

## Supplementary Materials for

### Transient ATM Kinase Inhibition Disrupts DNA Damage–Induced Sister Chromatid Exchange

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Fig. S1. KU60019 is another reversible inhibitor of ATM activity.

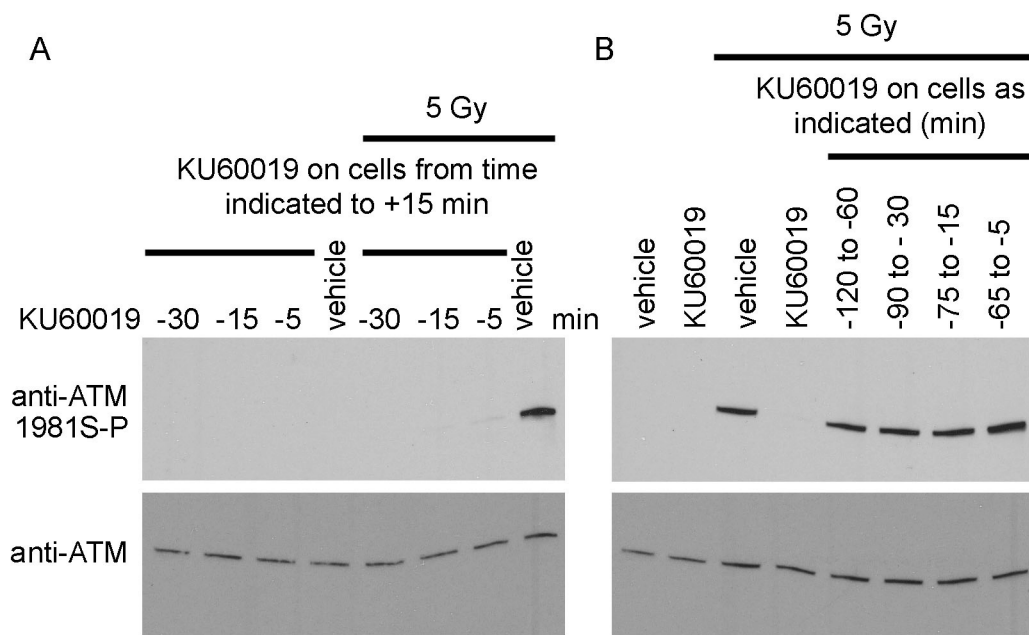
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Table S3. Chromosome aberrations induced by 2 Gy IR in late-S and G<sub>2</sub>-phase IMR90 fibroblasts (+15 to +75 min post-IR).

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Table S5. Chromosome aberrations induced by 2 Gy IR in Artemis-defective cells after inhibition of ATM for 1 hour.



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Fig. S1. KU60019 is another reversible inhibitor of ATM activity. (A) ATM kinase activity is inhibited in IMR90 fibroblasts 15 min following irradiation when 1  $\mu$ M KU60019 is added to fibroblasts 5 min prior to the irradiation. 1  $\mu$ M KU60019 was added at the time indicated before the irradiation and left on the cells until lysis. (B) ATM activity is undiminished in IMR90 fibroblasts 15 min following exposure to 5 Gy IR when 1  $\mu$ M KU60019 is removed from fibroblasts 5 min prior to the irradiation. Cells were exposed to 1  $\mu$ M KU60019 for a 1 h period culminating at the time indicated prior to the irradiation (-120 min to -60 min, -90 min to -30 min, -75 min to -15 min, and -65 min to -5 min). Media containing 1  $\mu$ M KU60019 was then removed and replaced with preconditioned media. 1  $\mu$ M KU60019 was added to the control samples at -30 min and left on the cells.

Table S1. Dose-dependent and KU55933-dependent increase in chromosome aberrations in IMR90 fibroblasts. TWA, total weighted aberrations; no, number; freq, frequency; pct, percent

| IR dose and time of drug or vehicle (veh) exposure | TWA/cell +/- SEM | total no. aberrations | chromatid breaks (freq [pct]) | chromosome breaks (freq [pct]) |
|--|------------------|-----------------------|-------------------------------|--------------------------------|
| veh +15 to +75                                     | 0.16 +/- 0.07    | 8                     | 8 [100]                       | 0 [0]                          |
| 0.1 Gy + veh +15 to +75                            | 0.25 +/- 0.08    | 10                    | 8 [80]                        | 2 [20]                         |
| 0.5 Gy + veh +15 to +75                            | 0.29 +/- 0.09    | 13                    | 12 [92]                       | 1 [8]                          |
| 1 Gy + veh +15 to +75                              | 0.33 +/- 0.09    | 15                    | 14 [93]                       | 1 [7]                          |
| 2 Gy + veh +15 to +75                              | 0.24 +/- 0.08    | 12                    | 12 [100]                      | 0 [0]                          |
| 5 Gy + veh +15 to +75                              | 1.11 +/- 0.18    | 44                    | 35 [80]                       | 9 [20]                         |
| 10 Gy + veh +15 to +75                             | 0.93 +/- 0.17    | 37                    | 28 [76]                       | 9 [24]                         |
| KU55933 +15 to +75                                 | 0.16 +/- 0.06    | 8                     | 8 [100]                       | 0 [0]                          |
| 0.1 Gy + KU55933 +15 to +75                        | 0.62 +/- 0.17    | 24                    | 17 [71]                       | 7 [29]                         |
| 0.5 Gy + KU55933 +15 to +75                        | 0.53 +/- 0.13    | 24                    | 22 [92]                       | 2 [8]                          |
| 1 Gy + KU55933 +15 to +75                          | 0.79 +/- 0.13    | 37                    | 35 [95]                       | 2 [5]                          |
| 2 Gy + KU55933 +15 to +75                          | 1.15 +/- 0.18    | 39                    | 22 [56]                       | 17 [44]                        |
| 5 Gy + KU55933 +15 to +75                          | 1.60 +/- 0.22    | 59                    | 40 [68]                       | 19 [32]                        |
| 10 Gy + KU55933 +15 to +75                         | 3.15 +/- 0.35    | 115                   | 75 [65]                       | 40 [35]                        |

Table S2. Dose-dependent and NU7441-dependent increase in chromosome aberrations in IMR90 fibroblasts.

| IR dose and time of drug or vehicle (veh) exposure | TWA/cell +/- SEM | total no. aberrations | chromatid breaks (freq [pct]) | chromosome breaks (freq [pct]) |
|--|------------------|-----------------------|-------------------------------|--------------------------------|
| veh +15 to +75                                     | 0.68 +/- 0.15    | 33                    | 32 [97]                       | 1 [3]                          |
| 0.1 Gy + veh +15 to +75                            | 0.68 +/- 0.12    | 31                    | 28 [90]                       | 3 [10]                         |
| 0.5 Gy + veh +15 to +75                            | 0.87 +/- 0.15    | 41                    | 38 [93]                       | 3 [7]                          |
| 1 Gy + veh +15 to +75                              | 0.65 +/- 0.14    | 30                    | 28 [93]                       | 2 [7]                          |
| 2 Gy + veh +15 to +75                              | 0.76 +/- 0.13    | 37                    | 36 [97]                       | 1 [3]                          |
| 5 Gy + veh +15 to +75                              | 1.06 +/- 0.17    | 49                    | 46 [94]                       | 3 [6]                          |
| 10 Gy + veh +15 to +75                             | 1.42 +/- 0.24    | 62                    | 51 [82]                       | 11 [18]                        |
| NU7441 +15 to +75                                  | 0.70 +/- 0.15    | 33                    | 30 [91]                       | 3 [9]                          |
| 0.1 Gy + NU7441 +15 to +75                         | 0.81 +/- 0.17    | 40                    | 40 [100]                      | 0 [0]                          |
| 0.5 Gy + NU7441 +15 to +75                         | 0.92 +/- 0.20    | 46                    | 45 [98]                       | 1 [2]                          |
| 1 Gy + NU7441 +15 to +75                           | 1.20 +/- 0.20    | 60                    | 57 [95]                       | 3 [5]                          |
| 2 Gy + NU7441 +15 to +75                           | 1.61 +/- 0.26    | 75                    | 64 [85]                       | 10 [13]                        |
| 5 Gy + NU7441 +15 to +75                           | 2.73 +/- 0.41    | 88                    | 74 [84]                       | 11 [13]                        |
| 10 Gy + NU7441 +15 to +75                          | 3.70 +/- 0.99    | 27                    | 21 [78]                       | 6 [22]                         |

Table S3. Chromosome aberrations induced by 2 Gy IR in late-S and G<sub>2</sub>-phase IMR90 fibroblasts (+15 to +75 min post-IR).

|                    | IR dose and time of drug or vehicle (veh) exposure | TWA/cell +/- SEM | total no. aberrations | percent positive | chromatid breaks (freq [pct]) | chromosome breaks (freq [pct]) |
|--------------------|--|------------------|-----------------------|------------------|-------------------------------|--------------------------------|
| <b>calyculin A</b> | veh +15 to +75                                     | 0.46 +/- 0.10    | 23                    | 34               | 23 [100]                      | 0 [0]                          |
|                    | KU55933 +15 to +75                                 | 0.43 +/- 0.10    | 21                    | 32               | 21 [100]                      | 0 [0]                          |
|                    | NU7441 +15 to +75                                  | 0.84 +/- 0.15    | 41                    | 48               | 41 [100]                      | 0 [0]                          |
|                    | KU55933/NU7441 +15 to +75                          | 0.67 +/- 0.13    | 32                    | 44               | 31 [97]                       | 1 [3]                          |
|                    | 2 Gy + veh +15 to +75                              | 0.84 +/- 0.18    | 42                    | 42               | 42 [100]                      | 0 [0]                          |
|                    | 2 Gy + KU55933 +15 to +75                          | 1.41 +/- 0.25    | 64                    | 54               | 59 [92]                       | 5 [8]                          |
|                    | 2 Gy + NU7441 +15 to +75                           | 1.85 +/- 0.24    | 84                    | 74               | 78 [93]                       | 6 [7]                          |
|                    | 2 Gy + KU55933/NU7441 +15 to +75                   | 2.49 +/- 0.26    | 113                   | 80               | 103 [91]                      | 10 [9]                         |
| <b>colcemid</b>    | veh +15 to +75                                     | 0.08 +/- 0.06    | 3                     | 4                | 2 [67]                        | 1 [33]                         |
|                    | KU55933 +15 to +75                                 | 0.02 +/- 0.02    | 1                     | 2                | 1 [100]                       | 0 [0]                          |
|                    | NU7441 +15 to +75                                  | 0.02 +/- 0.02    | 1                     | 2                | 1 [100]                       | 0 [0]                          |
|                    | KU55933/NU7441 +15 to +75                          | 0.06 +/- 0.04    | 2                     | 4                | 1 [50]                        | 1 [50]                         |
|                    | 2 Gy + veh +15 to +75                              | 0.13 +/- 0.07    | 3                     | 8                | 2 [67]                        | 1 [33]                         |
|                    | 2 Gy + KU55933 +15 to +75                          | 0.06 +/- 0.04    | 2                     | 4                | 1 [50]                        | 1 [50]                         |
|                    | 2 Gy + NU7441 +15 to +75                           | 0.43 +/- 0.13    | 13                    | 20               | 8 [62]                        | 5 [38]                         |
|                    | 2 Gy + KU55933/NU7441 +15 to +75                   | 0.20 +/- 0.08    | 7                     | 14               | 4 [57]                        | 3 [43]                         |

Table S4. Chromosome aberrations induced by 2 Gy IR in late-S and G<sub>2</sub>-phase IMR90 cells (+15 to +225 min post-IR).

|                    | IR dose and time of drug or vehicle (veh) exposure | TWA/cell +/- SEM | total no. aberrations | percent positive | chromatid breaks (freq [pct]) | chromosome breaks (freq [pct]) | Radials (freq [pct]) |
|--------------------|--|------------------|-----------------------|------------------|-------------------------------|--------------------------------|----------------------|
| <b>calyculin A</b> | veh +15 to +225                                    | 0.30 +/- 0.09    | 15                    | 22               | 15 [100]                      | 0 [0]                          | 0 [0]                |
|                    | KU55933 +15 to +225                                | 0.50 +/- 0.15    | 24                    | 28               | 23 [96]                       | 1 [4]                          | 0 [0]                |
|                    | NU7441 +15 to +225                                 | 0.36 +/- 0.09    | 18                    | 28               | 18 [100]                      | 0 [0]                          | 0 [0]                |
|                    | KU55933/NU7441 +15 to +225                         | 0.39 +/- 0.11    | 20                    | 26               | 20 [100]                      | 0 [0]                          | 0 [0]                |
|                    | 2 Gy + veh +15 to +225                             | 0.44 +/- 0.14    | 21                    | 28               | 20 [95]                       | 1 [5]                          | 0 [0]                |
|                    | 2 Gy + KU55933 +15 to +225                         | 0.76 +/- 0.15    | 34                    | 42               | 30 [88]                       | 4 [12]                         | 0 [0]                |
|                    | 2 Gy + NU7441 +15 to +225                          | 0.99 +/- 0.18    | 46                    | 52               | 42 [91]                       | 4 [9]                          | 0 [0]                |
|                    | 2 Gy + KU55933/NU7441 +15 to +225                  | 2.09 +/- 0.26    | 88                    | 80               | 69 [78]                       | 18 [21]                        | 1 [1]                |
| <b>colcemid</b>    | veh +15 to +225                                    | 0.00 +/- 0.00    | 0                     | 0                | 0 [0]                         | 0 [0]                          | 0 [0]                |
|                    | KU55933 +15 to +225                                | 0.04 +/- 0.04    | 1                     | 2                | 1 [100]                       | 0 [0]                          | 0 [0]                |
|                    | NU7441 +15 to +225                                 | 0.02 +/- 0.02    | 1                     | 2                | 1 [100]                       | 0 [0]                          | 0 [0]                |
|                    | KU55933/NU7441 +15 to +225                         | 0.02 +/- 0.02    | 1                     | 2                | 1 [100]                       | 0 [0]                          | 0 [0]                |
|                    | 2 Gy + veh +15 to +225                             | 0.08 +/- 0.06    | 2                     | 4                | 1 [50]                        | 1 [50]                         | 0 [0]                |
|                    | 2 Gy + KU55933 +15 to +225                         | 0.20 +/- 0.12    | 5                     | 4                | 3 [60]                        | 2 [40]                         | 0 [0]                |
|                    | 2 Gy + NU7441 +15 to +225                          | 0.28 +/- 0.10    | 8                     | 14               | 4 [50]                        | 4 [50]                         | 0 [0]                |
|                    | 2 Gy + KU55933/NU7441 +15 to +225                  | 0.67 +/- 0.18    | 18                    | 26               | 9 [50]                        | 9 [50]                         | 0 [0]                |

Table S5. Chromosome aberrations induced by 2 Gy IR in Artemis-defective cells after inhibition of ATM for 1 hour.

| IR dose and time of drug or vehicle (veh) exposure | TWA/cell +/- SEM | total no. aberrations | percent positive | chromatid breaks (freq [pct]) | chromosome breaks (freq [pct]) |
|--|------------------|-----------------------|------------------|-------------------------------|--------------------------------|
| veh +15 to +75                                     | 0.54 +/- 0.15    | 23                    | 26               | 19 [83]                       | 4 [17]                         |
| KU55933 +15 to +75                                 | 0.41 +/- 0.14    | 17                    | 18               | 14 [82]                       | 3 [18]                         |
| 2 Gy + veh +15 to +75                              | 0.61 +/- 0.14    | 28                    | 38               | 25 [89]                       | 3 [11]                         |
| 2 Gy + KU55933 +15 to +75                          | 1.38 +/- 0.23    | 57                    | 56               | 43 [75]                       | 14 [25]                        |