An Encouraging Progress Report on the Treatment of Progeria and its Implications for Atherogenesis

Running title: Oshima et al.; Progeria clinical trial

Junko Oshima, MD, PhD¹; Fuki M. Hisama, MD²; George M. Martin, MD¹, ³

¹Dept of Pathology; ²Division of Medical Genetics, University of Washington, Seattle, WA;
³Molecular Biology Institute, University of California at Los Angeles, Los Angeles, CA

Address for Correspondence:
Junko Oshima, MD, PhD
Department of Pathology
University of Washington
Box 357470, HSB K-543B
Seattle, WA, USA 98195
Tel: 206-616-4227
Fax: 206-685-8356
E-mail: picard@u.washington.edu

Journal Subject Codes: Atherosclerosis:[89] Genetics of cardiovascular disease, Treatment:[118] Cardiovascular pharmacology

Key words: Hutchinson-Gilford progeria syndrome, farnesyltrasnferase inhibitor, Editorial, atherosclerosis, genetics, human, clinical trial
There are no known human genetic syndromes that faithfully accelerate all of the common phenotypes associated with aging, but there are several striking disorders with multiple features ("segmental progeroid syndromes"). The suffix –"oid" is conceptually important, as it leaves open the possibility that the underlying mechanisms may differ to some extent from what is considered the usual pathogenesis. Two canonical examples are the Werner syndrome (WS) and the Hutchinson-Gilford Progeria Syndrome (HGPS). In both disorders atherosclerosis is a particularly striking feature, causing the majority of deaths, usually via a myocardial infarction (although cancer is also a common cause of death in WS, a later onset disorder). WS is an autosomal recessive disorder due to null mutations at a member of the RecQ family of DNA helicases and is associated with accelerated clonal senescence of somatic cells and genomic instability. HGPS is caused by a specific class of autosomal dominant mutations in lamin A, a component of the nuclear membrane. The causative mutations result in the synthesis of large amounts of an alternatively spliced toxic isoform known as progerin; it is thus a gain of function mutation.

During the maturation of prelamin A, a farnesyl group is added to a cysteine residue at the C-terminus and subsequently removed by a proteolytic enzyme, Zmpste24, to produce mature lamin A. In progerin, the proteolytic site for Zmpste24 is deleted due to a cryptic splice site generated by the mutation. A mouse model expressing the non-farnesylated progerin exhibited near-normal phenotypes, supporting the interpretation that progerin toxicity is mediated by the farnesyl moiety. The severities of the disease correlate with the amount of progerin, a function of the activity of the cryptic splice site. The size of the deletion also contributes to the severity of the phenotype. The majority of classical early-onset forms of HGPS result from a 50 amino acid deletion. Attenuated forms of the disease have been associated with...
alternative deletions that lead to lesser amounts of the progerin isoform 7, 8.

Of major biomedical interest have been reports that small amounts of progerin can be detected in human tissues, including human arteries, and that these amounts increase with age 9, 10. These observations now raise the important question of the extent to which progerin contributes to human atherogenesis.

In this issue of Circulation, Gordon et al. 11 describe small but robust increases in the survival of HGPS patients following treatments that interfere with the post-translational farnesylation of lamin A proteins. Remarkably, given the extreme rarity of this disorder 12, this team, assisted by the efforts of a highly successful Progeria Research Foundation (http://www.progeriaresearch.org/), managed to assemble a cohort of 161 untreated control HGPS patients and 43 HGPS patients who were treated with either a single farnesyltransferase inhibitor (FTI), lonafarnib, or with two additional agents that also inhibited farnesylation: a bisphosphonate (zoledronate), which inhibits the synthesis of farnesyl-pyrophosphate and a statin (pravastatin), which inhibits HMG-CoA reductase (see Fig. 1 11)

Recruitment of control disease subjects did not require genotyping, although some are listed in Table S3 11. In contrast to the experience of our International Registry of Werner Syndrome, however, in which we could only provide a molecular diagnosis of ~80% of clinical diagnoses, molecular confirmations of clinically diagnosed HGPS is much more robust, probably well in excess of 95%. Moreover, all treated subjects were in fact genotyped, some of whom had LMNA mutations that resulted in lower levels of progerin (see Table S5 11).

It is important to note that the treated group included patients with ANY duration of treatment, either during a previous 2 year trial of FTI monotherapy or during an ongoing 3.5 year trial of combination treatment. Given that feature of the study, the Kaplan-Meier curves of Fig.

3
2B are even more impressive – 21/43 death in the untreated group vs. 5/42 deaths in the treated group during the approximately five year period of this study.

The authors suggest that the increased longevity of the treated HGPS patients was due to a reduction of cardiovascular pathology, a very plausible hypothesis. Investigators engaged in survival studies must always keep in mind, however, the possibility that any intervention with the potential to induce dietary restriction might have a more general effect on the pathophysiology of aging. This is extraordinarily unlikely in the present case, however, given the precise target of the interventional agents and the fact that there was no evidence of weight loss in a previous preliminary trial of monotherapy.

The authors point out that the present data now provide the best available baseline data of HGPS survival for use by interventional trials with other agents. Among these future potential interventions is research that utilized a mouse model of HGPS to evaluate phenotypic improvements via the targeting of an enzyme, isoprenylcysteine carboxyl methyltransferase (ICMT), which is responsible for the methylation of isoprenylcysteine. After farnesylation of prelamin A and progerin, farnesylcysteine is methylated by isoprenylcysteine carboxyl methyltransferase (ICMT) prior to cleavage by Zmpste24. Zmpste24 deficient mice expressing hypomorphic ICMT showed striking Ameriolations of HGPS-like phenotypes and upregulation of mTOR signaling. In contrast to those mTOR results, however, Cao et al. showed that long-term treatment of HGPS cells with the mTOR inhibitor, rapamycin, facilitated the autophagic clearance of progerin and reversed cellular HPGS phenotypes. Therefore, mTOR inhibitors could prove to be useful in the treatment of age-related disorders, including the treatment of WS.

Since the advent of drug-eluting coronary stents which deliver sirolimus or everolimus to prevent restenosis, the use of systemic mTOR inhibitors have been approved by the Food and Drug
Administration for the treatment of renal and brain manifestations of the genetic condition, tuberous sclerosis, as well as for the treatment of HER2 negative breast cancer, pancreatic neuroendocrine tumors, and advanced renal cell carcinoma. There is also much current interest in the use of “rapalogs”, chemical agents that may have fewer side effects – for example, upon immune suppression \(^{18}\). Finally, there are reports of the use of RNAi against progerin mRNA that showed improvements of cellular HGPS phenotypes\(^{19,20}\). Such gene therapy, however, may not be practical at this point because of the limitations of delivery systems.

Returning to the consideration of the most biomedically significant HGPS phenotype – atherosclerosis - there are two issues that remain to be resolved. The first are histopathological observations of apparent phenotypic discordances with atherosclerosis as it usually occurs. Of particular interest are the unusually extensive peri-arterial adventitial fibrosis and the involvement of vascular smooth muscle alterations\(^9\). The latter issue has been partially addressed by in vitro studies indicating that aging normal vascular smooth muscle cells reiterate the aging phenotype of HGPS fibroblasts in culture and that this process of “cell senescence” was accelerated by the accumulation of prelamin A\(^{21}\). The former observation will require more research, however.

The second issue is the need for more robust animal models of HGPS that include atherosclerosis. To the best of our knowledge, atherosclerosis has yet to be demonstrated in any of the several published mouse models of HGPS. The use of alternative animal models, such as the miniature pig, would be highly desirable\(^{22}\). 

\[DOI: 10.1161/CIRCULATIONAHA.114.010648\]
**Acknowledgments:** We wish to dedicate this editorial to the memory of Sam Berns, a highly intelligent and heroic HGPS patient and the son of the first author of the manuscript addressed by this editorial. For one of Sam’s inspirational addresses, the reader is referred to the TED talk he presented shortly after his 17th birthday: https://www.youtube.com/watch?v=36m1o-tM05g.

**Conflict of Interest Disclosures:** None.

**References:**


An Encouraging Progress Report on the Treatment of Progeria and its Implications for Atherogenesis
Junko Oshima, Fuki M. Hisama and George M. Martin

Circulation. published online May 2, 2014;
Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2014 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7322. Online ISSN: 1524-4539

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circ.ahajournals.org/content/early/2014/05/02/CIRCULATIONAHA.114.010648

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation is online at:
http://circ.ahajournals.org/subscriptions/