Current Biology, Vol. 14, R613–R614, August 10, 2004, ©2004 Elsevier Ltd. All rights reserved. DOI 10.1016/j.cub.2004.07.042

Telomeres: Not All Breaks Are Equal

Dispatch

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ATM, Rad50 and Mre11 have been shown to prevent telomere fusion in *Drosophila*, thereby extending the protective role of DNA damage checkpoint proteins to non-canonical telomeres formed without telomerase. How do these proteins help chromosomal termini escape fusion through 'repair' while promoting repair of induced DNA breaks?

When a eukaryotic cell is exposed to a genotoxic treatment such as ionizing radiation that induces a DNA double-strand break, the response is led by two kinases related to the well known signalling enzyme phosphatidyl inositol 3-kinase. These are ATM, for ataxia telangiectasia mutated, and ATR, for ATM and Rad3-like, which activate DNA repair and cell-cycle arrest in the damaged cell [1]. ATM and ATR proteins are conserved in all eukaryotes examined. Components of the so-called MR protein complex, Mre11, Rad50 and the less-well conserved Nbs1, cooperate with ATM/ATR homologs in DNA damage responses, with MR proteins activating ATM/ATR homologs and *vice versa* [2–4].

ATM/ATR homologs and MR proteins also respond to the naturally occurring DNA double-strand 'breaks' — telomeres — at the ends of linear chromosomes. Human, mouse and yeast cells that are deficient in ATM/ATR homologs have shorter telomeres which often fuse to other telomeres or induced double-strand breaks [5,6]. Likewise, mutations in MR proteins lead to shortened and fused telomeres in yeast [7,8]. In these organisms, telomere length is maintained by telomerase, a reverse transcriptase which restores the GC-rich repeats at chromosome ends that would otherwise shorten after each round of DNA replication. The ends are further protected by the binding of proteins such as TRF2 homologs, and by DNA secondary structures. Genetic analyses suggest that ATM/ATR and MR proteins function to maintain telomere length or to prevent telomere fusion by telomerase-dependent and telomerase-independent mechanisms [9]. The exact nature of these mechanisms, and their relative contribution to telomere maintenance, needed clarification.

Six papers [10–15] — four in this issue of *Current Biology* [10,11,13,14] — have now reported that the *Drosophila* homologs of ATM (allelic to the previously described gene *telomere fusion*), Mre11 and Rad50 play a role in telomere maintenance. Mutants defective in each of these proteins show increased fusion of telomeres, with the proportion of mitotic cells showing at least one fusion event ranging from 50% to over 90%. *Drosophila* telomeres are unusual; they are made up of repeats of Het-A and TERT retrotransposons that are maintained by transposition rather than telomerase activity, and are protected by binding of two proteins, heterochromatin protein 1 (HP1) and HP1, ORC2 associated protein (HOAP). The new studies [10–15] thus clearly define telomerase-independent contributions of ATM and MR proteins to telomere maintenance and, specifically, in the prevention of fusions. Furthermore, it now appears that the role of ATM/ATR and MR proteins in telomere protection is universal, and occurs regardless of other mechanisms that lengthen telomeres or protect them. The consequences of the loss of this protection in *Drosophila* are devastating; fused telomeres seem to induce a breakage–fusion cycle and lead to apoptosis and organismal death. Thus, paradoxically, DNA damage checkpoints that have been shown to promote apoptosis in response to DNA damage are fulfilling an anti-apoptotic role indirectly through telomere protection.

A possible mechanism for telomere protection by ATM and MR proteins in *Drosophila* is suggested by two findings. First, Het-A sequences are still present at telomeres in mutants, even in those participating in fusion [10,12]. Although it was difficult to quantify the extent of sequences present, it is clear that total recession of Het-A repeats was not necessary for fusion. Second, two telomere-associated proteins, HOAP and HP1, are missing or present at reduced levels from telomeres in ATM, Rad50 and Mre11 mutants [10–12]. The reduction of HOAP is apparent in mitotic cells in Mre11 and Rad50 mutants, while both HOAP and HP1 are reduced on telomeres of polytene chromosomes in ATM, Mre11 and Rad50 mutants. As HOAP and HP1 are known to prevent telomere fusion in mitotically proliferating cells [16,17], reduction of these proteins may explain why telomeres fuse in ATM, Mre11 and Rad50 mutants. The fusions appear to be due, to a significant degree, to end-to-end joining of telomeres. Mutations in *Drosophila* ligase IV reduced, but did not abolish, telomere fusion in ATM mutants [10], similar to findings in budding yeast and mammals [8,18]. As ligase IV homologs are needed to repair a DNA double-stranded break by non-homologous end-joining [1], the latter may be the mechanism for at least some telomere fusion events.

Drosophila Mre11 and Rad50 mutants are defective in ionizing-radiation-induced repair of DNA [11]. Mre11 and Rad50 homologs also act in the repair of DNA double-strand breaks in yeast and mammals [1]. Thus, we revisit a question posed by previous work: how can the same proteins act to repair DNA double-strand breaks induced, for example, by radiation, while acting to preserve them stably as telomeres if the 'breaks' are chromosomal termini (Figure 1)? This question becomes all the more pressing because internally deleted chromosome ends, such as those generated by mobilizing of a transposable element, can recruit HOAP and HP1 to function stably as telomeres for generations in *Drosophila* [16,17]. Assuming that MR proteins are also needed for recruitment of telomeric proteins to internally deleted ends, what is different about those deletions that lead to protein recruitment and telomere

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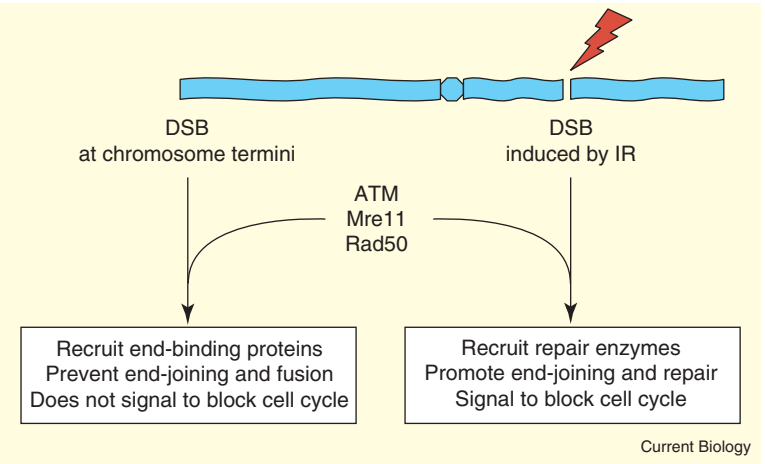

Figure 1.

A schematic illustration of the paradoxical roles of DNA damage checkpoint functions at either a telomere or an internal DNA double-strand break (DSB) on a chromosome (blue). The role of ATM and MR proteins in telomere protection has been shown previously for organisms in which telomerase maintains telomeres. New work in *Drosophila* [10–15], which uses transposition to maintain telomeres, illustrates clear telomerase-independent roles for these proteins in telomere protection.

formation, while other deletions are repaired through the activity of the same proteins? A related question is how HP1 and HOAP are recruited to telomeres in a MR-dependent manner but not to double-strand breaks caused by ionizing radiation, for instance? Conversely, if repair enzymes are recruited to a DNA double-strand break caused by damaging agents, why are they not recruited to chromosome ends?

In addition to activating DNA repair, DNA double-strand breaks also activate a checkpoint that causes cell-cycle delays. ATM/ATR and MR proteins are needed for this checkpoint in yeast and mammals [1,19]. The checkpoint role of *Drosophila* Mre11 and Rad50 remains to be examined, but *Drosophila* ATM may have a minor role that is apparent at shorter times after irradiation with mutants showing robust regulation of mitosis at later times [12–14]. The question then is: if double-strand breaks produce cell-cycle delay, why do not telomeres? ATM and Mre11 were recently shown to become co-localized at telomeric foci in senescent human cells [20]. Depletion of ATM caused these cells to re-enter the cell cycle. Thus, in senescent cells, ATM appears to recognize telomeres as DNA breaks and cause cell-cycle arrest. An intriguing possibility, then, is that onset of senescence may simply reflect a switch in which ATM goes from having a telomere-protective role that does not result in cell-cycle arrest, to a DNA-damage checkpoint role that does result in cell-cycle arrest and hence senesence. In any case, addressing how the same set of molecules, ATM and MR proteins, could initiate different sets of outcomes depending on where on the chromosome a 'break' is should prove to be challenging and rewarding.

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