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p53: Good Cop/Bad Cop

Minireview

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Activation of the p53 transcription factor in response to a variety of cellular stresses, including DNA damage and oncogene activation, initiates a program of gene expression that blocks the proliferative expansion of damaged cells. While the beneficial impact of the anticancer function of p53 is well established, several recent papers suggest that p53 activation may in some circumstances act in a manner detrimental to the long-term homeostasis of the organism. Here, we discuss the significant participation of p53 in three non-mutually exclusive theories of human aging involving DNA damage, telomere shortening, and oxidative stress. These "good cop/bad cop" functions of p53 appear to place it at the nexus of two opposing forces, cancer and aging. By extension, this relationship implies that therapies aimed to reduce cancer and postpone aging, and thereby increase longevity, will necessarily work either upstream or downstream, but not on the level of, p53.

The Good Cop

We long-lived metazoans find ourselves in a rough neighborhood. Each day, our component cells are bombarded by background ionizing radiation and UV sunlight, exposed to chemical mutagens such as aflatoxin, benzene, or N-nitrosamines, and infected by oncogenic pathogens such as EBV and HPV. Even in the absence of these external agents, there appear to be constant, significant levels of physiologic DNA damage with each cell division, which, if left unrepaired, can set the stage for tumor initiation. Proof that these tumorigenic influences are at work from cradle-to-grave comes from the PCR detection of occult oncogenic translocations such as TEL-AML1 in the blood of newborns (Mori et al., 2002) and the presence of low-grade prostate cancer foci in approximately 80% of men over the age of 80.

Over the last decade, it has become clear that the prime molecular policeman in repressing these miscreant elements is p53, the so-called "guardian of the genome." p53 enforces a variety of anticancer functions by encouraging cells to arrest or die in the face of DNA damage, hypoxia, oxidative stress, excessive mitogenic stimuli, or denuded telomeres (reviewed in Hahn and Weinberg, 2002). The preeminent importance of p53 in tumor suppression is underscored by the fact that any impairment of p53 function brought about by direct mutation, reduced gene dosage or by $p14^{ARF}$ inactivation or MDM2 overexpression is associated with increased tumor susceptibility. Therefore, we should be glad that p53 is at work keeping us safe from cancer, the worst sort of cellular organized crime. Unfortunately, however, just as an unfettered police force may erode civil liberties, so it now appears that excess p53 activity comes with an unwanted long-term cost: aging.

The Bad Cop

There are several, non-mutually exclusive theories of human aging, and it appears p53 has plausible links to three of the most familiar: unrepaired DNA damage, telomeric shortening, and oxidative stress.

DNA damage responses. An age-related decrease of DNA repair or increase in DNA damage has long been thought to play a role in human aging (Figure 1). For example, significant exposure to DNA damaging agents, such as chemo- or radiotherapy for malignancy, can induce aging-like phenotypes such as impaired wound healing, early menopause, alopecia, and secondary cancer predisposition. Also, genetic conditions with progeroid features such as ataxia-telangectasia, Fanconi anemia, and Werner and Bloom syndromes involve genes that sense DNA damage and/or participate in DNA repair processes. In mice, genetic deficiency of either nonhomologous end-joining (Ku80) or nucleotide excision repair (TTD), has been shown to engender signs of accelerated aging (de Boer et al., 2002; Vogel et al., 1999). Notably, both types of DNA damage (double-strand breaks and adducts) resulting from these genetic defects are sensed, at least in part, through p53. Thus, if human aging results from decreased DNA repair or increasing amounts of DNA damage with advanced age, it seems likely that p53 activation would play a role in the generation of age-related cellular responses.

Telomeres. Although the role of telomeric shortening in normal human aging is not well established, telomeric shortening likely contributes to end-organ failure in chronic diseases typified by increased cellular turnover (e.g., cirrhosis after chronic hepatitis). There is also evidence that compromised telomere maintenance may partially contribute to the impaired regenerative capacity and age-related phenotypes of some forms of dyskeratosis congenita (Vulliamy et al., 2001). In the mouse, the genetic analysis of telomere dynamics has established a link to cancer and aging with outcomes once again dictated by the status of p53. In mice with intact p53, critically shortened telomeres can constrain the cancer-prone condition of tumor suppressor mutations (Greenberg et al., 1999), yet elicit premature aging-like disorders such as decreased wound healing, premature graying, and diminished capacity to respond to acute and chronic stress (Rudolph et al., 1999). Strikingly, most of the organ compromise and morbidity associated with telomeric shortening can be attenuated by p53 loss (Chin et al., 1999). However, with elimination of p53 function, telomeric shortening is no longer an efficient anticancer mechanism, but rather fuels genomic instability and *accelerates* tumorigenesis (Chin et al., 1999). Therefore, in this system, p53 determines whether short telomeres will be sensed, leading to prematurely aged mice, or ignored, leading to tumor-prone animals. It seems likely that p53 would play a similar role in human dis-

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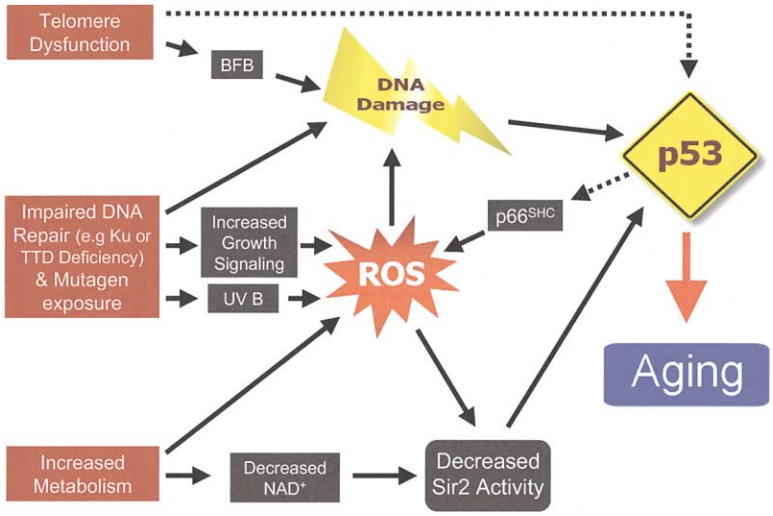

Figure 1. p53 Induces Aging in Response to Increased Metabolism, ROS, and DNA Damage

Telomere dysfunction engenders a bridge-fusion-breakage cycle (BFB) that induces a DNA-damage, p53-dependent checkpoint response. Telomere shortening may also induce p53 through a DNA-damage-independent mechanism (dotted line). Impaired DNA repair and mutagen exposure lead to unrepaired DNA damage, but also may be associated with increased ROS (e.g., as a result of UV exposure or increased growth signaling). Increased metabolism from caloric excess and/or insulin-like growth factor signaling induces ROS. The observation that Sir2 represses p53-mediated transactivation also suggests a DNA damage-independent link between metabolism and aging. Sir2 activity appears to be regulated by NAD levels and ROS levels as shown. $p66^{SHC}$ appears to be a downstream effector of p53 that mediates apoptosis and increases ROS levels. Loss of $p66^{SHC}$ leads to reduced levels of intracellular oxidants and extends murine lifespan, but $p66^{SHC-/-}$ mice are not tumor prone.

eases associated with critical telomeric shortening, as further evidenced by the impact of p53 in cell culture models of telomere dysfunction (Karlseder et al., 1999).

Debate exists as to whether the progeroid features of diseases like ataxia-telangectasia (associated with mutation of *ATM*) and Werner syndrome (associated with mutation in *WRN*) results from impaired DNA repair and/or altered telomere metabolism, since both ATM and WRN have been linked to processes of DNA repair and telomere maintenance. Interestingly, the deficiency in DNA repair and progeria are dissociated in *Atm*-deficient mice that display reduced DNA repair yet do not manifest the dramatic premature aging phenotype seen in AT patients. It could be that the biochemical function of ATM is partially fulfilled by some other protein in the mouse, or alternatively that differences in telomere reserve may influence the phenotype. Along these lines, mouse telomeres are much longer than those in humans, and the moderate degree of telomeric shortening evident in *Atm*-deficient mice would not be sufficient to activate p53. It will be of interest therefore, to determine if the effects of ATM loss would be altered in mice with shortened human-like telomeres, and thereby understand whether the progeroid features seen in AT patients result from altered telomere metabolism, impaired DNA repair, or both. Whichever theory of aging prevails in this disease, however, it seems likely that p53 will figure prominently as an important downstream mediator.

Oxidative Stress. Several lines of evidence from worms, flies, yeast, and mammals have linked oxidative stress, metabolism, and aging (reviewed in Guarente and Kenyon, 2000). Caloric restriction, or mutations that decrease glucose metabolism, will extend the lifespan of many species, and it has been suggested that in general a lower metabolic rate will decrease the production of toxic reactive oxygen species (ROS). In mammals at least, the response to oxidative stress appears to involve p53. While it has long been thought that p53 might sense free-radical-induced DNA damage, two direct links between ROS, metabolism, and p53 have recently been forged (Figure 1). One of the few genetic mutations known to increase the murine lifespan is loss of $p66^{SHC}$, a protein that appears to be involved in both the production of and the response to ROS (Migliaccio et al., 1999). In $p66^{SHC-/-}$ cells and mice, the p53-dependent apoptotic response to oxidative stress is impaired, and intracellular oxidants and oxidation-induced DNA damage are reduced (Migliaccio et al., 1999; Nemoto and Finkel, 2002; Trinei et al., 2002). $p66^{SHC}$ appears to be stabilized by p53, whereas p53 phosphorylation and acetylation are not affected in $p66^{SHC-/-}$ cells, and $p66^{SHC-/-}$ mice are not tumor prone (Trinei et al., 2002). These observations suggest that not only could p53 be a sensor of ROS, but also that p53 might regulate ROS levels (and therefore aging) through $p66^{SHC}$.

A further link between metabolism and aging has now been suggested by the observation that p53 interacts with, and is deacetylated by, the human ortholog of yeast Sir2 (Langley et al., 2002; Luo et al., 2001; Vaziri et al., 2001). Sir2 expression attenuates the ability of p53 to transactivate target genes and to induce growth arrest and apoptosis, while a dominant-negative form of Sir2 potentiates p53-mediated apoptosis in response to oxidative stress. It is not clear if this effect of Sir2 requires deactylation of p53 directly, or if Sir2 is tethered by p53 to the promoters of p53-responsive genes functioning to repress target gene transcription through the established histone deactylase activity of Sir2. The relationship between p53 and Sir2 is particularly intriguing, as *Sir2* encodes an NAD-dependent histone deacetylase whose overexpression extends the longevity of yeast, and whose function is required for lifespan extension by caloric restriction in this system (Lin et al., 2000). All of this has led to speculation that the dependence of Sir2 on NAD concentration links metabolism and oxidative stress to the activity of this enzyme (Guarente, 2001). In this model, free NAD is relatively scarce in times of caloric excess because it is sequestered for electron transport within the glycolytic pathway. In times of caloric restriction, however, NAD would be relatively abundant because of decreased glycolysis, which then would induce Sir2 activity and limit p53 activity. Of note, this

model of the role of p53 in aging does not depend upon DNA damage, as opposed to the aforementioned aging theories. However, it is not clear at present if Sir2 is predominantly regulated under physiologic conditions by NAD levels, ROS, or both. Nonetheless, these recent data clearly establish an interaction between p53 and Sir2, a protein known to modify longevity in lower organisms, and imply that Sir2 and NAD levels could couple oxidative stress and metabolism with p53 activity in mammals (Langley et al., 2002; Luo et al., 2001; Vaziri et al., 2001).

As these data suggest, p53 is of principal importance in sensing DNA damage, telomeric shortening, and oxidative stress (and in particular, its activity is regulated by the latter), and it seems clear that p53, acting as a "bad cop," contributes to mammalian aging. Direct evidence for this has been suggested by the generation of a hyperfunctional allele of p53 in the mouse germline (Tyner et al., 2002). These animals resist cancer, but develop an accelerated aging phenotype highly reminiscent of that seen in animals with DNA repair defects or telomere dysfunction. As these mice presumably have normal regulation of p53, with only increased transactivation of p53-dependent genes, this observation suggests that the level of p53 function determines the onset of aging phenotypes (and therefore sets longevity) in a direct way. This result is of particular importance as it suggests that some aspects of mammalian aging are not directly the consequence of ROS or damage to specific loci or genes per se, but rather stem from the organismal response to these factors. The conclusion suggested by these disparate lines of data is that p53, while cracking down on neoplasia, also limits the repair and regeneration of normal tissues.

Conclusions

The "good cop/bad cop" analogy of p53 raises the question of the extent to which aging depends on p53 per se. An extreme interpretation would be that all human aging is an untoward result of an anticancer mechanism. This view does not appear to be correct for several reasons. First, aging occurs in yeast, *Drosophila*, and *C. elegans*, whose life spans are generally not limited by tumorigenesis. Also, the p53 homologs of *C. elegans* and *Drosophila* are predominantly mediators of DNA damage-induced apoptosis without a clear role in the prevention of neoplasia. Therefore, aging seems to be older, in evolutionary terms, than either cancer or p53's role in preventing it. Likewise, there do not appear to be straightforward relationships, even among mammals, between life span, tumor susceptibility, and metabolic rates. In higher organisms, however, a principal cost of unrepaired DNA damage is increased tumorigenesis, and the p53-mediated checkpoint response to cellular stresses has been perfected in response to these evolutionary pressures. Donehower and colleagues have now shown that this de facto anticancer mechanism comes with an attendant cost of accelerated aging, independent of the stimulus of p53 induction (Tyner et al., 2002). Therefore, it seems fair to say that some, but certainly not all, aspects of human aging stem from the efforts of p53 to inhibit tumor growth.

It remains to be seen whether this new understanding of the dual role of p53 in cancer and aging will translate into therapeutic advances. In patients with cancer, where p53 activation is desirable and perhaps even necessary, the problem seems simpler. For example, our present new understanding of the downside of p53 activation might support the transient use of agents that prevent the induction of p53 or attenuate its function in specific clinical circumstances, for example to protect non-diseased tissues in patients undergoing chemo- or radiotherapy. In fact, Gudkov and coworkers have developed a small molecule inhibitor of p53, the use of which protects mice from the toxicity of radiotherapy, suggesting that many of the side effects of cancer treatment stem from p53's "bad cop" functions (Komarov et al., 1999). Likewise, several adjuncts to modern oncologic therapy (e.g., the administration of amifostine or autologous stem cell transplants) may work at the molecular level by sparing healthy tissue the effects of p53 activation. Of course, anticancer therapies that do not induce p53 have long been sought in medical oncology, but the success rate for identification of such compounds has been low. Despite the promise of receptor tyrosine kinase inhibitors, anti-angiogenesis therapy, and tumor vaccines, it is sobering to realize that nearly all curative regimens currently in use for advanced cancer depend at least in part on the use of DNA damaging agents.

In healthy individuals, however, the postponement of aging by attenuating p53 activity is a more difficult prospect, as the pharmacologic long-term inhibition of p53 function would be complicated by increased tumorigenesis. As $p66^{SHC-/-}$ mice do not appear to be tumor prone, Pelicci and colleagues have suggested it might be possible to extend longevity by targeting this downstream effector of p53 without perturbing other aspects of p53 function (Trinei et al., 2002). However, if $p66^{SHC}$ inhibition in humans induces some unappreciated toxicity, the remaining approaches to this problem appear to be upstream of p53 activation. Along these lines, Guarente has proposed that Sir2 has evolved as an adaptive means to extend longevity in times of reduced caloric intake and decreased metabolism in a variety of species (Guarente, 2001). If Sir2 does serve such a function in mammals as well as worms and yeast, then increased activity/expression of this protein would not be expected to increase tumorigenesis, despite attenuated p53 function. Last, if one were to limit DNA damage, oxidative stress and telomeric shortening, all oncogenic in some circumstances, both cancer *and* aging might be deferred. Clearly, remaining non-obese, avoiding genotoxins (e.g., UV light, tobacco), and consuming an adequate supply of anti-oxidants and NAD (also known as vitamin B3) would all appear reasonable medical advice. This plan, which is in essence "clean living," has long been ignored by humanity and additional insight into p53 function is not likely to rectify this problem. Perhaps, as our understanding of aging improves, it will be possible to offer more specific lifestyle advice or develop pharmacologic inhibitors of oxidative stress, DNA damage, and/or telomere shortening. In this way, we would reduce the workload of the "good cop" while keeping the "bad cop" off the streets.

Selected Reading

Chin, L., Artandi, S.E., Shen, Q., Tam, A., Lee, S.L., Gottlieb, G.J., Greider, C.W., and DePinho, R.A. (1999). Cell *97*, 527–538.

de Boer, J., Andressoo, J.O., de Wit, J., Huijmans, J., Beems, R.B., van Steeg, H., Weeda, G., van der Horst, G.T., van Leeuwen, W., Themmen, A.P., et al. (2002). Science *296*, 1276–1279.

Greenberg, R.A., Chin, L., Femino, A., Lee, K.H., Gottlieb, G.J., Singer, R.H., Greider, C.W., and DePinho, R.A. (1999). Cell *97*, 515–525.

Guarente, L. (2001). Trends Genet. *17*, 391–392.

Guarente, L., and Kenyon, C. (2000). Nature *408*, 255–262.

Hahn, W.C., and Weinberg, R.A. (2002). Nat. Rev. Cancer *2*, 331–341.

Karlseder, J., Broccoli, D., Dai, Y., Hardy, S., and de Lange, T. (1999). Science *283*, 1321–1325.

Komarov, P.G., Komarova, E.A., Kondratov, R.V., Christov-Tselkov, K., Coon, J.S., Chernov, M.V., and Gudkov, A.V. (1999). Science *285*, 1733–1737.

Langley, E., Pearson, M., Faretta, M., Bauer, U.M., Frye, R.A., Minucci, S., Pelicci, P.G., and Kouzarides, T. (2002). EMBO J. *21*, 2383–2396.

Lin, S.J., Defossez, P.A., and Guarente, L. (2000). Science *289*, 2126–2128.

Luo, J., Nikolaev, A.Y., Imai, S., Chen, D., Su, F., Shiloh, A., Guarente, L., and Gu, W. (2001). Cell *107*, 137–148.

Migliaccio, E., Giorgio, M., Mele, S., Pelicci, G., Reboldi, P., Pandolfi, P.P., Lanfrancone, L., and Pelicci, P.G. (1999). Nature *402*, 309–313.

Mori, H., Colman, S.M., Xiao, Z., Ford, A.M., Healy, L.E., Donaldson, C., Hows, J.M., Navarrete, C., and Greaves, M. (2002). Proc. Natl. Acad. Sci. USA *12*, 8242–8247.

Nemoto, S., and Finkel, T. (2002). Science *295*, 2450–2452.

Rudolph, K.L., Chang, S., Lee, H.W., Blasco, M., Gottlieb, G.J., Greider, C., and DePinho, R.A. (1999). Cell *96*, 701–712.

Trinei, M., Giorgio, M., Cicalese, A., Barozzi, S., Ventura, A., Migliaccio, E., Milia, E., Padura, I.M., Raker, V.A., Maccarana, M., et al. (2002). Oncogene *21*, 3872–3878.

Tyner, S.D., Venkatachalam, S., Choi, J., Jones, S., Ghebranious, N., Igelmann, H., Lu, X., Soron, G., Cooper, B., Brayton, C., et al. (2002). Nature *415*, 45–53.

Vaziri, H., Dessain, S.K., Ng Eaton, E., Imai, S.I., Frye, R.A., Pandita, T.K., Guarente, L., and Weinberg, R.A. (2001). Cell *107*, 149–159.

Vogel, H., Lim, D.S., Karsenty, G., Finegold, M., and Hasty, P. (1999). Proc. Natl. Acad. Sci. USA *96*, 10770–10775.

Vulliamy, T., Marrone, A., Goldman, F., Dearlove, A., Bessler, M., Mason, P.J., and Dokal, I. (2001). Nature *413*, 432–435.