LAPTM5
A Novel Target in an Old Fight against Tubular Senescence

Michael S. Balzer

JASN 35: 1624–1626, 2024. doi: https://doi.org/10.1681/ASN.0000000524

The rising life expectancy and mankind's increasing desire to further extend human health span present the nephrology community with new challenges. These challenges include understanding not only physiological aging but also aging in the context of disease. Aging is driven by several biological processes, often referred to as the hallmarks of aging. These hallmarks meet three criteria: (*1*) They manifest as age progresses, (2) their acceleration hastens aging when intensified experimentally, and (*3*) therapeutic interventions targeting these hallmarks can slow, halt, or even reverse the aging process.[1] Within the kidneys, cellular senescence stands out as both a key feature of normal aging and a significant driver of kidney disease progression.[2,3]

Cellular senescence was first identified in the early 1960s by Hayflick and Moorhead, who observed cultured fibroblasts entering a state of irreversible cell cycle arrest while remaining viable. Senescent cells secrete a variety of factors collectively termed the senescence-associated secretory phenotype (SASP), which can influence nearby cells and tissues and have effects at distant sites. Senescence can be triggered by various stimuli, including, but not limited to, oncogene activation, DNA damage, oxidative and mechanical stress, mitochondrial dysfunction, and epigenetic changes.[4] Initially considered a mechanism for cancer suppression, senescence is now recognized as a contributor to aging and age-related diseases.[5] Recent research has focused on the accumulation of senescent cells in CKD and how this contributes to fibrosis, dysfunction, and higher mortality.[4]

Another important hallmark of aging is the disruption of protein homeostasis or proteostasis, leading to the accumulation of damaged or misfolded proteins. These proteins often aggregate into intracellular inclusion bodies or extracellular amyloid plaques.[1] The lysosome plays a crucial role in maintaining proteostasis by degrading these proteins through chaperone-mediated autophagy. In this process, proteins are first bound by heat shock protein HSC70 and then transported to lysosome-associated membrane proteins, which facilitate their translocation into the lysosome for degradation. Abnormalities in lysosomal function have been linked to increased cellular senescence and subsequent aging-related kidney fibrosis.

In this issue of *JASN*, Liu *et al.* investigate the role of LAPTM5, a lysosome-related protein initially identified for its function in immune cells, in a kidney parenchymal context.[6] The study reveals that *Laptm5* is significantly upregulated in kidney tubular epithelial cells in both human and murine CKD. Conditional knockout of *Laptm5* in kidney tubules results in reduced fibrosis in several CKD models, suggesting that LAPTM5 contributes to kidney fibrosis by promoting tubular senescence. Mechanistically, LAPTM5 appears to inhibit WWP2-mediated ubiquitination of NICD1, the active intracellular domain of Notch1, which plays a role in regulating cellular senescence.

The authors first demonstrated that LAPTM5 was expressed at relatively low levels in the kidney under normal conditions. However, its expression was significantly upregulated in mouse models of CKD, including aristolochic acid nephropathy, ischemia-reperfusion injury, and unilateral ureteral obstruction. LAPTM5 was shown to colocalize with markers for proximal tubules, distal convoluted tubules, and macrophages, indicating that its expression was increased in these cells during CKD. *In vitro* experiments confirmed that LAPTM5 expression was upregulated in response to stimuli such as aristolochic acid and TGF-β in a rat kidney epithelial cell line.

Having established that global *Laptm5* knockout mice were protected against kidney injury and fibrosis, the authors generated tubule-specific *Laptm5* knockout mice to explore the role of LAPTM5 in kidney fibrosis more precisely. These mice exhibited reduced markers of senescence, such as senescence-associated β-galactosidase (SA-β-gal), p53, p21, and p16, along with decreased SASP production in aged mice. In CKD models (aristolochic acid nephropathy, ischemia-reperfusion injury, and unilateral ureteral obstruction), *Laptm5*-deficient mice had less kidney damage, both functionally and structurally, and showed reduced signs of senescence, including lower levels of SA-β-gal and γ-H2A.X, and higher levels of antiaging protein Klotho. Human kidney tissue samples from patients with diabetic kidney disease and hypertensive

Department of Nephrology and Medical Intensive Care, Charité – Universitätsmedizin Berlin, Berlin, Germany; and Berlin Institute of Health at Charité – Universitätsmedizin Berlin, BIH Biomedical Innovation Academy, BIH Charité Clinician Scientist Program, Berlin, Germany

Correspondence: Dr. Michael S. Balzer, email: michael-soeren.balzer@charite.de

Published Online Ahead of Print: October 15, 2024

See related article, "Lysosomal-Associated Protein Transmembrane 5, Tubular Senescence, and Progression of CKD," on pages 1655–1670.

nephropathy supported these findings, showing a correlation between LAPTM5 expression and markers of tubular senescence. Notably, LAPTM5 levels were inversely correlated with eGFR, although the study did not examine correlations with fibrosis score.

The study also investigated the interaction between senescent tubular epithelial cells and interstitial fibroblasts, which are key players in fibrosis. *In vitro* experiments showed that senescent epithelial cells expressing high levels of LAPTM5 could activate fibroblasts, a process reversed by silencing *Laptm5*. *In vivo* fibroblasts in *Laptm5*-deficient mice showed reduced activation, further suggesting that LAPTM5 promotes fibroblast activation and fibrosis through a paracrine mechanism.

The authors then explored the molecular mechanisms underlying these observations, focusing on Notch signaling. *Laptm5* overexpression increased levels of NICD1, the active intracellular domain of Notch1, whereas *Laptm5* knockdown reduced NICD1 levels. Importantly, this effect was post-transcriptional, as *Notch1* mRNA levels remained unchanged. Further experiments revealed that LAPTM5 inhibited the ubiquitination and subsequent degradation of NICD1 by promoting the lysosomal degradation of WWP2, an E3 ubiquitin ligase. This finding was supported by mass spectrometry and coimmunoprecipitation studies showing a direct interaction between LAPTM5 and WWP2. Disrupting this interaction led to increased NICD1 ubiquitination and degradation, preventing the accumulation of NICD1 and reducing senescence.

To confirm the relevance of these findings *in vivo*, the authors used shRNA to knock down *Laptm5* in CKD mice and observed protective effects against kidney fibrosis. This supports the notion that LAPTM5 plays a central role in promoting senescence and fibrosis in CKD, and targeting this pathway could offer a novel therapeutic strategy for preventing CKD progression.

Despite the promising results, the study has some limitations. First, accurately identifying senescent cells remains a challenge. According to the 2019 consensus guidelines from the International Cell Senescence Association,[7] identifying senescence requires multiple criteria, including the absence of proliferation markers in cells expressing SA-β-gal or lipofuscin, along with increased expression of p16 and p21 and decreased lamin B1 expression. In addition, a full SASP signature should be demonstrated. No single marker can definitively identify a senescent cell, as certain markers, such as SA-β-gal, can also be expressed by other cell types like macrophages. Multiple tissue types upregulate p16 and p21, particularly in the context of cellular proliferation.[4] Meticulous exclusion of proliferative signatures, which have been described previously for the CKD models the authors used in their study, are important going forward. Of note, single-cell methodologies have tremendously advanced our ability to differentiate cell phenotypes of adaptation, maladaptation, senescence, and proliferation on a whole transcriptome–level, aiding in the identification of senescent cells.[8,9]

Second, the human CKD samples analyzed in this study were exclusively from patients with diabetic kidney disease and hypertensive nephropathy. These forms of CKD are often characterized by glomerular injury, raising questions about the cell types contributing to the observed changes in bulk RNA sequencing data. Given that LAPTM5 expression is much higher in immune and endothelial cells than in tubular cells, as shown by recent single-cell RNA-seq datasets, it is not yet clear whether targeting of LAPTM5 would primarily affect tubular cells. This specificity is crucial for the development of therapies against senescent cells in kidney disease with fewer off-target effects and reduced toxicity. Nevertheless, the authors provide compelling evidence that LAPTM5 plays a role in tubular senescence and kidney health, even as its absolute expression levels are relatively low in these cells.

In summary, this study explored a novel mechanism in CKD involving the lysosome-associated protein LAPTM5, linking tubular senescence to CKD progression. Mechanistically, LAPTM5 promoted lysosomal degradation of WWP2, leading to reduced ubiquitination and accumulation of NICD1 in tubular cells, facilitating a senescent phenotype for paracrine activation of fibroblasts. By highlighting LAPTM5 as a key mediator of tubular epithelial cell senescence and kidney fibrosis, this study opens new avenues for research and potential therapeutic interventions targeting LAPTM5 to slow or prevent CKD progression. Future studies should explore how LAPTM5 expression is regulated in response to different CKD stimuli, such as the potential role of transcription factor E-box binding,[10] and further investigate the molecules involved in cell–cell communication between senescent tubular cells and fibroblasts.

Disclosures

Disclosure forms, as provided by each author, are available with the online version of the article at http://links.lww.com/JSN/E862.

Funding

M.S. Balzer: Deutsche Forschungsgemeinschaft, Else Kröner-Fresenius-Stiftung, Dr. Werner Jackstädt-Stiftung, and Berlin Institute of Health.

Acknowledgments

The author apologizes to all researchers whose work could not be cited owing to the limited number of allowed citations. Because Dr. Michael S. Balzer is an Editorial Fellow of *JASN*, he was not involved in the peer-review process for this manuscript. Another editor oversaw the peer-review and decision-making process for this manuscript.

The content of this article reflects the personal experience and views of the author and should not be considered medical advice or recommendation. The content does not reflect the views or opinions of the American Society of Nephrology (ASN) or *JASN*. Responsibility for the information and views expressed herein lies entirely with the author.

Author Contributions

Conceptualization: Michael S. Balzer.
Writing – original draft: Michael S. Balzer.
Writing – review & editing: Michael S. Balzer.

References

1. López-Otín C, Blasco MA, Partridge L, Serrano M, Kroemer G. Hallmarks of aging: an expanding universe. *Cell.* 2023;186(2):243–278. doi:10.1016/j.cell.2022.11.001
2. Ferenbach DA, Bonventre JV. Mechanisms of maladaptive repair after AKI leading to accelerated kidney ageing and CKD. *Nat Rev Nephrol.* 2015;11(5):264–276. doi:10.1038/nrneph.2015.3
3. Rex N, Melk A, Schmitt R. Cellular senescence and kidney aging. *Clin Sci.* 2023;137(24):1805–1821. doi:10.1042/CS20230140
4. Mylonas KJ, Ferenbach DA. Targeting senescent cells as therapy for CKD. *Kidney360.* 2024;5(1):142–151. doi:10.34067/KID.0000000000000316
5. Campisi J, d'Adda di Fagagna F. Cellular senescence: when bad things happen to good cells. *Nat Rev Mol Cell Biol.* 2007;8(9):729–740. doi:10.1038/nrm2233
6. Liu X, Zhan P, Zhang Y, et al. Lysosomal-associated protein transmembrane 5, tubular senescence, and progression of CKD. *J Am Soc Nephrol.* 2024;35(12):1655–1670. doi:10.1681/ASN.0000000000000446
7. Gorgoulis V, Adams PD, Alimonti A, et al. Cellular senescence: defining a path forward. *Cell.* 2019;179(4):813–827. doi:10.1016/j.cell.2019.10.005
8. O'Sullivan ED, Mylonas KJ, Bell R, et al. Single-cell analysis of senescent epithelia reveals targetable mechanisms promoting fibrosis. *JCI Insight.* 2022;7(22):e154124. doi:10.1172/jci.insight.154124
9. Balzer MS, Doke T, Yang YW, et al. Single-cell analysis highlights differences in druggable pathways underlying adaptive or fibrotic kidney regeneration. *Nat Commun.* 2022;13(1):4018. doi:10.1038/s41467-022-31772-9
10. Yuan H, Zheng C, Zhu L, et al. Contribution of TFEB-mediated autophagy to tubulointerstitial fibrosis in mice with adenine-induced chronic kidney disease. *Biomed Pharmacother.* 2021;133:110949. doi:10.1016/j.biopha.2020.110949