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Telomere-dependent senescence

To the editor:

Rubin[1,2] has argued in your commentary and elsewhere that the limited division capacity of primary human cells in vitro is due to the accumulation of "damage" during cell culture. For fibroblasts, this possibility has now been disproven. Telomerase on its own can extend the in vitro lifespan of human fibroblasts[3–5] with no detectable change in phenotype[6,7]. These otherwise normal cells are maintained under identical culture conditions to controls and in Rubin's model should thus still "sustain cumulative damage during serial subcultivations"[2]. The fact that they continue to divide indefinitely is proof that telomere erosion is directly involved in fibroblast senescence and that any other damage that occurs in vitro is insufficient to halt cell division.

Rubin[8] used data on the relationship between fibroblast replicative potential and donor age[9] to argue in a recent letter that there is no ". . .fixed limit to cell division in vivo, much less a mechanism such as telomere length to count divisions"[8]. The data in question[9] show that by choosing a biopsy site with minimal photodamage and excluding donors with conditions such as diabetes known to promote cell turnover, there is no significant decline in division potential in vitro with age[9]. Put simply, if donors and biopsy sites with minimal cell turnover are selected, minimal cell turnover is observed.

These data support earlier observations on tissues with different turnover rates and illustrate the point that telomere erosion is the way fibroblasts count cell division, not the way human bodies count years[10,11]. It is a non-sequitur[8] to use data consistent with a slowly ticking mitotic clock to conclude that the clock is absent.

The concept that telomere erosion limits the division capacity of telomerase-negative cells is supported experimentally[3–5] and is also a logical and unavoidable outcome of the fundamental biology of telomeres and DNA replication[12]. As first postulated by one of us in 1971[13], terminal sequence loss during DNA replication is an unavoidable outcome of the enzymology of DNA polymerases. Functional telomeres are essential for long-term chromosome viability in eukaryotes. Telomerase-negative primary human cells show telomere erosion during in vitro culture at a rate similar to that seen in the telomerase knockout mouse in vivo[14]. Telomere erosion will eventually compromise an essential chromosomal element, and thus cell division in the absence of telomere maintenance must eventually lead to a situation incompatible with continued proliferation[15].

If one argues that there is no limit to cell division in vivo, one must then explain what feature of DNA replication or chromosome biology would allow a telomerase-negative cell in vivo with no telomere maintenance to have an unlimited division capacity. No one argues that every human cell shows the telomere-dependent senescence seen in fibroblasts, or that every animal cell behaves like its human equivalent[16].

The challenges for the future are to explore the details of telomere-dependent senescence, additional telomere-independent clocks such as those in rodents[17], the contribution cell senescence makes to human ageing, the role telomerase plays in allowing the high divisional capacities of some stem cells[18], and the ways in which cancer cells abrogate these barriers to unlimited cell division.

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Harry Rubin replies:

The authors and cosigners of the two letters[1,2] responding to my several critiques on cellular aging[3–6] have consistently ignored the main thrust of that critique, which is that primary fibroblasts senesce in culture in a stochastic manner rather than with a uniform, genetically fixed lifespan. Half of the clones undergo 16 divisions or less[7,8], rather than the uniform 50 for all the cells usually quoted in the senes-