Response to Comment on: Thallas-Bonke et al. (2008) Inhibition of NADPH Oxidase Prevents Advanced Glycation End Product–Mediated Damage in Diabetic Nephropathy Through a Protein Kinase C-α–Dependent Pathway: *Diabetes* 57:460–469, 2008

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We thank Dr. Yamagishi for his constructive comments (1) on our recent article (2). In response, we agree that it is possible for protein kinase C–α (PKC-α) to be a downstream signaling molecule of NADPH oxidase, further validating this PKC isoform as an important therapeutic target for the treatment of progressive diabetic nephropathy. In the schema of Fig. 7 in our recent study, a question mark was attached to the arrow, suggesting that NADPH oxidase is downstream of PKC-α. However, this issue remains to be fully clarified. Indeed, in our previous study (3), we reported that the renoprotective effects of treatments attenuating the accumulation of advanced glycation end products (AGEs), such as alagebrium, may in part occur via inhibition of PKC-α activation. Dr. Yamagishi raises the possibility that PKC-α activation may be a downstream event of the production of reactive oxygen species (ROS) via NADPH oxidase. We agree that this postulate cannot be excluded but is currently based primarily on in vitro studies.

We did not present any data in our study indicating that inhibition of PKC-α suppresses in vivo superoxide generation and reduces levels of phosphorylated $p47^{phox}$. This relates to the lack of availability of a specific PKC-α inhibitor for use in diabetic animal models and, therefore, we must rely on studies from the PKC-α knockout model (4) to determine whether PKC-α is an appropriate therapeutic target. It is likely that PKC-α inhibition in diabetes would reduce albuminuria and renal vascular endothelial growth factor expression end points that were specifically examined in our study. Therefore, we agree that the overall theme of Dr. Yamagishi's letter emphasizing important bidirectional interactions among putative mediators of renal injury in diabetes is consistent with our own work, suggesting that combination therapies able to target multiple pathways are likely to ultimately be more successful than those targeting one specific molecule/pathway (5).

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