Pathogenic mutations identified by a multimodality approach in 117 Japanese Fanconi anemia patients

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Supplemental Data

Supplemental Note: Case 66 Presentation

A 23-year-old man was admitted to the hospital because of a giant mediastinal tumor. He was born to unrelated healthy parents and had no significant past medical history. Physically, he presented with short stature (155cm, -2.7SD) and severe microcephaly (49.4cm, -5SD). Hematological and bone marrow examination were normal (neutrophils, 3.88×10^9/L; hemoglobin, 14.1 g/dl; hematocrit, 42.2%; reticulocytes, 1.5%; platelets 244×10^9/L). On the basis of a CT scan and mediastinal tumor biopsy, he was diagnosed with mediastinal T-cell lymphoblastic lymphoma (T-LBL). Induction chemotherapy consisting of cyclophosphamide, vincristine, daunorubicin, prednisone, and l-asparaginase was performed and he suffered from severe sepsis due to prolonged pancytopenia. After hematological recovery, a mitomycin C induced chromosomal breakage test showed an increased rate of chromosomal breakage. Targeted-exome sequencing identified the splice site mutation c.475+1G>A and the missense mutation c.7847C>T in *FANCD1 (BRCA2)*. He was diagnosed as Fanconi anemia.

After induction chemotherapy, the T-LBL achieved a partial remission but he was found to have adenosquamous lung cancer and bilateral renal tubule-papillary adenoma. He underwent focal radiation therapy to the mediastinal lesion and reduced-intensity chemotherapy. However, he relapsed and died of T-LBL 15 months after the initial chemotherapy.

Supplemental Table 1. Summary of 22 FA-related genes

FA gene (Alternative name)	OMIM No.	FA /FBOC/FA-like*	Functions	Distribution of genetic subtyping		
				this study (2018)	Rockefeller Fanconi Anemia Mutation Database (2008)[1]	National Network of the Italian Association of Pediatric Hematology and Oncology (2014)[2]
FANCA	607139	FA	Component of the FA core complex	58%	57%	85%
FANCB	300515	FA	Component of the FA core complex	3%	0.9%	1%
FANCC	613899	FA	Component of the FA core complex	1%	15%	3%
FANCD1 (BRCA2)	600185	FBOC, FA	HR repair, mediator fucntion for RAD51, Protects stalled replication fork	2%	2.9%	0%
FANCD2	613984	FA	Monoubiquitylated by the FA core complex, Forms ID2 complex, Regulates the DNA damage response	0%	3.9%	2%
FANCE	613976	FA	Component of the FA core complex	1%	2.3%	0%
FANCF	613897	FA	Component of the FA core complex	1%	2%	0%
FANCG	602956	FA	Component of the FA core complex	25%	11.0%	9%
FANCI	611360	FA	Monoubiquitylated by the FA core complex, Forms ID2 complex, Regulates the DNA damage response	2%	1.7%	0%
FANCJ (BRIP1)	605882	FBOC,FA	HR repair, DNA helicase	0%	2.4%	0%
FANCL	608111	FA	Component of the FA core complex, E3 ubiquitin ligase	0%	0.1%	0%
FANCM	609644	FBOC, FA-associated**	Component of the FA core complex, DNA translocase	0%	0%	0%
FANCN (PALB2)	610355	FBOC, FA	HR repair, Facilitaties BRCA2 function	1%	0.8%	0%
FANCO (RAD51C)	602774	FBOC, FA-like	RAD51 paralog, HR repair, Stabilizes RAD51 nucleoprotein filament	0%	—***	0%
FANCP (SLX4)	613278	FA	Resolutes Holliday junctions, Nuclease regulation, Incises DNA-ICL damage	2%	—***	0%
FANCQ (ERCC4)	133520	FA	DNA repair nuclease	0%	—***	0%
FANCR (RAD51)	179617	FA-like	HR repair, Protects stalled replication fork	0%	—***	—***
FANCS (BRCA1)	113705	FBOC, FA-like	HR repair, Promotes RAD51 recruitment	0%	—***	—***
FANCT (UBE2T)	610538	FA	E2 ubiquitin-conjugating enzyme	2%	—***	—***
FANCU (XRCC2)	600375	FA-like	RAD51 paralog, HR repair, Stabilizes RAD51 nucleoprotein filament	0%	—***	—***
FANCV (MAD2L2/REV7)	604094	FA	Translesion DNA synthesis	0%	—***	—***
FANCW (RFWD3)	614151	FA	HR repair, E3 ligase	0%	—***	—***

* FA-like genes cause a chromosome fragility syndrome with FA-related malformations but without bone marrow failure.[3]

** FANCM was originally thought to be FA gene but it turned out that biallelic FANCM mutations do not cause any overt FA phenotype[4] but early onset cancer.[5, 6]

*** These genes were not identified at the time of the publication.[1, 2]

FA, Fanconi anemia; FBOC, familial breast and ovarian cancer; HR, homologous recombination; ID2 complex, FANCD2-FANCI heterodimer; ICL, interstrand crosslink

Supplemental Table 2. The list of *FANC* genes variants, *ALDH2* genotype, and clinical information in 117 Japanese patients with Fanconi anemia

Family No.	Case No.	Sex	Affected gene	Methods for identifying the mutations	Mutation 1		Mutation 2		*ALDH2* genotype*	Hematological/Oncologic phenotype	Age at BMF/Malignancy diagnosis (months)	FA-features**	VACTERL-H	References /Comments
					cDNA	Protein	cDNA	Protein						
1	1	F	*FANCA*	WES	c.2870G>A	p.W957X	**c.2723_2725TCT>GCC**	p.LS908_909RP	GG	MDS	121/335	Yes	No	5, 10
2	2	M	*FANCA*	WES	c.1303C>T	p.R435C	c.1303C>T	p.R435C	GA	AML	unknown/289	Yes	No	8, 10
3	3	F	*FANCA*	WES	c.2170A>C	p.T724P	c.505G>T	p.E169X	GA	MDS	unknown/143	Yes	No	8, 10
4	4	M	*FANCA*	WES, MLPA	c.2546delC	p.S849FfsX40	ex30del	-	GA	AA	37	Yes	No	5, 8, 10
5	5	F	*FANCA*	WES	c.1303C>T	p.R435C	c.4168-1G>C	aberrant splicing	GA	AA	26	Yes	No	5, 8, 10
6	6-1	M	*FANCA*	WES, MLPA	c.3765+1G>T	aberrant splicing	ex30del	-	GG	AA	96	Yes	No	5, 10
	6-2	F	*FANCA*	MLPA, Sanger	c.3765+1G>T	aberrant splicing	ex30del	-	GG	AA	51	Yes	No	5, 10
7	7	M	*FANCA*	WES	c.4240_4241delAG	p.S1414LfsX10	c.2602-1G>A	aberrant splicing	GG	AML	41/115	Yes	No	5, 7, 8, 10
8	8	M	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.2546delC	p.S849FfsX40	GA	AA	38	Yes	No	5, 10
9	9-1	M	*FANCA*	MLPA, Sanger	c.978_979delGA	p.Q326HfsX12	ex30del	-	GA	MDS	60/192	Yes	No	5, 7, 10
	9-2	F	*FANCA*	MLPA, Sanger	c.978_979delGA	p.Q326HfsX12	ex30del	-	GG	AA	92	Yes	No	5, 7, 10
	9-3	F	*FANCA*	WES, MLPA	c.978_979delGA	p.Q326HfsX12	ex30del	-	GG	AA	45	Yes	No	5, 7, 10
10	10-1	F	*FANCA*	WES	c.2602-2A>T	aberrant splicing	c.4198C>T	p.R1400C	GG	AA	120	Yes	No	5, 8, 10
	10-2	F	*FANCA*	WES	c.2602-2A>T	aberrant splicing	c.4198C>T	p.R1400C	GA	AA	48	Yes	No	5, 10
11	11	M	*FANCA*	WES, MLPA	c.3568C>T	p.Q1190X	**ex11-15dupli**	-	GG	AA	297	Yes	No	5, 10
12	12	M	*FANCA*	WES	c.3919_3920insT	p.Q1307LfsX6	c.2546delC	p.S849FfsX40	GG	MDS	144/145	Yes	No	5, 8, 10
13	13	F	*FANCA*	WES, MLPA	c.2546delC	p.S849FfsX40	ex1-28del	-	GG	MDS	72/72	Yes	No	5, 7, 8, 10
14	14	M	*FANCA*	WES	c.2602-1G>A	aberrant splicing	c.2602-2A>T	aberrant splicing	GG	AA	134	Yes	No	5, 8, 10
15	15	F	*FANCA*	WES	**c.1007-2A>G**	aberrant splicing#	c.4168-2A>G	aberrant splicing	GA	MDS	48/60	No	No	10
16	16	F	*FANCA*	WES	c.2546delC	p.S849FfsX40	**c.3965T>G**	p.V1322G	GA	AA	24	Yes	No	5, 7, 8, 10
17	17	F	*FANCA*	WES	**c.190_191insT**	p.E65RfsX6	**c.190_191insT**	p.E65RfsX6	not examined	CVID	No	Yes	No	9
18	18-1	M	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	**c.4042_4043insC**	p.I1348TfsX77	AA	MDS	0/12	Yes	Yes	5, 7, 10
	18-2	F	*FANCA*	Targeted-seq	c.2546delC	p.S849FfsX40	**c.4042_4043insC**	p.I1348TfsX77	GG	AML	69/69	Yes	No	7, 8, 10
19	19-1	M	*FANCA*	Targeted-seq	**c.283+2T>C**	aberrant splicing #	c.2730_2731delCT	p.W911DfsX31	GA	AA	30	Yes	No	7, 8, 10
	19-2	M	*FANCA*	Targeted-seq	**c.283+2T>C**	aberrant splicing#	c.2730_2731delCT	p.W911DfsX31	GA	AA	16	Yes	No	7, 8, 10
20	20	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	**c.3781_3785delTTCTT**	p.F1261LfsX15	AA	MDS	7/7	Yes	No	5, 10
21	21	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	**c.3931_3932delAG**	p.S1311X	GA	AA	21	Yes	No	5, 7, 10
22	22-1	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.4168-2A>G	aberrant splicing	GG	AA	106	Yes	No	5, 10
	22-2	M	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.4168-2A>G	aberrant splicing	GA	MDS	28/168	Yes	No	5, 10
23	23	F	*FANCA*	Sanger	c.2593delA	p.I879LfsX24	c.2840C>G	p.S947X	GA	AA/HNSCC	53/457	No	No	5, 10
24	24	M	*FANCA*	Sanger, MLPA	c.2546delC	p.S849FfsX40	ex1-3del	-	GA	AA	22	Yes	No	5, 7, 10
25	25	M	*FANCA*	Sanger	c.2602-2A>T	aberrant splicing	c.2527T>G	p.Y843D	GG	AA	78	Yes	No	5, 7, 10
26	26	M	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.2546delC	p.S849FfsX40	GG	AA	114	Yes	No	5, 10
27	27	F	*FANCA*	Sanger	c.2602-2A>T	aberrant splicing	c.2602-2A>T	aberrant splicing	GG	AML	62/311	Yes	No	5, 7, 10
28	28	F	*FANCA*	Sanger	c.4124-4125delCA	p.T1375SfsX49	c.2290C>T	p.R764W	GG	AML	156/156	Yes	No	5, 10
29	29	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	**c.3765+827_3814del**	-	GG	AA	72	Yes	No	5, 10

30	30	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.2546delC	p.S849FfsX40	GG	AA	70	Yes	Yes	5, 7, 10
31	31	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	**c.1567-1G>A**	aberrant splicing#	GG	MDS	82/82	Yes	No	5, 10
32	32	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.3720_3724del	p.E1240DfsX36	GG	AA	88	Yes	No	5, 10
33	33	M	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.3720_3724del	p.E1240DfsX36	GG	MDS	68/105	Yes	No	10
34	34	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.2602-1G>A	aberrant splicing	GA	AML	60/282	Yes	No	10
35	35-1	M	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.2546delC	p.S849FfsX40	AA	MDS	0/4	Yes	No	7, 10
	35-2	M	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	c.2546delC	p.S849FfsX40	GA	AA	21	Yes	No	7, 10
36	36	M	*FANCA*	Sanger	c.44_69del	p.P15RfsX13	c.2170A>C	p.T724P	GG	MDS/HNSCC	108/348/348	Yes	No	5, 10
37	37	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	**c.3295C>T**	p.Q1099X	GG	MDS	49/189	Yes	Yes	5, 10
38	38	M	*FANCA*	WES, MLPA	c.2840C>G	p.S947X	ex24-28del	-	GG	AA	60	Yes	No	5, 10
39	39	F	*FANCA*	Targeted-seq, MLPA	**c.462T>G**	p.Y154X	ex6del	-	GG	AA	unknown	No	No	8
40	40	F	*FANCA*	Sanger	c.2602-1G>A	aberrant splicing	not detected	-	GG	AML	108/384	Yes	No	5, 10
41	41	M	*FANCA*	Sanger, MLPA	c.2546delC	p.S849FfsX40	ex37del	-	GG	AML	136/176	Yes	No	5, 10
42	42	F	*FANCA*	WES, MLPA	c.4199G>C	p.R1400P	ex16_17del	-	GG	AML	61/61	Yes	No	5, 10
43	43	M	*FANCA*	Targeted-seq	c.2T>C	p.M1T	c.15G>A	p.W5X	GA	AA	37	No	No	7, 10
44	44	M	*FANCA*	Targeted-seq	c.2546delC	p.S849FfsX40	c.2972delT	p.F991SfsX35	GA	MDS	50/73	Yes	No	7, 10
45	45-1	F	*FANCA*	Targeted-seq, MLPA	ex1_43del	-	ex19_29 del	-	GG	AA	108	Yes	No	10
	45-2	F	*FANCA*	Targeted-seq, MLPA	ex1_43del	-	ex19_29 del	-	GA	AA	12	Yes	No	10
46	46	F	*FANCA*	WES, MLPA	c.2546delC	p.S849FfsX40	ex1_5del	-	GG	AA	unknown	Yes	No	New case
47	47	M	*FANCA*	WES	c.2546delC	p.S849FfsX40	c.2546delC	p.S849FfsX40	GA	unknown	unknown	unknown	unknown	8
48	48	F	*FANCA*	WES	c.4015_4017del CTC	p.L1339del	c.3638_3639delCT	p.P1213RfsX64	GA	unknown	unknown	unknown	unknown	8
49	49	F	*FANCA*	WES	c.2546delC	p.S849FfsX40	c.2546delC	p.S849FfsX40	GG	AA	71	Yes	No	8
50	50	F	*FANCA*	WES	c.2546delC	p.S849FfsX40	c.2546delC	p.S849FfsX40	GG	AA	71	Yes	No	8
51	51	F	*FANCA*	WES	**c.1464C>A**	p.Y488X	**c.1464C>A**	p.Y488X	GG	AA	157	Yes	No	8
52	52	F	*FANCA*	WES, MLPA	c.978_979delGA	p.Q326HfsX12	ex30del	-	GG	unknown	unknown	unknown	unknown	New case
53	53	M	*FANCA*	WES, MLPA	c.978_979delGA	p.Q326HfsX12	ex30del	-	GG	unknown	unknown	unknown	unknown	New case
54	54	F	*FANCA*	WES	c.2546delC	p.S849FfsX40	not detected	-	not examined	AA	85	No	No	8
55	55	F	*FANCA*	WES, MLPA	c.2546delC	p.S849FfsX40	ex30_31del	-	GA	AA	80	Yes	No	8
56	56	F	*FANCA*	WES	**c.2316+2T>A**	aberrant splicing#	not detected	-	GG	AA	59	Yes	No	8
57	57	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	not detected	-	GA	MDS	unknown/234	No	No	New case
58	58	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	not detected	-	GG	AA	82	Yes	No	New case
59	59	F	*FANCA*	Sanger	c.2546delC	p.S849FfsX40	not detected	-	GG	AA	80	Yes	No	New case
60	60	M	*FANCB*	aCGH, Sanger	**chrX: g.14730104-14904216del**	complete loss	-	-	GG	AA	58	Yes	Yes	5 Current study identified causative FA gene mutation.
61	61	M	*FANCB*	aCGH, Sanger	c**hrX: g.14810970-14932973del**	complete loss	-	-	GA	MDS	24/51	Yes	Yes	5, 10 Current study identified causative FA gene mutation.
62	62	M	*FANCB*	WES, RNA-seq	**c.1497G>T**	aberrant splicing (p.S500AfsX14)	-	-	GG	AA	96	Yes	No	5 Current study identified causative FA gene mutation.
63	63	M	*FANCB*	WES	**c.516G>A**	p.W172X	-	-	not examined	unknown	unknown	unknown	unknown	8

64	64	F	*FANCC*	WGS	**c.1154+5G>A**	aberrant splicing (p.S386X)	**c.1154+5G>A**	aberrant splicing (p.S386X)	GG	AA	40	Yes	Yes	5, 7 Current study identified causative FA gene mutation.
65	65	F	*FANCD 1*	WES	**c.517-2A>G**	aberrant splicing#	**c.6952C>T**	p.R2318X	GG	immmature teratoma	No/9	Yes	No	8
66	66	M	*FANCD 1*	Targeted-seq	**c.475+1G>A**	aberrant splicing#	**c.7847C>T**	p.S2616F	GA	T-LBL, Lung cancer	No/508	No	No	8
67	67	F	*FANCE*	WES	**c.419T>C**	p.L140P	**c.648delC**	p.R219DfsX77	GA	CVID	No	Yes	No	9
68	68	F	*FANCF*	WES	**c.484_485delCT**	p.L162DfsX103	**c.66C>A**	p.Y22X	GG	AA	43	Yes	No	8
69	69	M	*FANCG*	WES	c.1066C>T	p.Q356X	c.307+1G>C	aberrant splicing	GA	MDS	12/61	Yes	Yes	5, 8, 10
70	70	F	*FANCG*	WES	c.1066C>T	p.Q356X	c.1066C>T	p.Q356X	GG	AA	66	Yes	No	5, 8, 10
71	71	M	*FANCG*	WES	c.1066C>T	p.Q356X	c.1066C>T	p.Q356X	GG	AA	72	Yes	No	5, 8, 10
72	72-1	M	*FANCG*	WES	c.91C>T	p.Q31X	c.307+1G>C	aberrant splicing	GG	AA	27	Yes	No	5, 8, 10
	72-2	M	*FANCG*	WES	c.91C>T	p.Q31X	c.307+1G>C	aberrant splicing	GG	AA	60	Yes	No	5, 10
73	73-1	M	*FANCG*	WES	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GA	AA	48	Yes	Yes	5, 8, 10
	73-2	M	*FANCG*	WES	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GA	AA	39	Yes	No	5, 10
74	74	F	*FANCG*	WES	c.1066C>T	p.Q356X	c.1066C>T	p.Q356X	GA	AA	24	Yes	No	5, 7, 8, 10
75	75	M	*FANCG*	WES	**c.907_908del**	p.L303GfsX5	c.307+1G>C	aberrant splicing	GA	AA	21	Yes	No	5, 8, 10
76	76	F	*FANCG*	WES	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GA	AA	69	Yes	No	5, 8, 10
77	77	F	*FANCG*	WES	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GA	AA	18	Yes	No	5, 7, 8, 10
78	78	F	*FANCG*	WES	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GG	MDS	78/78	Yes	No	7, 8, 10
79	79	F	*FANCG*	WES	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GG	AA	37	Yes	No	10
80	80	M	*FANCG*	WES	c.1066C>T	p.Q356X	c.1066C>T	p.Q356X	GA	AA	24	Yes	No	10
81	81	F	*FANCG*	WES	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	AA	MDS	0/18	Yes	No	7, 8, 10
82	82	M	*FANCG*	WES	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	AA	AA	0	Yes	No	7, 8, 10
83	83	F	*FANCG*	WES	c.1066C>T	p.Q356X	c.307+1G>C	aberrant splicing	GA	MDS	24/38	Yes	No	7, 8, 10
84	84	M	*FANCG*	Sanger	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GA	AA	38	Yes	No	5, 10
85	85	M	*FANCG*	Sanger	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GA	AA	36	Yes	No	5, 10
86	86	F	*FANCG*	Sanger	c.307+1G>C	aberrant splicing	c.1066C>T	p.Q356X	GG	AA	50	Yes	No	5, 7, 10
87	87	M	*FANCG*	Sanger	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GG	AA	28	No	No	5, 7, 10
88	88	M	*FANCG*	WES	**c.1386delC**	p.W463GfsX55	**c.1637-15G>A**	VUS	GG	MDS	69/120	Yes	No	5, 7, 10
89	89	F	*FANCG*	Sanger	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GG	AA	46	Yes	No	7, 10
90	90	M	*FANCG*	WES	c.194delC	p.P65LfsX7	c.307+1G>C	aberrant splicing	GG	AA	29	Yes	No	8
91	91	F	*FANCG*	WES	c.1066C>T	p.Q356X	c.307+1G>C	aberrant splicing	GG	AA/MDS	30/66	unknown	unknown	8
92	92	M	*FANCG*	Sanger	c.194delC	p.P65LfsX7	c.194delC	p.P65LfsX7	GA	AA	unknown	Yes	No	New case
93	93	M	*FANCG*	Sanger	c.307+1G>C	aberrant splicing	c.307+1G>C	aberrant splicing	GG	AA	67	Yes	No	New case
94	94	M	*FANCG*	Sanger	c.1066C>T	p.Q356X	not detected	-	GG	MDS	unknown/396	Yes	No	New case
95	95	M	*FANCG*	Sanger	c.1066C>T	p.Q356X	c.1066C>T	p.Q356X	GA	AA	75	Yes	No	New case
96	96	M	*FANCI*	WES	**c.158-2A>G**	aberrant splicing (p.S54FfsX5)	**c.288G>A**	aberrant splicing (p.C56FfsX8)	GA	AA	7	Yes	Yes	5 Current study identified causative FA gene mutations.
97	97	M	*FANCI*	WES	**c.3346_3347ins T**	p.S1116FfsX16	**c.3006+3A>G**	aberrant splicing#	GA	AA	15	Yes	No	5, 7 Current study identified causative FA

														gene mutations.
98	98	M	*FANCN*	WES, RNA-seq	**c.3350+5C>T**	aberrant splicing (p.G1068VfsX5)	**c.3350+5C>T**	aberrant splicing (p.G1068VfsX5)	GA	Wilms tumor	No/12	Yes	No	10 Current study identified causative FA gene mutation.
99	99-1	M	*FANCP*	WES	**c.343delA**	p.S115AfsX11	**c.343delA**	p.S115AfsX11	AA	MDS	0/4	Yes	Yes	5, 7, 8, 10
	99-2	F	*FANCP*	Sanger	**c.343delA**	p.S115AfsX11	**c.343delA**	p.S115AfsX11	GG	Normal	No at 8	Yes	No	7
100	100	F	*FANCT*	WES, aCGH	c.4C>G	p.Q2E	chr1:g202288583_202309772del	complete loss	GA	AA	87	Yes	No	5, 6, 7
101	101	M	*FANCT*	WES	c.4C>G	p.Q2E	c.180+5G>A	aberrant splicing (p.E37RfsX49)	GA	AML	41/41	Yes	No	5, 6, 10
102	102	F	unclassified	-	-	-	-	-	GA	MDS	12/108	Yes	No	5, 10
103	103-1	F	unclassified	-	-	-	-	-	not examined	unknown	unknown	unknown	unknown	New case
	103-2	M	unclassified	-	-	-	-	-	not examined	unknown	unknown	unknown	unknown	New case
104	104	M	unclassified	-	-	-	-	-	not examined	unknown	unknown	unknown	unknown	New case

Novel mutations (not included in the Rockefeller FA mutation data base) are indicated in boldface type.

#Effects of these splicing mutations are unverified.

*The *ALDH2* wild type and the inactivating mutation (p.Glu504Lys) allele is referred to as G and A, respectively[8]. **FA features include physical abnormalities such as short stature, malformations or skin pigmentation.

AA, aplastic anemia; *ALDH2*, aldehyde dehydrogenase-2; AML, acute myeloid leukemia; aCGH, array-CGH; BMF, bone marrow failure; CVID, common variable immunodeficiency; FA, Fanconi anemia; HNSCC, head and neck squamous cell carcinoma; MDS, myelodysplastic syndrome; MLPA, multiplex ligation-dependent probe amplification; T-LBL, T cell lymphoblastic lymphoma; VACTERL-H, VACTERL-H, vertebral anomalies, anal atresia, cardiac anomalies, tracheal-esophageal fistula, esophageal atresia, renal structural abnormalities, limb anomalies, and hypocephalus; VUS, variation of unknown significant; WES, whole-exome sequencing; WGS, whole-genome sequencing

References in this table (the numbers are the same as the references in the main text)

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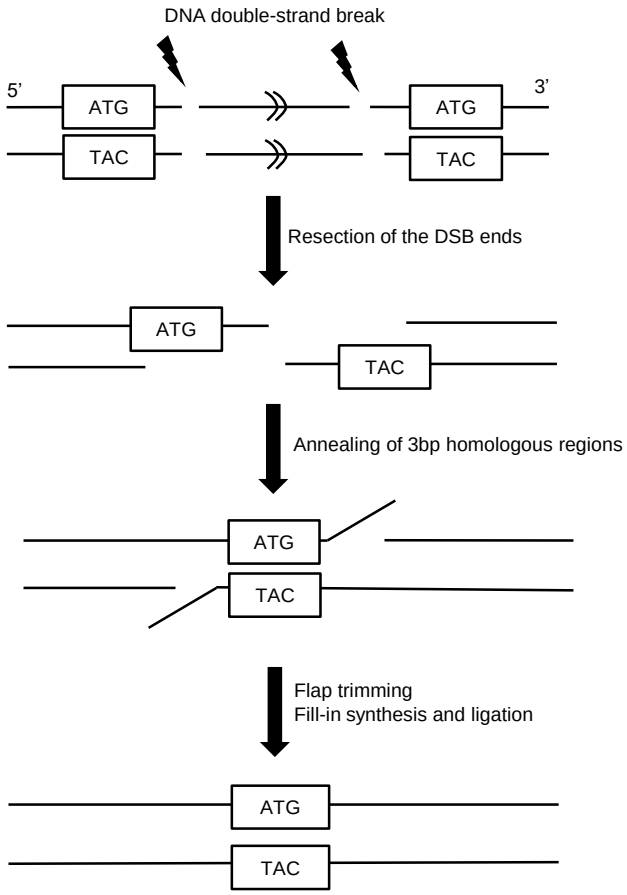
Supplemental Table 3. 55 different *FANCA* variants detected in Japanese FA-A patients

DNA change	Location	Effect	Comments	No. of alleles	No. of patients	No.of unrelated families
missense mutations						
c.2T>C	exon 1	p.M1T	known mutation	1	1	1
c.1303C>T	exon 14	p.R435C	known mutation	3	2	2
c.2170A>C	exon 24	p.T724P	known mutation	2	2	2
c.2290C>T	exon 25	p.R764W	known mutation	1	1	1
c.2527T>G	exon 27	p.Y843D	known mutation	1	1	1
c.2723_2725TCT>GCC	exon 28	p.LS908_909RP	novel mutation	1	1	1
c.3965T>G	exon 40	p.V1322G	novel mutation	1	1	1
c.4198C>T	exon 42	p.R1400C	known mutation	2	2	1
c.4199G>C	exon 42	p.R1400P	known mutation	1	1	1
nonsense mutations						
c.15G>A	exon 1	p.W5X	known mutation	1	1	1
c.462T>G	exon 5	p.Y154X	novel mutation	1	1	1
c.505G>T	exon 5	p.E169X	known mutation	1	1	1
c.1464C>A	exon 15	p.Y488X	novel mutation	2	1	1
c.2840C>G	exon 29	p.S947X	known mutation	2	2	2
c.2870G>A	exon 30	p.W957X	known mutation	1	1	1
c.3295C>T	exon 33	p.Q1099X	novel mutation	1	1	1
c.3568C>T	exon 36	p.Q1190X	known mutation	1	1	1
small insertions/deletions						
c.44-69del	exon 1	p.P15RfsX40	known mutation	1	1	1
c.190_191insT	exon 3	p.E65RfsX6	novel mutation	2	1	1
c.978_979delGA	exon 11	p.Q326HfsX12	known mutation	5	5	3
c.2546delC	exon 27	p.S849FfsX40	known mutation	41	33	30
c.2593delA	exon 27	p.I879LfsX24	novel mutation	1	1	1
c.2730_2731delCT	exon 28	p.W911DfsX31	known mutation	2	2	1
c.2972delT	exon 30	p.F991SfsX35	known mutation	1	1	1
c.3638_3639delCT	exon 37	p.P1213RfsX64	known mutation	1	1	1
c.3720_3724 del	exon 37	p.E1240DfsX36	known mutation	2	2	2
c.3781_3785delTTCTT	exon 38	p.F1261LfsX15	novel mutation	1	1	1
c.3919_3920insT	exon 39	p.Q1307LfsX6	novel mutation	1	1	1
c.3931-3932delAG	exon 39	p.S1311X	novel mutation	1	1	1
c.4015_4017delCTC	exon 41	p.L1339del	known mutation	1	1	1
c.4042_4043insC	exon 41	p.I1348TfsX77	novel mutation	2	2	1
c.4124-4125delCA	exon 41	p.T1375SfsX49	known mutation	1	1	1
c.4240_4241delAG	exon 42	p.S1414LfsX10	known mutation	1	1	1
splicing mutations						
c.283+2T>C	intron 3	aberrant splicing	novel mutation	2	2	1
c.1007-2A>G	intron 11	aberrant splicing	novel mutation	1	1	1
c.1567-1G>A	intron 16	aberrant splicing	novel mutation	1	1	1
c.2316+2T>A	intron 25	aberrant splicing	novel mutation	1	1	1
c.2602-2A>T	intron 27	aberrant splicing	known mutation	6	5	4
c.2602-1G>A	intron 27	aberrant splicing	known mutation	4	4	4
c.3765+1G>T	intron 37	aberrant splicing	known mutation	2	2	1
c.4168-1G>C	intron 41	aberrant splicing	known mutation	1	1	1
c.4168-2A>G	intron 41	aberrant splicing	known mutation	3	3	2
large deletions						
ex1-3 del	—	—		1	1	1
ex1-5 del	—	—		1	1	1
ex1-28 del	—	—		1	1	1
ex1-43 del	—	—		2	2	1
ex6 del	—	—		1	1	1
ex16-17 del	—	—		1	1	1
ex19-29 del	—	—		2	2	1
ex24-28 del	—	—		1	1	1
ex30 del	—	—		8	8	5
ex30-31 del	—	—		1	1	1
ex37 del	—	—		1	1	1
c.3765+827_3814 del	intron 37-exon 38	—	novel mutation	1	1	1
large duplication						
ex11-15 dupi	—	—		1	1	1
Total				130		

Supplemental Table 4. Seven different *FANCG* variants found in Japanese FA-G patients

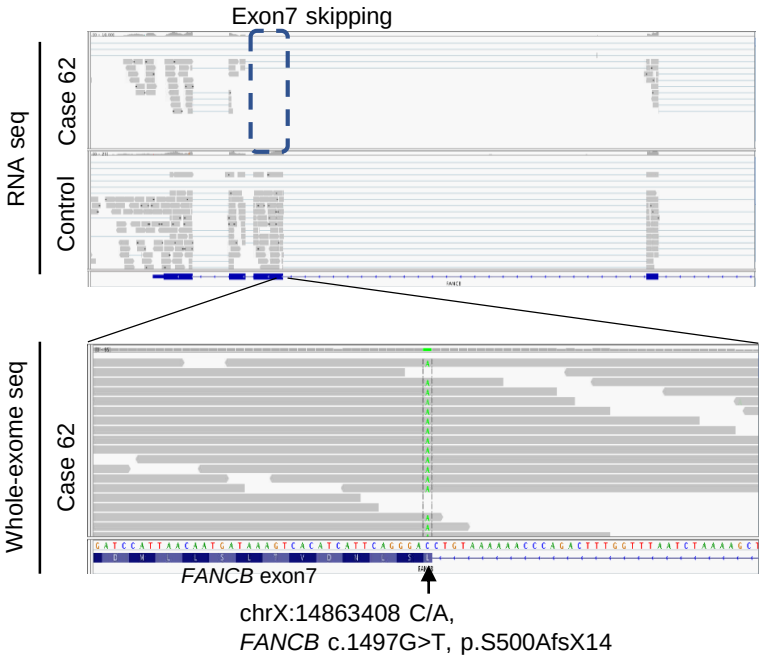
DNA change	Location	Effect	Comments	No. of alleles	No. of patients
nonsense mutations					
c.91C>T	exon 2	p.Q31X	known mutation	2	2
c.1066C>T	exon 8	p.Q356X	known mutation	15	10
small deletions					
c.194delC	exon 3	p.P65LfsX7	known mutation	3	2
c.907_908del	exon 7	p.L303GfsX5	novel muation	1	1
c.1386delC	exon 10	p.W463GfsX55	novel muation	1	1
splicing mutations					
c.307+1G>C	intron 3	aberrant splicing	known mutation	34	21
c.1637-15G>A	intron 12	VUS	novel mutation	1	1
Total				57	

variant of unknown significance

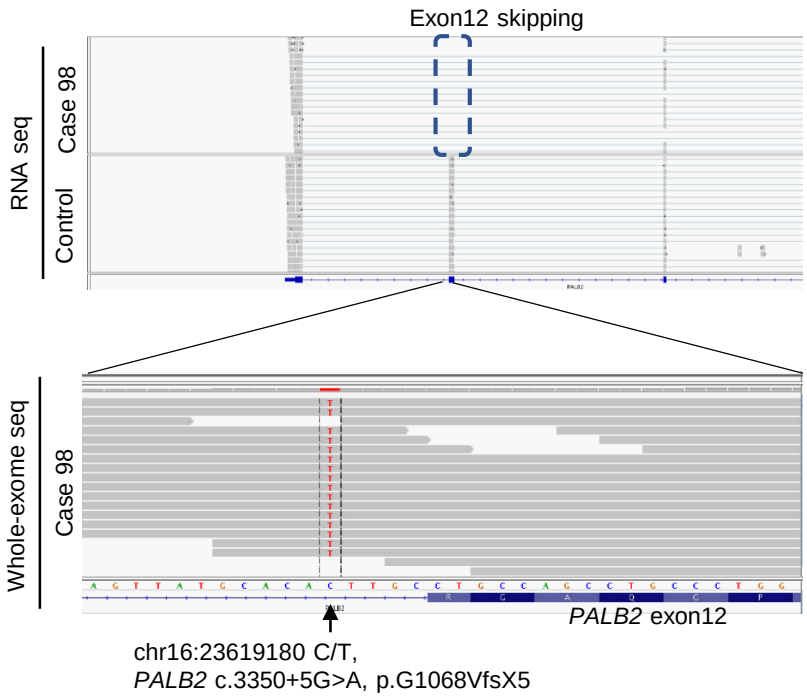


Supplemental Figure 1. Proposed model for mechanism of microhomology-mediated end joining (MMEJ) to repair DNA double-strand break (DSB). This repair model consists of at least five steps: resection of the DSB ends by nuclease digestion, annealing of 3bp homologous regions, removal of heterologous flaps, and fill-in synthesis and ligation[7]. The mutation is speculated to be created by two DSBs and subsequent religation of the two distant ends by MMEJ repair.

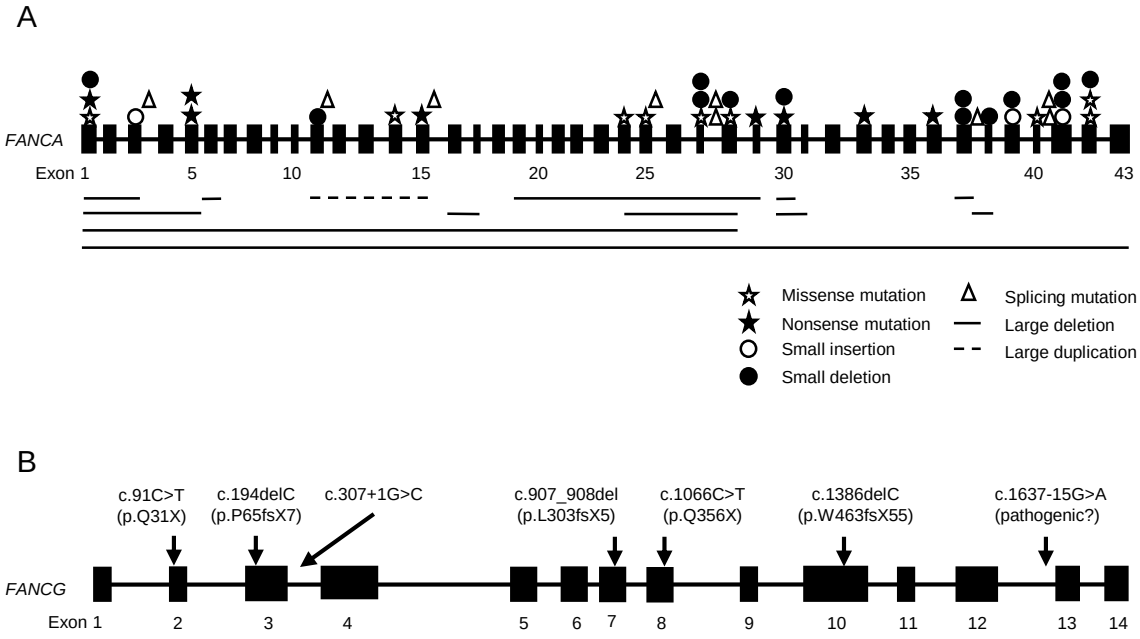
A



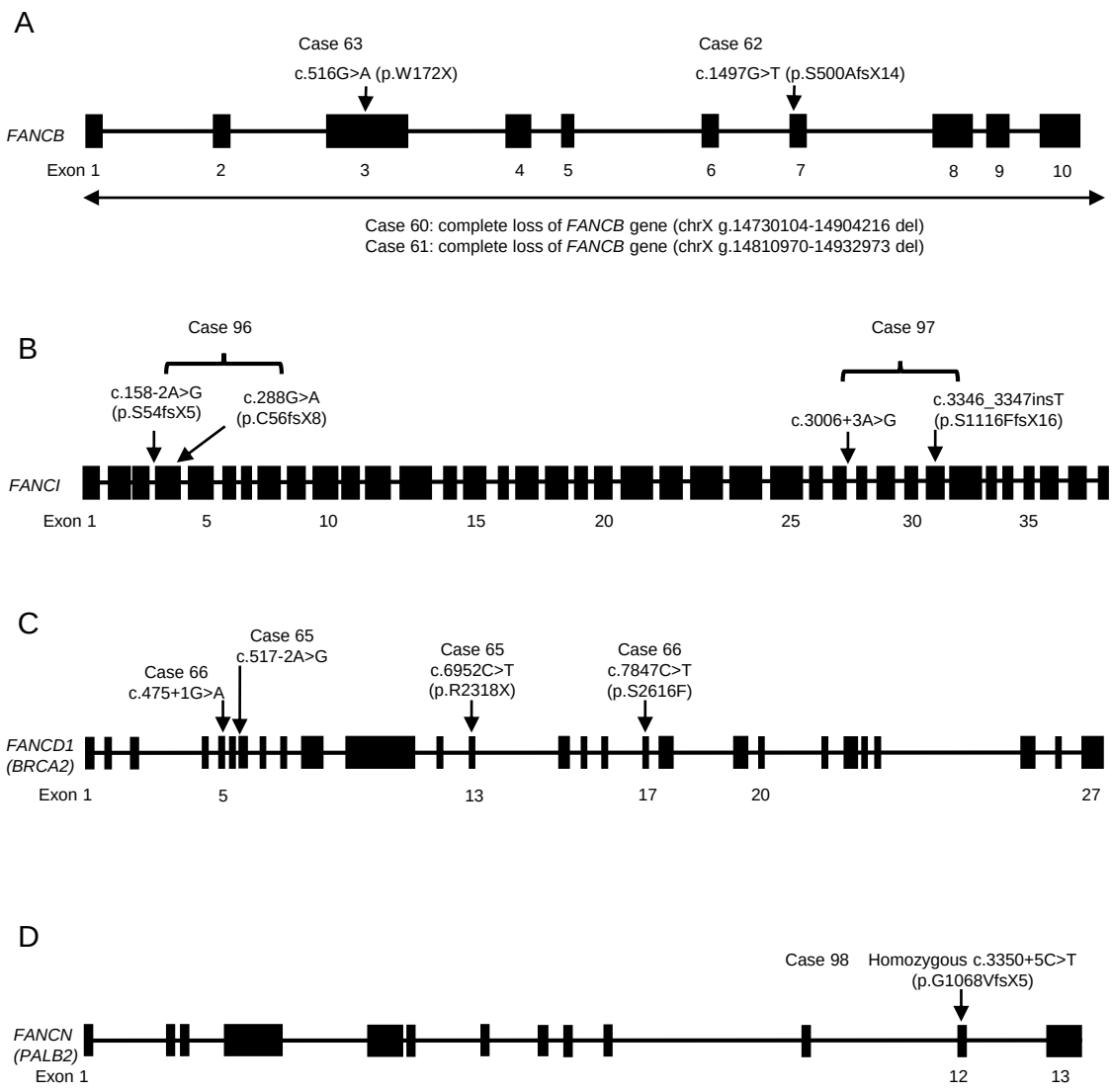
B



Supplemental Figure 2. Display of a cross section of RNA-sequencing (top) and Whole-exome sequencing (bottom). RNA sequence reads of exon 7 in *FANCB* and exon 12 in *FANCN* were absent for Case 62 (A) and Case 98 (B), respectively, which enabled us to identify exon skipping. WES analysis revealed a synonymous mutation (*FANCB* c.1497G>T) in Case 62, resulting in skipping of exon7, and a homozygous mutation (*PALB2* c.3350+5G>A) in intron 12 in Case 98, resulting in skipping of exon 12. These mutation variants were also verified by PCR and Sanger sequencing.



Supplemental Figure 3. Localization of mutation variants found in *FANCA* (A) or *FANCG* (B).



Supplemental Figure 4. Localization of mutation variants in *FANCB* (A), *FANCI* (B), *FANCD1 (BRCA2)* (C), and *FANCN (PALB2)* (D).

Supplemental References

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