PD-1, a New Player in Podocyte Age-Related Senescence

Paulina X. Medina Rangel, PhD, Shuta Ishibe, MD

PII: S0272-6386(22)01042-3

DOI: https://doi.org/10.1053/j.ajkd.2022.10.007

Reference: YAJKD 57817

To appear in: *American Journal of Kidney Diseases*

Received Date: 26 September 2022

Accepted Date: 6 October 2022

Please cite this article as: Medina Rangel PX, Ishibe S, PD-1, a New Player in Podocyte Age-Related Senescence, *American Journal of Kidney Diseases* (2022), doi: https://doi.org/10.1053/j.ajkd.2022.10.007.

PD-1, a New Player in Podocyte Age-Related Senescence

Paulina X. Medina Rangel, PhD, and Shuta Ishibe, MD

Yale University School of Medicine, Section of Nephrology, New Haven, Connecticut, USA 06520

Corresponding Author-contact shuta.ishibe@yale.edu

Commentary on Pippin, JW, Kaverina, N, Wang, Y, Eng, DG, Zeng, Y, Tran, U, Loretz, CJ, Chang, A, Akilesh, S, Poudel, C, Perry, HS, O'Connor, C, Vaughan, JC, Bitzer, M, Wessely, O, Shankland, SJ: Upregulated PD-1 signaling antagonizes glomerular health in aged kidneys and disease. J Clin Invest, 2022;132(16):e156250. doi:10.1172/JCI156250.

Life expectancy has improved considerably over the past decade and consequently the aging population continues to increase. As one ages, kidney function decreases, as evidenced by the decline in the glomerular filtration rate (GFR), due to nephron loss. The loss of podocytes, the post-mitotic cells that line the outermost surface of the glomerulus, is thought to be a contributor to this process. An unmet need exists to understand the molecular mechanism of podocyte aging and develop therapeutic interventions to delay the process of age-related declines in kidney function.

In a recent issue of JCI, Pippin *et al* identified the importance of podocyte associated PD-1 signaling in aging-related changes in the kidney and the therapeutic potential of interfering with PD-1 signaling.[1] PD-1 (programmed cell death protein) and its ligands, PD-L1 and PD-L2 represent one of the immune checkpoints that prevent autoimmunity. The activation of PD-1/PD-L1 signaling is utilized by many malignant cells to evade detection by T cells.[2] This has led to development of immune checkpoint inhibitors for PD-1 or PD-L1, that restore T cell recognition to detect tumor cells, and are widely used in cancer therapy. While PD-1 signaling has been extensively studied in cancer research, little is known about its role in non tumor cells, such as the podocyte.

What Does This Important Study Show?

Pippin *et al* demonstrate that PD-1 may play a vital role in age-related kidney disorders.[1, 3] They first identified podocyte associated PD-1 upregulation in aged mice using unbiased RNA-seq analysis and observed a positive correlation between PD-1 pathway expression and the decline of kidney function. By blocking PD-1/PD-L1 signaling using an antibody directed against PD-1 (aPD1ab), the authors show that

podocyte senescence and the inflammatory phenotype improved, resulting in an enhancement of podocyte life span. Moreover, the authors performed transcriptome profiling of aPD1ab-treated mice and elucidated that the primary effects of interfering with PD-1 signaling was the restoration of the podocyte metabolic profile (*i.e.* oxidative phosphorylation, amino acid, fatty acid and glucose metabolism).

This study demonstrates in a very elegant manner that aged podocytes exhibit authentic features of senescence including SA-β-galactosidase activity,[4] altered metabolism,[5] and positive immunoreactivity for p16 and p19.[6, 7] The hallmark of senescent cells is the secretion of proinflammatory cytokines, growth factors and matrix metalloproteases, a phenomenon known as the senescence-associated secretory phenotype (SASP).[8] The SASP plays an essential role in reinforcing senescence in an autocrine and paracrine fashion and stimulates the recruitment of the immune system to eliminate damaged cells.[8, 9] Interestingly, in this investigation aged podocytes used PD-1/PD-L1 expression that is also used by cancer cells to evade being detected by T-cells.

One interpretation of these captivating results is that podocytes may use the PD-1 mechanism to facilitate their surveillance and avoid being cleared by the immune system, since as post-mitotic cells, podocytes cannot be replaced by new cells, thus their loss would be detrimental.[10] The renoprotection by aPD1ab may be due to senescent podocytes being unable to escape the immune system and thus being eliminated. The fact that injured podocytes can damage healthy podocytes and other glomerular cells has been hypothesized.[11] In the absence of senescent podocyte, there is no longer SASP that potentially affects the neighboring cells, resulting in a better glomerular health [Figure 1]. Furthermore, the elimination of senescent podocytes with aPD1ab can explain the improvement of the podocyte metabolic profile. Interestingly, a similar mechanism may occur in the liver, where senescent hepatocytes also benefit from the inhibition of PD-1 activity as the authors observed. In addition, cellular senescence of hepatocytes has been reported to be involved in several liver diseases.[12]

Additional investigation utilizing a podocyte specific conditional knockout of PD-1 would further validate these findings and its role in the aged kidney. Since there is a lack of PD-1 expression in the young glomerulus, a phenotype in the PD-1 KO model at early life stages would not be expected. However, as the mice ages, loss of PD-1 in podocytes may prolong kidney health by preventing podocyte induced injury. Further studies that elucidate the role of PD-1 in cellular senescence will be highly valuable.

How Does This Study Compare with Prior Studies?

With an aging population that continues to increase, it is not surprising that over the past five years, the notion of podocyte senescence has become an increasing focus of research. Podocyte senescence has emerged as a new mechanism that may lead to podocyte loss and a decline kidney function; therefore, several investigations have attempted to elucidate its mechanism. Some studies have identified the downregulation of 'protective' genes in the aged podocytes. Lee and colleagues observed a reduction of sirtuin 1 (*Sirt1*) in podocytes of aging mice.[13] *Sirt1*, a longevity gene, was shown to protect podocytes from oxidative stress, inflammation, and regulate the activity of many transcriptional factors. In a similar manner, Zhang *et al* demonstrated the protective role of C/EBPα, a member a family of transcription factors of C/EBPs that regulate energy metabolism, inflammation, and autoimmunity in podocytes. Moreover, the loss of C/EBPα in podocytes resulted in glomerulosclerosis.[14]

Pippin *et al* correlated podocyte-specific PD-1 upregulation with the aged phenotype, while Fang *et al* demonstrated that glycogen synthase kinase 3β (GSK3β) is also overexpressed in senescent podocytes resulting in a functional and histological decline observed with aging.[15] The authors identified the importance of GSK3β, a major molecular target of lithium which has been shown to delay aging and increase longevity.

Furthermore, GSK3β has also been identified as a key upstream kinase that regulates PD-1 expression and that the inhibition of GSK3β *in vivo* blocked PD-1 expression.[16] These findings together further suggest

that GSK3β and PD-1 may modulate podocyte senescence through a similar pathway. Moreover, the fact that the genes identified as being down or up-regulated in the aged podocytes play a role in podocyte metabolism and inflammation, suggest that podocyte senescence signaling may occur through a similar cellular processes.

What Are the Implications for Nephrologists?

The study by Pippin *et al* demonstrated that pharmacological antagonism of the PD-1 pathway has major benefits on podocyte lifespan and kidney histology. Yet, adverse renal effects have been shown by onco-nephrologists when using immune checkpoint inhibitors in cancer therapy including glomerular damage, acute kidney injury and acute interstitial nephritis.[17, 18] The mechanisms underlying the kidney complications are believed to be related to T-cell activation and infiltration in the kidney. Perhaps, targeted therapies capable of blocking PD-1 specifically in podocytes in the future may be needed. Furthermore, the essence of mouse models may not necessarily mean that blocking PD-1 will have a clinical role in patients but opens new and exciting discoveries about the role of immune checkpoints in podocyte-aging. Hopefully this study will motive further investigations to elucidate the role of the immune system in aging, the mechanism of podocyte senescence, or the identification of the SASP molecules that are responsible for glomerular damage.

In sum, the beauty of these discoveries also lies in the fact that the authors demonstrated that PD-1 signaling is not only involved in the podocyte aging, but also in diseases such as focal segmental glomerulosclerosis (FSGS). Accordingly, blocking PD-1 activity improved the outcome of FSGS in murine models. These findings suggest that what we learn from podocyte aging may also have implications for other glomerular diseases including collapsing glomerulopathy, minimal change disease and diabetic glomerulopathy, opening a new field of study in podocytes for investigators worldwide.

Article Information

Support: NIH RO1DK083294 and RO1DK093269 (SI).

Financial Disclosure: The authors declare that they have no relevant financial interests.

Peer Review: Received September 26, 2022 in response to an invitation from the journal. Accepted October 6, 2022 after editorial review by an Associate Editor and a Deputy Editor.

References

1. Pippin, JW, Kaverina, N, Wang, Y, Eng, DG, Zeng, Y, Tran, U, Loretz, CJ, Chang, A, Akilesh, S, Poudel, C, Perry, HS, O'Connor, C, Vaughan, JC, Bitzer, M, Wessely, O, Shankland, SJ: Upregulated PD-1 signaling antagonizes glomerular health in aged kidneys and disease. *J Clin Invest,* 2022;132(16):e156250. doi:10.1172/JCI156250.
2. Sharpe, AH, Pauken, KE: The diverse functions of the PD1 inhibitory pathway. *Nat Rev Immunol,* 18**:** 153-167, 2018.
3. Yu, SM, He, JC: Aged glomeruli: a link between PD-1 and podocytes. *J Clin Invest,* 2022;132(16):e162330. doi:10.1172/JCI162330
4. Lee, BY, Han, JA, Im, JS, Morrone, A, Johung, K, Goodwin, EC, Kleijer, WJ, DiMaio, D, Hwang, ES: Senescence-associated beta-galactosidase is lysosomal beta-galactosidase. *Aging Cell,* 5**:** 187-195, 2006.
5. Wiley, CD, Campisi, J: From Ancient Pathways to Aging Cells-Connecting Metabolism and Cellular Senescence. *Cell Metab,* 23**:** 1013-1021, 2016.
6. Haber, DA: Splicing into senescence: the curious case of p16 and p19ARF. *Cell,* 91**:** 555-558, 1997.
7. Krishnamurthy, J, Torrice, C, Ramsey, MR, Kovalev, GI, Al-Regaiey, K, Su, L, Sharpless, NE: Ink4a/Arf expression is a biomarker of aging. *J Clin Invest,* 114**:** 1299-1307, 2004.
8. Coppe, JP, Desprez, PY, Krtolica, A, Campisi, J: The senescence-associated secretory phenotype: the dark side of tumor suppression. *Annu Rev Pathol,* 5**:** 99-118, 2010.
9. Acosta, JC, Banito, A, Wuestefeld, T, Georgilis, A, Janich, P, Morton, JP, Athineos, D, Kang, TW, Lasitschka, F, Andrulis, M, Pascual, G, Morris, KJ, Khan, S, Jin, H, Dharmalingam, G, Snijders, AP, Carroll, T, Capper, D, Pritchard, C, Inman, GJ, Longerich, T, Sansom, OJ, Benitah, SA, Zender, L, Gil, J: A complex secretory program orchestrated by the inflammasome controls paracrine senescence. *Nat Cell Biol,* 15**:** 978-990, 2013.
10. Reiser, J, Sever, S: Podocyte biology and pathogenesis of kidney disease. *Annu Rev Med,* 64**:** 357-366, 2013.
11. Wiggins, RC: The spectrum of podocytopathies: a unifying view of glomerular diseases. *Kidney Int,* 71**:** 1205-1214, 2007.
12. Aravinthan, AD, Alexander, GJM: Senescence in chronic liver disease: Is the future in aging? *J Hepatol,* 65**:** 825-834, 2016.
13. Chuang, PY, Cai, W, Li, X, Fang, L, Xu, J, Yacoub, R, He, JC, Lee, K: Reduction in podocyte SIRT1 accelerates kidney injury in aging mice. *Am J Physiol Renal Physiol,* 313**:** F621-F628, 2017.
14. Zhang, L, Zhou, F, Yu, X, Zhu, Y, Zhou, Y, Liu, J, Liu, Y, Ma, Q, Zhang, Y, Wang, W, Chen, N: C/EBPalpha deficiency in podocytes aggravates podocyte senescence and kidney injury in aging mice. *Cell Death Dis,* 10**:** 684, 2019.
15. Fang, Y, Chen, B, Gong, AY, Malhotra, DK, Gupta, R, Dworkin, LD, Gong, R: The ketone body beta-hydroxybutyrate mitigates the senescence response of glomerular podocytes to diabetic insults. *Kidney Int,* 100**:** 1037-1053, 2021.

16. Taylor, A, Harker, JA, Chanthong, K, Stevenson, PG, Zuniga, EI, Rudd, CE: Glycogen Synthase Kinase 3 Inactivation Drives T-bet-Mediated Downregulation of Co-receptor PD-1 to Enhance CD8(+) Cytolytic T Cell Responses. *Immunity,* 44**:** 274-286, 2016.
17. Gupta, S, Cortazar, FB, Riella, LV, Leaf, DE: Immune Checkpoint Inhibitor Nephrotoxicity: Update 2020. *Kidney360,* 1**:** 130-140, 2020.
18. Shankland, SJ, Pippin, JW, Wessely, O: Is treating with anti PD-1 to improve glomerular health come without a cost? Reply. *J Clin Invest*, 2022.

Figure legend

Figure 1- PD-1 expression and inhibition in podocyte senescence. **(A)** Senescent podocytes express PD-1 and affect surrounding cells through the senescence-associated secretory phenotype (SASP). Podocytes may express PD-1 to evade detection by immune cells. **(B)** An antibody directed against PD-1 (aPD1ab) allows senescent podocytes to be recognized by the immune system and be cleared. In the absence of senescent podocytes, there is no longer SASP that affect other glomerular cells. GBM, glomerular basement membrane