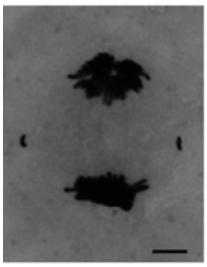
Supplementary figure legends

Supplementary Figure 1. Induction of lagging chromatids or chromatid bridges at anaphase by H_2O_2 in HCT116 cells. Asterisks indicate significant differences as compared to the control [t-test, $P < 0.05$ (*), 0.01 (**), and 0.001 (***)]. A, Lagging chromatids at anaphase in a HCT116 cell 4 h after H_2O_2 treatment. Bar, 10 μm. B, H_2O_2-induced aberrant anaphase cells with lagging chromosomes and/or chromosome bridges in HCT116 cells. Cells grown on a chamber slide were treated with H_2O_2 for 30 min and grown for 4 to 6 h. C, Induction of aneuploidy by H_2O_2 in HCT116 cells. Cells were treated with H_2O_2 for 30 min and grown for two cell cycles. The background chromosome number is 45 in HCT116 cells. Therefore cells not containing 45 chromosomes are considered aneuploid cells.

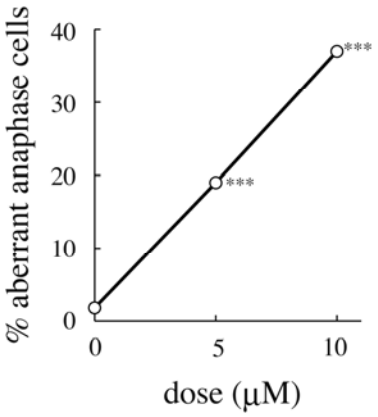
Supplementary Figure 2. H_2O_2-induced chromosomal aberrations in thymocytes. Nonirradiated thymocytes (5×10^6 cells per dish) were cultured with the proliferation stimuli for 24 h at 37°C, treated with H_2O_2 for 1 h, and then cultured with catalase for 31 h. Open circles denote individual mouse data. Closed circles with error bars denote the mean values ± standard deviations.

Supplementary Figure 3. Examples of karyotypic changes detected by chromosome painting in thymocytes and resultant thymic lymphomas. A, 10w-1 thymocyte painted with probes for chromosomes 8-14. Arrows indicate t(2;12). Chromosome 2 was not painted in this figure. B, The same translocation was observed in 10w-1-2 thymic lymphoma derived from 10w-1 thymocytes. In A and B, cells possessed trisomy 15, but chromosome 15 was not painted in these figures. C, 10w-7 thymocyte painted with probes for chromosomes 15-Y. Arrow indicates isochromosome 15. D, The same isochromosome 15 was detected in 10w-7-1 thymic lymphoma derived from 10w-7 thymocytes. In C and D, cells also bore t(1;12), but t(1;12) was not detected in these figures.

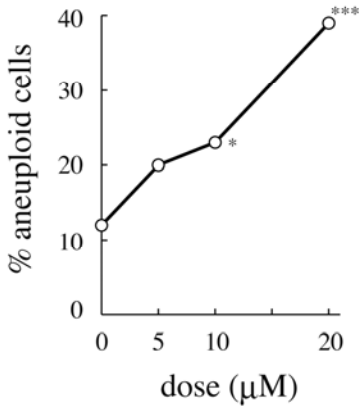
A

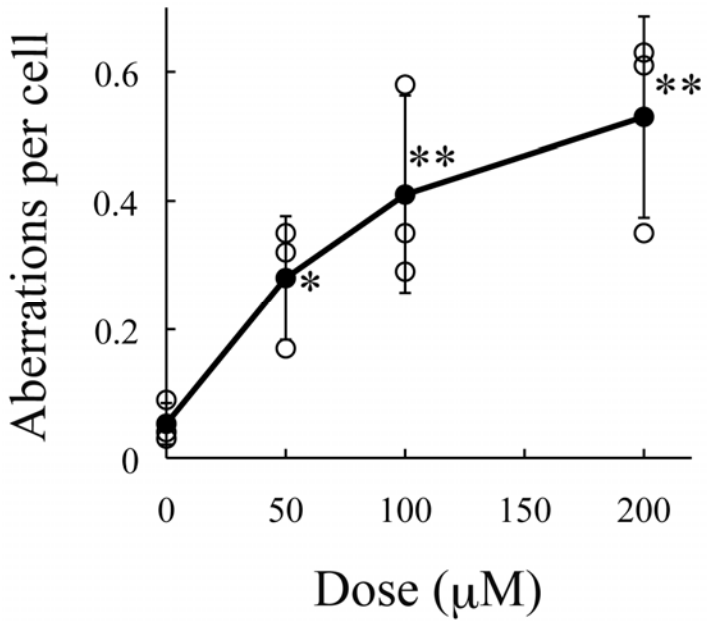


B

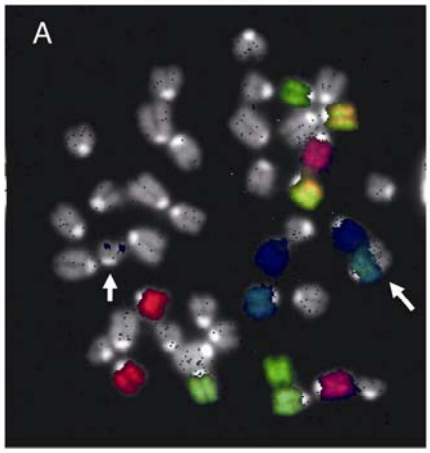

% aberrant anaphase cells
40
30
20
10
0
***
***
0
5
10
dose (μM)

C

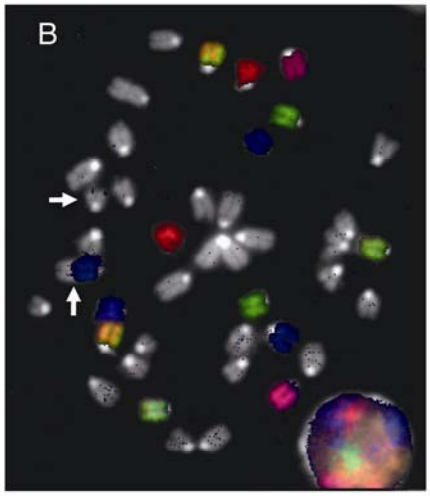

% aneuploid cells
40
30
20
10
0
***
*
0
5
10
20
dose (μM)


Aberrations per cell
0.6
0.4
0.2
0
0
50
100
150
200
Dose (μM)
*
**
**

10w-1 thymocyte
41, +15, t(2;12)

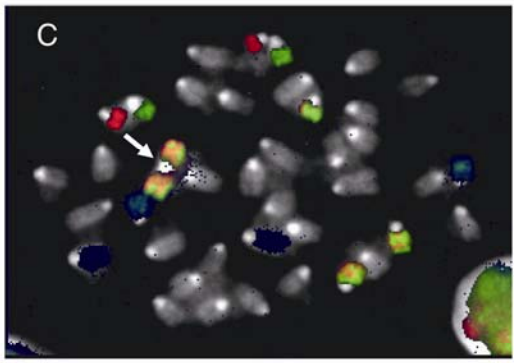


10w-1-2 thymic lymphoma

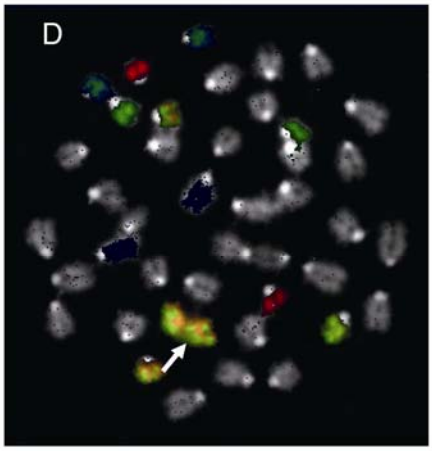


probe: chromosomes 8-14

10w-7 thymocyte
40, i(15), t(1;12)



10w-7-1 thymic lymphoma



probe: chromosomes 15-Y

Supplementary Table 1. Aneuploidy in post-irradiation thymocytes and thymic lymphomas.

Time after irradiation	No. of thymocytes (mice) examined	% aneuploid cells	No. of cells with different chromosome number											
			38 (%)	39 (%)	40 (%)	41 (%)	42 (%)	43 (%)	44 (%)	45 (%)	46 (%)	47 (%)	48 (%)	49 (%)
Nonirradiated	200 (5)	0.5			199 (99.5)	1 (0.5)								
0 week (2 h)	200 (5)	3.0	1 (0.5)	3 (1.5)	194 (97)	2 (1)								
2 weeks	360 (9)	14.4		9 (2.5)	308 (86)	36 (10)	7 (2)							
4 weeks	320 (8)	34.1	1 (0.3)	2 (0.6)	211 (66)	101 (32)	4 (1)	1 (0.3)						
6 weeks	360 (9)	34.7	3 (1)	8 (2)	235 (65)	112 (31)	2 (0.6)							
8 weeks	600 (15)	28.3		11 (2)	429 (72)	146 (24)	12 (2)	1 (0.2)	1 (0.2)					
10 weeks	280 (7)	18.9		8 (3)	227 (81)	44 (16)	1 (0.4)							
13 weeks	320 (8)	33.1		8 (3)	214 (67)	72 (23)	26 (8)							
Thymic lymphomas	560 (14)	61.6		2 (0.4)	215 (38)	286 (51)	10 (2)	6 (1)	0 (0)	0 (0)	0 (0)	3 (0.5)	24 (4)	14 (3)

Supplementary Table 2. Comparison of percent aneupoidy between thymocytes incubated with Colcemid for 3 h and thymocytes cultured with PMA, ionomycin, and 2-mercaptoethanol for 48 h.

Mice[a]	Treatments of thymocytes	No. of metaphases examined	Aneuploid cells (%)	No. of metaphase cells with different chromosome number			
				39	40	41	42
13w-6	Colcemid, 3 h	40	2 (5.0)	0	38	2	0
	PMA, 48 h	40	2 (5.0)	0	38	2	0
13w-7	Colcemid, 3 h	40	28 (70.0)	0	12	26	2
	PMA, 48 h	40	27 (67.5)	1	13	26	0
13w-9	Colcemid, 3 h	40	12 (30.0)	0	28	12	0
	PMA, 48 h	40	15 (37.5)	0	25	15	0
13w-10	Colcemid, 3 h	40	28 (70.0)	0	12	3	25
	PMA, 48 h	40	27 (67.5)	0	13	1	26
13w-11	Colcemid, 3 h	40	10 (25.0)	0	30	10	0
	PMA, 48 h	40	13 (32.5)	3	27	10	0
13w-12	Colcemid, 3 h	40	0 (0)	0	40	0	0
	PMA, 48 h	40	1 (2.5)	1	39	0	0
13w-14	Colcemid, 3 h	40	18 (45.0)	2	22	16	0
	PMA, 48 h	40	22 (55.0)	0	18	22	0
13w-15	Colcemid, 3 h	40	2 (5.0)	0	38	2	0
	PMA, 48 h	40	1 (2.5)	0	39	1	0

[a] Mice were irradiated at 1.8 Gy 4 times at 1-week intervals. Aneuploidy was examined in thymocytes 13 weeks after irradiation.

Supplementary Table 3. Chromosomal aberrations in descendants of irradiated thymocytes.

Weeks after irradiation	No. of cells (mice) examined	No. of aberrant cells (%)	Mean No. of aberrations per cell	Chromatid-type				Chromosome-type							
				G	B	E	SM	G	B	Dic	R	F	DM	Pul	CF
Nonirradiated	1912 (10)	126 (6.6)	0.069	103	12	7	0	2	0	0	0	0	1	1	6
0	353 (5)	145 (41.1)	0.808	38	12	14	6	1	0	83	16	122	14	2	5
2	1000 (5)	102 (10.2)	0.113	60	6	6	3	7	0	7	0	2	5	1	16
4	1000 (5)	86 (8.6)	0.103	40	16	7	4	3	1	5	0	8	9	2	8
6	1717 (9)	177 (10.3)	0.115	125	10	14	7	8	1	1	0	4	1	0	27
8	2937 (15)	424 (14.4)	0.163	342	40	19	7	21	3	4	2	9	4	2	27
10	1400 (7)	200 (14.3)	0.162	158	22	19	0	3	1	4	0	6	1	1	12
13	1101 (7)	146 (13.3)	0.155	123	12	5	3	3	0	2	2	11	4	2	4

Abbreviations: G, gap; B, break; E, exchange; Dic, dicentric; R, ring; F, fragment; DM, double minute; SM, single minute; Pul, pulverization; CF, centromeric fusion.

Supplementary Table 4. *Notch1* rearrangements in thymocyte clones after 4 consecutive irradiations of 1.8 Gy γ-rays.

Weeks after irradiation	Mouse designation	Rearranged sequences in thymocyte clones[a]	Frequency
0	0w-1	4926·G·16675	7.6×10^{-6}
	0w-3	4921·GCTA·16674	7.6×10^{-6}
	0w-4	4921·GA·16674	1.3×10^{-5}
2	2w-1	8186·CGCGAGGGG·16676	4.4×10^{-6}
4	4w-9	4924·G·14786	7.6×10^{-6}
8	8w-3	4923·GGTA·16674	1.3×10^{-4}
		4923·AGGGG·16674	9.9×10^{-5}
		8189·A·16674	4.4×10^{-5}
	8w-6	4926·GAGATAC·16676	1.1×10^{-4}
		8189·AGGG·16675	1.2×10^{-4}
		8190·CA·16674	4.4×10^{-5}
	8w-11	4925·GAGGAGT·16676	9.9×10^{-6}
	8w-13	8190·GAGGGAA·16675	7.6×10^{-6}
		8186·CCC·16676	4.4×10^{-6}

[a] Rearranged sequences are expressed as breakpoint sequence ·inserted nucleotides· breakpoint sequence.

Supplementary Table 5. Examples of *TCRβ* rearrangements in thymocyte clones and resultant thymic lymphomas.

Mice examined	*TCRβ* rearrangements in thymocyte clones	Thymic lymphomas examined	*TCRβ* rearrangements
6w-8	–[a]	6w-8-1	D2/J2.3
		6w-8-2	D2/J2.3
			D2/GCGGGTA/J2.1
		6w-8-3	D2/GCGGGTA/J2.1
8w-14	D2/T/J2.3	8w-14-1	D2/J2.5
	D2/J2.5	8w-14-2	D2/J2.5
		8w-14-3	D2/J2.5
		8w-14-4	D2/T/J2.3
			D2/J2.5
8w-16	D1/J1.1	8w-16-1	D1/J1.1
	D2/T/J2.4		D2/T/J2.4
		8w-16-2	D1/J1.1
			D2/T/J2.4
		8w-16-3	D1/J1.1
			D2/T/J2.4
		8w-16-4	D1/J1.1
			D2/T/J2.4
		8w-16-6	D1/J1.1
			D2/T/J2.4
8w-17	D1/GA/J1.4	8w-17-1	D1/GA/J1.4
	D2/CGCGGG/J2.2		D2/CGCGGG/J2.2
	D2/T/J2.7		D2/T/J2.7
		8w-17-2	D1/GA/J1.4
			D2/CGCGGG/J2.2
			D2/T/J2.7
		8w-17-3	D1/GA/J1.4
			D2/CGCGGG/J2.2
			D2/T/J2.7
		8w-17-4	D1/GA/J1.4
			D2/CGCGGG/J2.2
			D2/T/J2.7
10w-3	D1/GGAA/J1.4	10w-3-1	D2/TACG/J2.5
	D2/CGT/J2.3	10w-3-2	D2/TACG/J2.5
	D2/TACG/J2.5	10w-3-3	D2/TACG/J2.5
		10w-3-4	D2/TACG/J2.5
10w-7	D1/CCCCTTCGGG/J1.3	10w-7-1	D1/CCCCTTCGGG/J1.3
		10w-7-2	D1/CCCCTTCGGG/J1.3
		10w-7-3	D1/CCCCTTCGGG/J1.3
		10w-7-4	D1/CCCCTTCGGG/J1.3

[a] The minus symbol indicates the absence of clonal *TCRβ* rearrangements in thymocytes.

Supplementary Table 6. Rearranged sequences of *Notch1* gene in resultant thymic lymphomas.

Thymic lymphomas	No. of *Notch1* rearrangements	Rearranged sequences[a]
6w-7-1	2	4924·G·16675, ?
6w-8-1	4	4925·16676, 8190·T·15582, 8190·16677, 8190·16681
6w-8-2	1	4924·16676
6w-8-3	3	4925·16676, 11179·15437, ?
6w-9-1	1	4926·AA·16675
6w-9-2	1	4926·GGAGG·16675
6w-9-3	5	4926·14781, 4926·GGAC·16675, 4926·GGGA·22145, 4926·GGGA·22147, 8073·TC·16006
6w-9-4	0	
6w-9-5	2	4923·GTTA·16674, 8073·C·16006
6w-10-2	1	4926·A·16674
6w-10-3	1	? in juxtamembrane extracellular domain
6w-10-4	1	4926·16674
8w-13-1	0	
8w-13-2	2	4926·TGGG·16676, 4926·GGACGGTATATG·16675
8w-13-3	1	4926·GGGGGG·16676
8w-14-1	1	?
8w-14-2	2	4926·GTG·16676, ?
8w-14-3	1	4926·TGG·16674
8w-14-4	1	4923·T·16674
8w-16-1	1	? in juxtamembrane extracellular domain
8w-16-2	0	
8w-16-3	1	4923·A·16674
8w-16-4	1	? in juxtamembrane extracellular domain
8w-16-6	0	
8w-17-1	5	4923·GTTTTA·22147, 4923·GTTTTA·22149, 4926·GTG·16676, 4925·GCG.16677, 8185·TG·16676
8w-17-2	7	4926·A·16674, 4925·AAA·22146, 4926·GTT·22146, 8185·GATCTCT·15809, 8187·CTGA·16674, 8188·CCTA·16674, ?
8w-17-3	5	4926·GGGCAG·16676, 4926·GCC·22149, 8190·T·16009, 8190·GGG·16676, ?
8w-17-4	1	4924·GGGAG·16675
10w-1-1	1	4923·GCCGCCAG·16675
10w-1-2	2	4921·CCCCCCTG·16675, 8190·ATTTTTGCATG·16676
10w-1-4	2	4926·TAGCTTT·16678, ?
10w-1-5	2	4926·GGGAT·16676, ?
10w-3-1	2	?, ?
10w-3-2	2	4921·CTATAA·16677, ?
10w-3-3	3	4925·16677, 8187·GTAAAAG·16674, ?
10w-3-4	1	4926·AGGTA·16674
10w-4-1	2	4922·TA·16674, ?
10w-7-1	1	?
10w-7-2	1	4926·TGGGGATAGG·16676
10w-7-3	0	
10w-7-4	0	

[a] Rearranged sequences are shown as breakpoint sequence·inserted nucleotides·breakpoint sequence. The question marks indicate an unknown sequence.