MTRFIL MAP2K3 PSD4 PSD4 HLA-DPAI AARS JAK3 JAK3 JAK3 CHGA DUSP26 KIFIB KIFIB NAH11 TEC	MTRFIL MAP2K3 PSD4 PSD4 HLA-DPAI AARS JAK3 JAK3 JAK3 CHGA CHGA DUSP26 DUSP26 DUSP26 KIFIB DUSP26 TEC TEC	MTRFIL MAP2K3 PSD4 HLA-DPAI AARS JAK3 JAK3 CHGA CHGA CHGA DUSP26 DUSP26 KIF1B DUSP26 TEC PRH1-PRR4 PRH1-PRR4 PRH1-PRR4	MTRFIL MAP2K3 PSD4 HLA-DPAI AARS JAK3 JAK3 CHGA DUSP26 CHGA DUSP26 KIF1B DUSP26 FRF1B PRH1-PRR4 PRH1-PRR4 PRH1-PRR4 NENF
		1	1	rs149540839		rs62057674	rs62057674 rs1553408097	rs62057674 rs1553408097 rs386699859	rs62057674 rs1553408097 rs386699859	rs62057674 rs1553408097 rs386699859 rs397839895	rs62057674 rs1553408097 rs386699859 rs386699859 rs397839895	rs62057674 rs1553408097 rs386699859 rs397839895 rs397839895	rs62057674 rs1553408097 rs386699859 rs397839895 rs397839895 rs397839895	rs62057674 rs1553408097 rs386699859 rs396699859 rs397839895 rs397839895 rs3831405 rs57952953	rs62057674 rs1553408097 rs386699859 rs396699859 rs397839895 rs397839895 rs3831405 rs57952953 rs11282767	rs62057674 rs1553408097 rs386699859 rs386699859 rs397839895 rs397839895 rs3831405 rs57952953 rs11282767 rs11282767	rs62057674 rs1553408097 rs386699859 rs396699859 rs397839895 rs397839895 rs397839895 rs37952953 rs57952953 rs11282767	rs62057674 rs1553408097 rs386699859 rs386699859 rs397839895 rs3831405 rs57952953 rs11282767 rs11282767
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;		8	13	6		17	2	17 17 2 6 6	17 2 6 6 16	17 2 6 6 16 19	17 2 6 6 16 19 19 14	17 2 6 6 6 16 19 19 14 8	17 2 2 16 19 19 19 18 8	17 2 2 6 6 6 16 19 19 14 14 18 8 8 8 8	17 2 6 6 6 16 19 14 14 8 8 8 8 8 8 8 8 8 8 8 8 8	17 17 2 2 2 16 16 19 17 1 18 8 8 8 11 1 12 1	17 2 2 16 16 19 14 1	17 17 2 2 16 19 12 19 19 19 11 12 13
		5FU							ACNU	ACNU	ACNU	ACNU	ACNU	ACNU	ACNU	ACNU	ACNU ADR	ACNU ADR

PLOS ONE

(Continued)

Modelia Modelia <t< th=""><th>Chr</th><th>Position^a</th><th>rsID^b</th><th>Gene</th><th>Allele Ref./Variant</th><th>Location</th><th>Amino acid change</th><th>Prediction</th><th>of functional fect</th><th></th><th>Gastric ca</th><th>ncer</th><th></th><th>All xeno</th><th>grafts</th></t<>	Chr	Position ^a	rsID ^b	Gene	Allele Ref./Variant	Location	Amino acid change	Prediction	of functional fect		Gastric ca	ncer		All xeno	grafts
0606011 560603497 KLU0 ·1A intron intron </th <th></th> <th></th> <th></th> <th></th> <th></th> <th></th> <th></th> <th>SIFT</th> <th>PolyPhen2</th> <th>z</th> <th>P value</th> <th>rs^c</th> <th>z</th> <th>P value</th> <th>rs^c</th>								SIFT	PolyPhen2	z	P value	rs ^c	z	P value	rs ^c
30007473 v. MARCP v/ABCCP v/ABCCP vector 1180106*8 NA 12 0486 0486 0470 0480 0470 0480 0470 0410		103664311	rs36083487	KLF10	-/A	intron				12	1.01E- 03	0.823	79	7.18E- 01	0.041
238738 · · · · · · · · · · · · · · · · · · ·		232087473	1	ARMC9	Ð/-	exon	I180Dfs*8	NA	NA	12	1.48E- 03	-0.808	79	3.73E- 01	-0.102
1143759 ····· ····· ····· ····· ····· ····· ······ ······ ······ ······ ······ ····· ······ ······ ······· ······· ······· ········ ·········· ············ ··············		29287938	1	C2orf71	-/TGC	intron				12	8.74E- 03	0.717	79	3.15E- 01	0.114
7065155 ··· STOXI ··· 10000860 ···· MHBPI ···· ···· ···· ···· ···· ···· ···· ···· ···· ···· ···· ···· ···· ···· ···· ···· ···· ··· ··· <td< td=""><td></td><td>71433759</td><td>1</td><td>SDK2</td><td>9/-</td><td>intron</td><td></td><td></td><td></td><td>12</td><td>8.82E- 03</td><td>0.716</td><td>79</td><td>5.20E- 01</td><td>0.074</td></td<>		71433759	1	SDK2	9/-	intron				12	8.82E- 03	0.716	79	5.20E- 01	0.074
3830131 FTHAMIS2 C/T exon C/3 <c< th=""> FT FT<td></td><td>70652195</td><td>1</td><td>STOX1</td><td>Ш/-</td><td>intron</td><td></td><td></td><td></td><td>12</td><td>9.68E- 03</td><td>0.710</td><td>79</td><td>8.40E- 01</td><td>-0.023</td></c<>		70652195	1	STOX1	Ш/-	intron				12	9.68E- 03	0.710	79	8.40E- 01	-0.023
1364.064 ··· <i>STKLD1 T/C</i> inton <i>T/C inton inton</i>		28203133	rs774954578	THEMIS2	C/T	exon	C43C			12	1.94E- 03	-0.796	17	9.65E- 01	-0.005
1000866··· $MH13$ C/T exon $E_1/91E$ <		136246047	1	STKLD1	T/C	intron				8	5.43E- 03	0.866	68	6.02E- 01	0.064
113639 ··· RHBDF1 G/A exon S1365 ··· 8 6.95 0.82 0.82 0.80 1892787 s1993872 5AX01 T/C $3UTR$ $3UTR$ $3UTR$ $3UTR$ $3UTR$ 2026 0.836 0.85 0.85 0.730 7 3.05 1892787 s1993872 $5AY01$ T/C UTC $3UTR$ $3UTR$ $3UTR$ 10000 10000 100000 $1000000000000000000000000000000000000$		10209869	1	MYH13	C/T	exon	E1791E			10	5.56E- 03	-0.799	53	4.45E- 01	-0.107
189038725 takes SAXO1 T/C 3UTR 3UTR MAD T G00 G00<		113639	1	RHBDF1	G/A	exon	S136S			8	6.95E- 03	-0.854	50	7.22E- 01	-0.052
1896059 \$\$386811653 \$\$IRPA \$\$CTAC \$\$exon \$\$V132T \$\$NA\$ \$\$NA\$ <td></td> <td>18927887</td> <td>rs199938722</td> <td>SAXOI</td> <td>I/C</td> <td>3'UTR</td> <td></td> <td></td> <td></td> <td>12</td> <td>6.99E- 03</td> <td>-0.730</td> <td>17</td> <td>3.46E- 02</td> <td>-0.241</td>		18927887	rs199938722	SAXOI	I/C	3'UTR				12	6.99E- 03	-0.730	17	3.46E- 02	-0.241
5350339 1534924760 SOATZ GC/TT exon A202A T.46E 0.73 5.29E 0.73 5.29E 0.73 0.73 0.73 0.73 0.73 0.73 0.73 0.71 0.73 0.71 0.73 0.71 0.73 0.73 0.71 0.73 0.71 0.73 0.71 0.73 0.73 0.71 0.73 </td <td></td> <td>1896059</td> <td>rs386811663</td> <td>SIRPA</td> <td>GT/AC</td> <td>exon</td> <td>V132T</td> <td>NA</td> <td>NA</td> <td>12</td> <td>7.15E- 03</td> <td>0.729</td> <td>27</td> <td>4.27E- 01</td> <td>0.092</td>		1896059	rs386811663	SIRPA	GT/AC	exon	V132T	NA	NA	12	7.15E- 03	0.729	27	4.27E- 01	0.092
40886465 HIPK4 T/G exon Y4785 Deleterious Probaby damaging 11 7.35- 0.75 7 2 00-1 01 <th< td=""><td></td><td>53509339</td><td>rs34924760</td><td>SOAT2</td><td>GC/TT</td><td>exon</td><td>A202A</td><td></td><td></td><td>12</td><td>7.46E- 03</td><td>0.726</td><td>27</td><td>5.29E- 01</td><td>0.073</td></th<>		53509339	rs34924760	SOAT2	GC/TT	exon	A202A			12	7.46E- 03	0.726	27	5.29E- 01	0.073
45649504 rs72019726 <i>PPIR37</i> -/GTAA intron intron 1 286E -0.750 7 7.09E -0.10 02 -0.10 -0.10 <t< td=""><td></td><td>40886465</td><td>1</td><td>HIPK4</td><td>T/G</td><td>exon</td><td>Y478S</td><td>Deleterious</td><td>Probably damaging</td><td>11</td><td>7.73E- 03</td><td>-0.751</td><td>74</td><td>2.60E- 01</td><td>-0.133</td></t<>		40886465	1	HIPK4	T/G	exon	Y478S	Deleterious	Probably damaging	11	7.73E- 03	-0.751	74	2.60E- 01	-0.133
147695284 $rs3217238$ $LOCI02546294$ $-/TCA$ $ncRNA_intron$ $ncRNA_intron$ 12 8 0.72 7 $6.63E$ 0.722 7 $6.63E$ 0.702 7 $6.63E$ 0.702 0.712 7 $6.63E$ 0.702 0.712 <		45649504	rs72019726	PPP1R37	-/GTAA	intron				11	7.86E- 03	-0.750	75	7.09E- 02	-0.210
655318 - DNHD1 G/A exon B971E N B 2.9E- 0.845 61 2.79E- 0.141 131185358 rs56988335 MIR1268A -/TGTCCACTG ncRNA_intron E971E N N 12 8.45E- 0.719 77 0.09 131185358 rs56988335 MIR1268A -/TGTCCACTG ncRNA_intron E2716/rs19 NA 12 8.45E- 0.719 77 0.19 93399168 - - - B - NA 12 8.45E- 0.717 77 0.17 93399168 - - B - NA 12 8.67E- 0.717 77 0.17 0.1 1895950 rs386811661 SIRPA CCT/GTC exon D95_L96 NA 12 8.67E- 0.714 77 0.11 1895950 rs386811661 SIRPA CCT/GTC exon D95_L96 NA 12 9.07E 0.11 0.113		147695284	rs3217238	LOC102546294	-/TCA	ncRNA_intron				12	8.07E- 03	-0.722	17	6.63E- 03	-0.307
131185358 rs6988335 <i>MIR1268A</i> -/TGTCCACTG ncRNA_intron ncm 12 8.45E 0.719 77 5.50E 0.009 9339168 - - - -/A exon E271Gfs*19 NA 12 8.67E 0.717 77 0.19E 0.179 9339168 - - -/A exon E271Gfs*19 NA NA 12 8.67E 0.717 77 0.19E 0.179 1895950 rs386811661 SIRPA CCT/GTC exon D95_L96 NA NA 12 8.67E 0.714 77 0.19E 0.113 1895950 rs386811661 SIRPA CCT/GTC exon D95_L96 NA 12 9.07E 0.714 77 3.30E 0.113 33451023 - DUSP26 -/T intron D95_L96 NA 12 9.07E 77 1.48E 0.016 33451023 - DUSP26 -/T intron		6555318	1	DNHDI	G/A	exon	E971E			8	8.29E- 03	-0.845	61	2.79E- 01	0.141
93399168 CHGA -/A exon E271Gfs*19 NA NA 12 8.67E -0.717 77 1.19E -0.175 1895950 rs386811661 SIRPA CCT/GTC exon D95_L96 NA 12 9.07E- 0.714 77 1.19E- -0.113 1895950 rs386811661 SIRPA CCT/GTC exon D95_L96 NA 12 9.07E- 0.714 77 3.30E- 0.113 33451023 DUSP26 -/T intron 12 9.07E- 0.708 77 1.48E- 0.166		131185358	rs56988335	MIR1268A	-/TGTCCACTG	ncRNA_intron				12	8.45E- 03	0.719	17	5.50E- 01	0.069
1895950 rs386811661 SIRPA CCT/GTC exon D95_L96 NA 12 9.076- 0.714 77 3.30E- 0.113 33451023 - DUSP26 -/T intron delinsES 0.1 2.98E- -0.708 77 1.48E- 0.166 33451023 - DUSP26 -/T intron 12 9.98E- -0.708 77 1.48E- -0.166		93399168	1	CHGA	-/A	exon	E271Gfs*19	NA	NA	12	8.67E- 03	-0.717	17	1.19E- 01	-0.179
33451023 - <i>DUSP26</i> -/T intron 12 9.98E- 0.708 77 1.48E- 0.166 01		1895950	rs386811661	SIRPA	CCT/GTC	exon	D95_L96 delinsES	NA	NA	12	9.07E- 03	0.714	27	3.30E- 01	0.113
		33451023	1	DUSP26	-μ	intron				12	9.98E- 03	-0.708	77	1.48E- 01	-0.166

Table 12. (Continued)

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ografts	le r _s ^c	0.139	0.039	- 0.039	0.109	0.053	0.125	- 0.003	- 0.008	- 0.028	0.091	- 0.033	0.207	- 0.007	- 0.162	0.253	- 0.080	- 0.107	0.051	0.085
All xend	P valu	2.33E 01	7.34E 01	7.36E 01	3.40E 01	6.42E 01	3.45E 01	9.80E 01	9.44E 01	8.06E 01	4.25E 01	7.75E 01	9.03E 02	9.54E 01	1.86E 01	3.77E 02	5.76E 01	3.87E 01	6.82E 01	4.89E
	z	75	62	62	79	79	59	79	79	12	79	79	68	68	68	68	51	68	68	68
ncer	rs ^c	-0.821	0.753	0.741	-0.734	0.734	-0.818	-0.720	-0.720	0.716	-0.716	-0.711	-0.835	-0.830	0.817	-0.812	-0.955	0.800	-0.791	-0.776
astric ca	P value	3.63E- 03	4.73E- 03	5.87E- 03	6.62E- 03	6.62E- 03	7.00E- 03	8.31E- 03	8.31E- 03	8.82E- 03	8.82E- 03	9.59E- 03	1.39E- 03	1.58E- 03	2.13E- 03	2.38E- 03	3.01E- 03	3.12E- 03	3.70E- 03	4.96E-
	z	10	12	12	12	12	6	12	12	12	12	12	11	11	11	11	9	11	11	11
of functional ffect	PolyPhen2			NA	NA		NA				NA	NA			NA	NA			NA	NA
Prediction	SIFT			NA	NA		NA				NA	NA			NA	NA			NA	NA
Amino acid change	l			T617A	M59Hfs*32		G27X				P393Q	G1938E			V320Gfs*20	G1938E	V711V		A251 delins GAAPPAPPP	N236del
Location		intron	intron	exon	exon	intron	exon	intron	intron	intron	exon	exon	intron	intron	exon	exon	exon	intron	exon	exon
Allele Ref./Variant		-/GCAGCGG	-/A	TT/CC	5/-	Т/-	Т/Э	-/GCTGAGACGG	-/CTGAGACGGG	-/AGCCCCAGCT GGGCGAGGC	TG/CT	GC/AA	CA/TG	-/TTGAAA	-/A	GC/AA	G/A	-/A	-/GCGGAGGGGGGG GTGGCGCCGCCC	TTT/-
Gene		SELENOW	GPNMB	FAM214A	OR6P1	MUC16	TXNDC15	SYCEI	SYCEI	CTSB	BTNL2	LRRK1	PSD4	KIF1B	UGGTI	LRRK1	PCDHA5	GFPT1	C2orf81	PKD1L2
rsID ^b		rs34940677	1	rs386783993	1	1	1	rs3831169	ı	rs145929462	rs28362676	rs386787404	rs1553408097	rs3831405	I	rs386787404	1	rs57860122	rs768089535	rs796089514
Position ^a		48282078	23293095	52901283	158533221	9000065	134210196	135368490	135368491	11705381	32362702	101606889	113953976	10384177	128878011	101606889	140203493	69597065	74642267	81242149
Chr		19	~	15	-	19	5	10	10	~	9	15	2	-	2	15	5	5	5	16
Drug		MMC	1	1	1	1	1	1	<u>.</u>	1	<u>.</u>	<u>.</u>	MTX	1	<u>.</u>	1	1	1	1	

xenografts	value r _s ^c	.89E- 0.047 01	.72E- 0.104 01	.27E- 0.041 01	.27E- 0.092 01	.77E0.033 01	.47E0.073 01		.73E0.049 01	73E0.049 01 0.186 01 0.186	73E- -0.049 01 01 11E- 0.186 01 0.75E- 01 0.065	7.73E0.049 01 -0.186 01 -0.186 01 -0.065 01 -0.065 01 -0.076 01 -0.076	7.73E0.049 01	7.73E0.049 01 1.11E- 0.186 01 01 1.75E- 0.065 01 1.77E0.076 01 37E0.039 01 01 01 01 01	7.3E0.049 01 1.11E- 0.186 01 1.75E- 0.065 01 1.77E0.076 01 3.760.039 01 01 3.37E0.007 01 01 01 01 01 01 01 01 01 01 01 01 01	7.3E- -0.049 01 0.186 01 0.186 7.5E- 0.065 01 0.17E 01 0.01 37E- -0.076 01 0.01 37E- -0.039 01 0.01 37E- -0.030 01 0.01 253E- -0.007 01 0.01 01 0.01 01 0.01 01 0.010 01 0.011 01 0.010 01 0.011 01 0.011 01 0.011 01 0.011 01 0.011 01 0.011	7.3E- -0.049 01 01 01 0.186 01 0.186 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.07 01 0.07 01 0.01 01 0.01 01 0.035 01 0.035 025 -0.034 02 -0.234	73E- -0.049 01 0.186 01 0.186 01 0.005 01 0.005 01 0.005 01 0.005 01 0.005 01 0.005 01 0.005 01 0.007 01 0.033 01 0.007 01 0.007 01 0.007 01 0.01 01 0.1 01 0.1 01 0.1 01 0.1 01 0.1 02 0.055 01 0.055 01 0.055	7.3E- -0.049 01 0.186 01 0.186 01 0.01 .75E- 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.076 01 0.039 01 0.031 .375E- -0.039 01 0.01 01 0.1 01 0.1 02 .0.351 02 .0.055 01 .0.055 01 .0.055 01 .0.055 01 .0.055 01 .0.055 01 .0.055 01 .0.085 01 .0.085 01 .0.085	7.31 -0.049 01 0.116 01 0.186 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.065 01 0.076 01 0.039 01 0.039 01 0.031 01 0.031 02 -0.034 02 -0.035 01 -0.035 01 -0.035 01 -0.035 01 -0.035 01 -0.035 01 -0.035 01 -0.035 01 -0.037 01 -0.027
NI	N N	76 6	76 3	76 7	76 4	76 7	71 5	-	76 6	76 6 75 1	76 66 75 1 76 5	76 6 75 1 76 5 76 5 76 5	76 5 75 1 76 5 76 5 76 5 76 5	76 6 75 1 75 1 76 5 76 5 76 5 76 5	76 5 1 75 1 75 1 76 5 7 5 1 76 7 7 5 1 76 5 7 7 5 1	76 6 75 1 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5	76 6 75 1 75 1 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 76 5 77 78	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	76 6 75 1 75 1 76 5 76 5 76 5 76 7 77 7 76 7 77 7 76 7 76 7 76 7
tcer	rs ^c	0.935	0.931	0.924	0.917	-0.818	-0.859		-0.801	0.752	0.752	0.801 0.752 0.750	0.801 0.752 0.750 0.758 -0.748	0.801 0.752 0.750 0.748 -0.716 0.712	0.801 0.752 0.750 0.748 -0.716 0.712 0.712	0.801 0.752 0.750 0.750 -0.748 -0.716 0.712 -0.709 -0.789	0.801 0.752 0.750 0.750 0.748 0.716 0.712 0.712 0.712 0.712	0.801 0.752 0.750 0.750 0.716 0.716 0.712 0.712 0.712 0.712 0.709	0.801 0.752 0.750 0.756 0.716 0.716 0.712 0.712 0.712 0.709 0.709 0.709	0.801 0.752 0.750 0.748 0.716 0.715 0.712 0.709 -0.769 -0.769 -0.774 0.774 0.774
tric can	value	32E- 06	08E- 05	73E- 05	67E- 05	15E 03	43E-		73E-	73E- 03 76E- 03	73E- 03 76E- 03 96E- 03 03	73E- 03 76E- 03 96E- 96E- 03 03 03	73E- 73E- 76E- 03 03 03 19E- 19E- 78E- 03 03 03 03 03 03 03 03 03 03 03 03 03	738- 7785- 766- 766- 766- 03 03 03 03 03 03 03 03 03 03	735- 7765- 766- 766- 03 03 03 03 03 03 03 03 03 03 03 03 03	73E- 73E- 03 03 03 03 04 04 05 03 03 04 19E- 19E- 103 19E- 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103 103	73E- 03	735- 735- 03 03 03 03 03 03 03 03 03 03 03 03 03 0	73E- 73B- 03 03 76B- 76B- 03 03 19E- 19E- 19E- 78B- 19E- 78B- 19E- 19E- 19E- 19E- 19B- 19B- <	73E- 73B- 03 03 03 03 96B- 96B- 96B- 19B- 96B- 19B- 93B- 19B- 19B- 19B- 103 33B- 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03 103 03
Gas	Å N	12 8.	12 1.	12 1.	12 2.	12 1.	10 1.		12 1.	12 1. 12 4.	12 1. 12 4. 12 4.	12 1. 12 4. 12 4. 12 4. 12 5.	12 1. 12 4. 12 4. 12 5. 12 8.	12 1. 12 4. 12 5. 12 8. 12 9.	12 1.2 4. 1.2 12 1.2 4. 4. 1.2 12 1.2 5. 5. 4. 1.2 12 1.2 8. 5. 9. 9. 9.	12 1. 12 4. 12 4. 12 5. 12 9. 12 9.	12 12<	12 12<	12 12 4 1 12 11 12 <td>12 12 4 1 12 11 12 12 13 13 14 14 15 15 16</td>	12 12 4 1 12 11 12 12 13 13 14 14 15 15 16
of functional fect	PolyPhen2							-	NA	NA NA	NA NA	NA NA NA	NA NA NA	NA NA NA	NA NA NA NA	NA NA NA NA	NA NA NA NA	NA NA NA NA	NA NA NA NA Benign	NA NA NA NA NA Benign
Prediction eff	SIFT								NA	NA NA	NA NA	NA NA NA	NA NA NA	NA NA NA	NA NA NA NA	NA NA NA NA	NA NA NA	NA NA NA	NA NA NA NA Deleterious	NA NA NA NA NA Deleterious
Amino acid change									G39Rfs*15	G39Rfs*15 D470fs*0	G39Rfs*15 D470fs*0	G39Rfs*15 D470fs*0 E130del	G39Rfs*15 D470fs*0 E130del	G39Rfs*15 D470fs*0 E130del	G39Rfs*15 D470fs*0 E130del E130del X478delinsX	G39Rfs*15 D470fs*0 E130del B130del X478delinsX	G39Rfs*15 D470fs*0 E130del B130del X478delinsX	G39Rfs*15 D470fs*0 E130del X478delinsX	G39Rfs*15 D470fs*0 E130del E130del X478delinsX X14R	G39Rfs*15 D470fs*0 E130del E130del X478delinsX S114R
Location		intron	intron	intron	intron	intron	intron		exon	exon exon	exon exon intron	exon exon intron exon	exon exon intron exon intron	exon exon intron exon intron intron	exon exon intron exon intron intron exon	exon exon intron exon intron intron exon exon	exon exon intron exon intron intron exon exon intron	exon exon intron exon intron intron exon intron 3'UTR intron	exon exon intron exon intron intron exon intron intron exon exon exon exon	exon exon intron exon intron intron intron intron exon exon exon exon intron
Allele Ref./Variant		G/À	G/A	T/G	A/G	9/-	-/CTTAT		-/G	-/C	-/G -/C -/CTGAGGC	-/G -/C -/CTGAGGC TTC/-	-/G -/C -/CTGAGGC TTC/- -/C	-/G -/C -/CTGAGGC -/C -/C -/C GC/AG	-/G -/C -/CTGAGGC TTC/- -/C GC/AG GC/AG	-/G -/C -/CTGAGGC -/C GC/AG GC/AG -/CT -/CT -/CT	-/G -/C -/CTGAGGC TTC/- -/C GC/AG GC/AG -/CT -/CT -/CT	-/G -/C -/CTGAGGC -/C -/C GC/AG GC/AG GC/AG -/CT -/CT -/CT -/A	-/G -/C -/C TTC/- TTC/- -/C GC/AG GC/AG GC/AG GC/AG -/C -/C -/C -/C -/C -/C	-/G -/C -/CTGAGGC TTC/- TTC/- -/C GC/AG GC/AG GC/AG -/A -/A -/GTAA -/GTAA T/G CCT/-
Gene		MAP2K3	MAP2K3	MAP2K3	MAP2K3	DIP2A	IRAKIBPI		ACTL9	ACTL9 PLOD3	ACTL9 PLOD3 SLC24A1	ACTL9 PLOD3 SLC24A1 TRIM52	ACTL9 PLOD3 SLC24A1 TRIM52 USP19	ACTL9 PLOD3 SLC24A1 TRIM52 USP19 SLC45A4	ACTL9 PLOD3 SLC24A1 TRIM52 USP19 SLC45A4 MYBPH	ACTL9 PLOD3 SLC24A1 TRIM52 USP19 SLC45A4 MYBPH PSD4	ACTL9 PLOD3 SLC24A1 TRIM52 USP19 SLC45A4 MYBPH PSD4 PSD4 CCDC86	ACTL9 PLOD3 SLC24A1 TRIM52 USP19 USP19 SLC45A4 MYBPH PSD4 PSD4 CCDC86 PPP1R37	ACTL9 PLOD3 SLC24A1 TRIM52 USP19 USP19 SLC45A4 MYBPH PSD4 PSD4 PSD4 CCDC86 PPP1R37 FARP1	ACTL9 PLOD3 SLC24A1 TRIM52 USP19 USP19 SLC45A4 MYBPH PSD4 PSD4 PSD4 CCDC86 PPP1R37 FARP1 FARP1 TMEM63B
rsID ^b		rs73302038	rs66486636	rs73302043	rs73302034	rs74854320	rs66520304		1		- - rs111310111	- - rs111310111 rs3073543	- - rs111310111 rs3073543	- - rs111310111 rs3073543 rs386730897	- rs111310111 rs3073543 rs386730897	- rs111310111 rs3073543 rs386730897 rs386730897 rs1553408097	- rs111310111 rs3073543 rs386730897 rs386730897 rs1553408097 rs1553408097	- rs111310111 rs3073543 rs386730897 rs386730897 rs1553408097 rs1553408097 rs1553408097	- rs111310111 rs3073543 rs3073543 rs386730897 rs1553408097 rs1553408097 rs1553408097 rs1553408097 rs1553408097 rs1553408097	- rs111310111 rs3073543 rs3073543 rs386730897 rs386730897 rs386730897 rs386730897 rs386730897 rs386730897 rs1553408097 rs10537719 rs10537719
Position ^a		21215682	21215637	21215700	21215643	47985555	79595168		8808938	8808938 100853907	8808938 100853907 65931909	8808938 100853907 65931909 180687440	8808938 100853907 65931909 180687440 49148887	8808938 100853907 65931909 180687440 49148887 142231944	8808938 100853907 65931909 180687440 49148887 112231944 112231944 203137787	8808938 100853907 65931909 180687440 180687440 49148887 142231944 142231944 142231944 113953976	8808938 100853907 65931909 180687440 49148887 49148887 142231944 142231944 142231944 113953976 06617832	8808938 100853907 65931909 180687440 49148887 49148887 142231944 142231944 142231944 113953976 113953976 60617832 60617832	8808938 100853907 65931909 180687440 4914887 4914887 142231944 142231944 142231944 142231944 1422319787 113953976 60617832 60617832 88866915	8808938 100853907 65931909 180687440 49148887 49148887 112231944 142231944 112231944 113953976 60617832 60617832 45649504 45649504 88896915 98896915
Chr		17	17	17	17	21	6		19	7	19 7 15	19 7 15 5	19 3 5 15	8 3 5 15 7 19	19 15 7 19 1 8 3 5 5 7 19	19 2 1 1 1	19 19 11 2 1 8 3 5 5 1 1 1 11 1 8 8 3 3 5 5 1 <td>19 1</td> <td>19 1</td> <td>19 1</td>	19 1	19 1	19 1
Drug		VCR									<u> </u>					ALL	ALB	ALL	ALL	ALL

^b rsID from the NCBI database of genetic variation (dbSNP). "-", this variant is not identified in dbSNP.

5FU, 5-fluorouracil; ACNU, nimustine; ADR, adriamycin; CPM, cyclophosphamide; DDP, cisplatin; MMC, mitomycin C; MTX, methotrexate; VCR, vincristine; VLB, vinblastine; ncRNA, ⁵ Spearman correlation coefficient had been calculated to estimate positive (resistant) or negative (sensitive) correlation between the VAF and sensitivity to each anticancer drug. noncoding RNA; del, deletion; ins, insertion; fs, frameshift.

Table 12. (Continued)

statistically significant variants or perform a subgroup analysis for all tumor types. Second, the results need to be confirmed using a larger number of samples, along with a functional analysis of the identified genes.

In conclusion, using whole-exome sequencing and a PDX model, we identified 162 genetic variants as possible susceptibility factors for sensitivity to one or more of the nine tested cyto-toxic anticancer drugs. This method and the results presented herein may contribute to the development of personalized treatments for the prescription of optimal chemotherapy regimens. Although the underlying mechanisms should be further investigated using a larger number of clinical samples and molecular analysis, we propose that our findings may help to contribute to understanding the mechanisms of chemoresistance and chemosensitivity, and aid in the improved prognosis and quality of life for patients with cancer.

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