Reviewer’s report

Title: Renal Injury is Accelerated by Global Hypoxia-inducible Factor 1 Alpha Deficiency in a Mouse Model of STZ-induced Diabetes

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Reviewer: Pinelopi Kapitsinou

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Authors

In the present study, Bohuslavova and colleagues have investigated the role of HIF-1α in the early development of diabetic kidney disease. To address this issue, the authors induced diabetes by streptozotocin administration in mice lacking one copy of the Hif-1 gene (Hif1a+/-). Remarkably, the mutants showed increased susceptibility to diabetes as indicated by higher glucose levels than controls. Focusing on the early diabetic kidney phenotype, the authors have observed higher serum creatinine and phosphorous levels in Hif1a+/- mice compared to controls at 6 weeks after induction of diabetes. Morphologic analysis showed similar amounts of advanced glycation end products, collagen content as well as αSMA and pH3 expression in diabetic Hif1a+/- mice and controls. On the other hand, glomeruli in Hif1a+/- mice demonstrated higher VEGF expression and significant reduction in the number of mature podocytes assessed by WT1 staining. Finally, gene expression analysis showed small reduction in the expression of HIF-1 regulated genes in Hif1a+/- kidneys at baseline while Adm, Nphs2 and Sox9 transcripts were significantly up-regulated in Hif1a+/- mice compared to controls following induction of diabetes.

Critique

The role of hypoxia signaling in diabetic kidney disease remains understudied and there are major gaps in understanding how hypoxia modulates diabetic complications. Therefore, the present study addresses a significant issue and the application of a genetic model provides a powerful approach to examine the role of HIF-1 in the diabetic kidney disease. Overall, the paper is well written with appropriate methodology and statistical analysis. I have the following suggestions.

1. Can the authors provide measurements of albuminuria? Given that they detect changes mainly in the glomeruli, ACR would be a more direct readout of the diabetic kidney damage.

2. How do the authors explain the higher creatinine values in the Hif1a+/- mice? Creatinine overall correlates with tubulointerstitial damage, which though seems to be similar in diabetic Hif1a+/- mice and controls at this stage.

4. Timing and context may have critical impact on HIF activation in different models of diabetes. Could the authors characterize the HIF response in diabetic kidneys at 6 weeks following STZ administration? HIF-1 immunostaining or immunoblot could be informative.

5. How do the authors explain the increased expression of VEGF in the glomeruli of Hif1a+/- mice? How do they reconcile their findings with prior studies demonstrating diminished VEGF expression in diabetic tissues (e.g. Chou et al. Circulation 2002;105: 373-379, Frank et al J Biol Chem 1995;270:12607-12613 and Thangarajah et al. PNAS 2009;106:13505-13510)?

Are the methods appropriate and well described?
If not, please specify what is required in your comments to the authors.

Yes

Does the work include the necessary controls?
If not, please specify which controls are required in your comments to the authors.

Yes

Are the conclusions drawn adequately supported by the data shown?
If not, please explain in your comments to the authors.

Yes

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