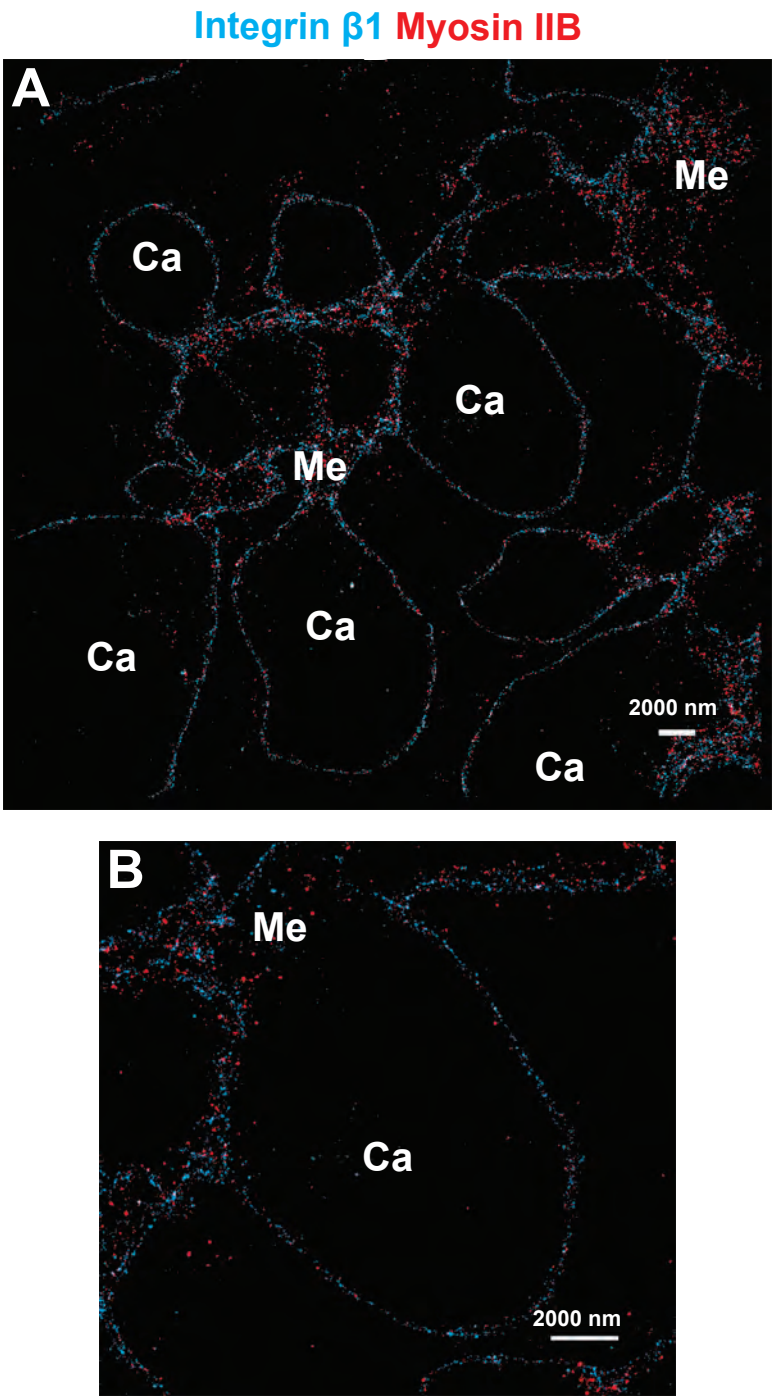
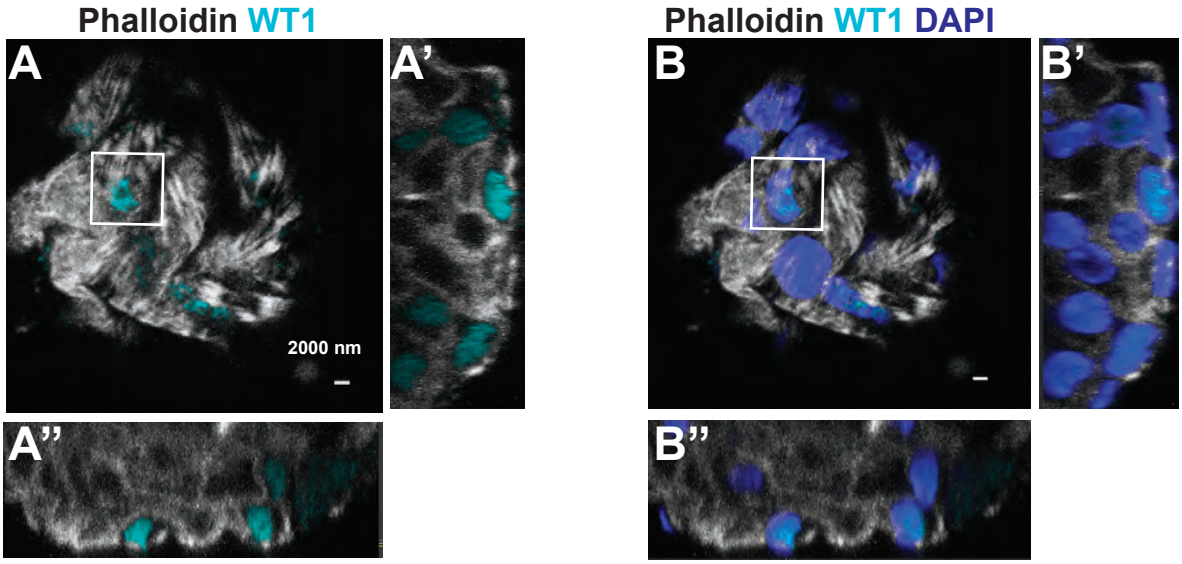
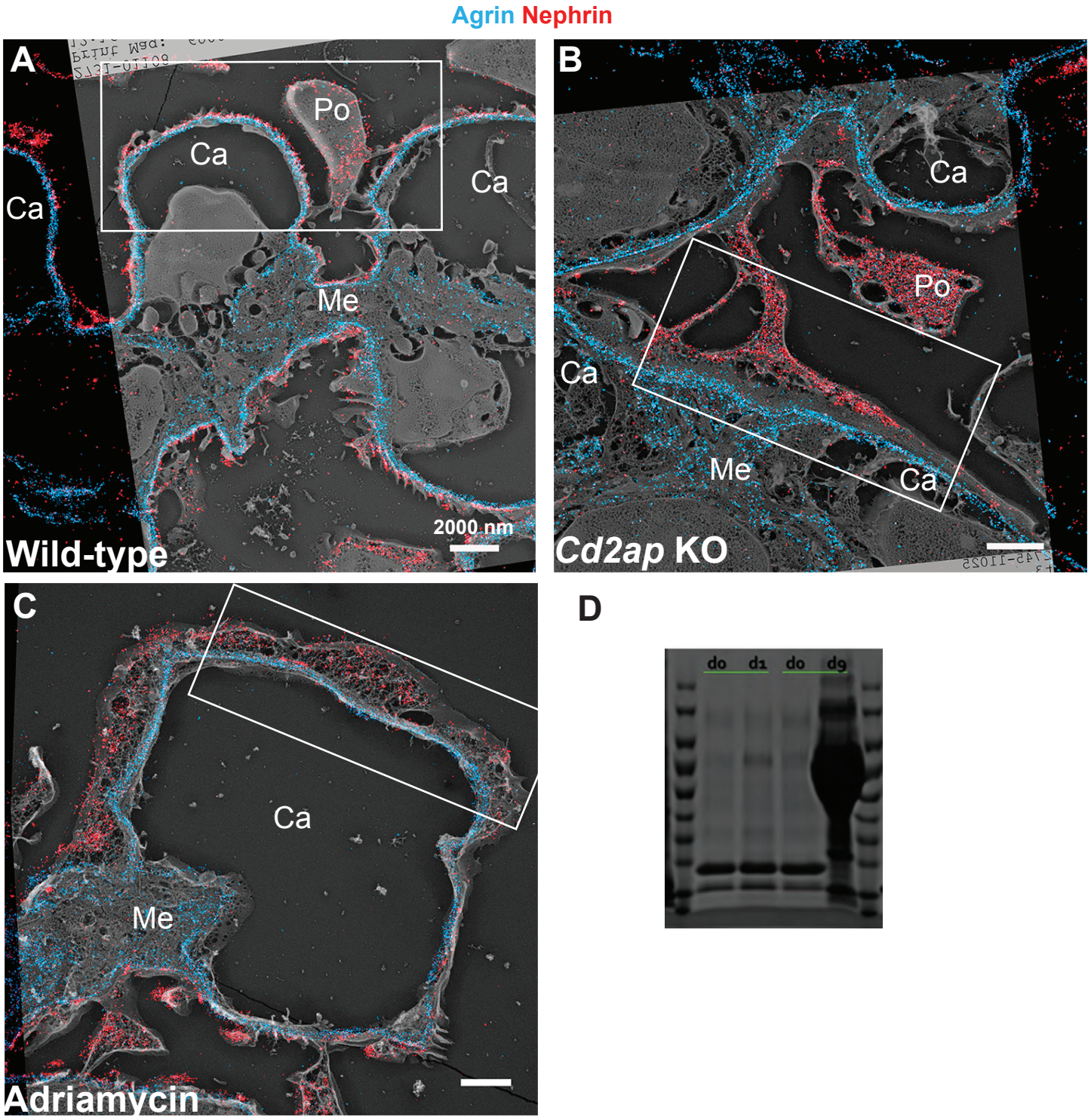


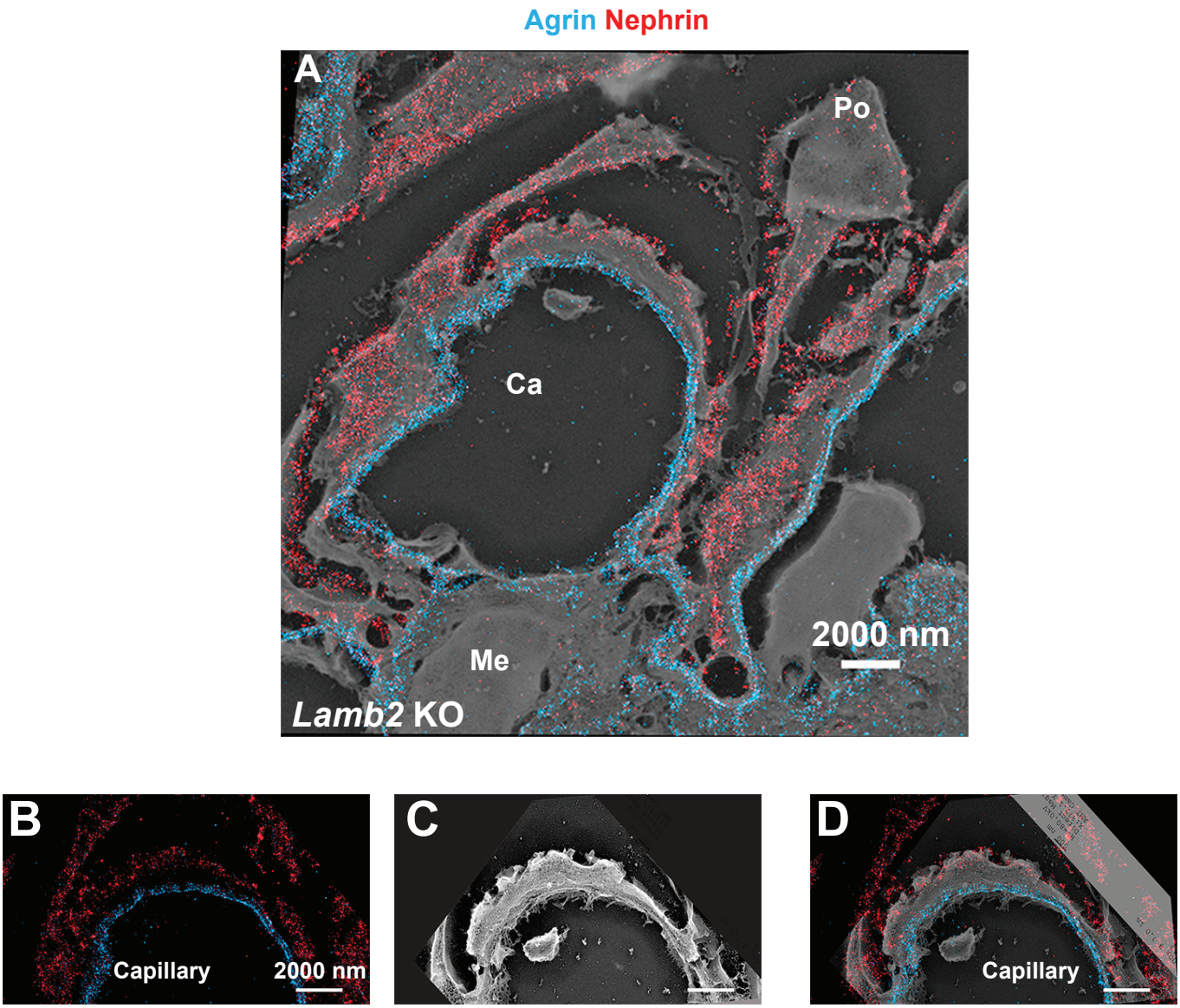
Supplemental Figure 1. STORM imaging of myosin IIB spatial distribution in healthy mouse glomerulus.
(A and B) Overview (A) and zoomed-in (B) STORM images of a WT mouse glomerulus stained for integrin β1 (blue) and myosin IIB (red) shows myosin IIB only in mesangial cells. Ca: capillary, Me: mesangial cells



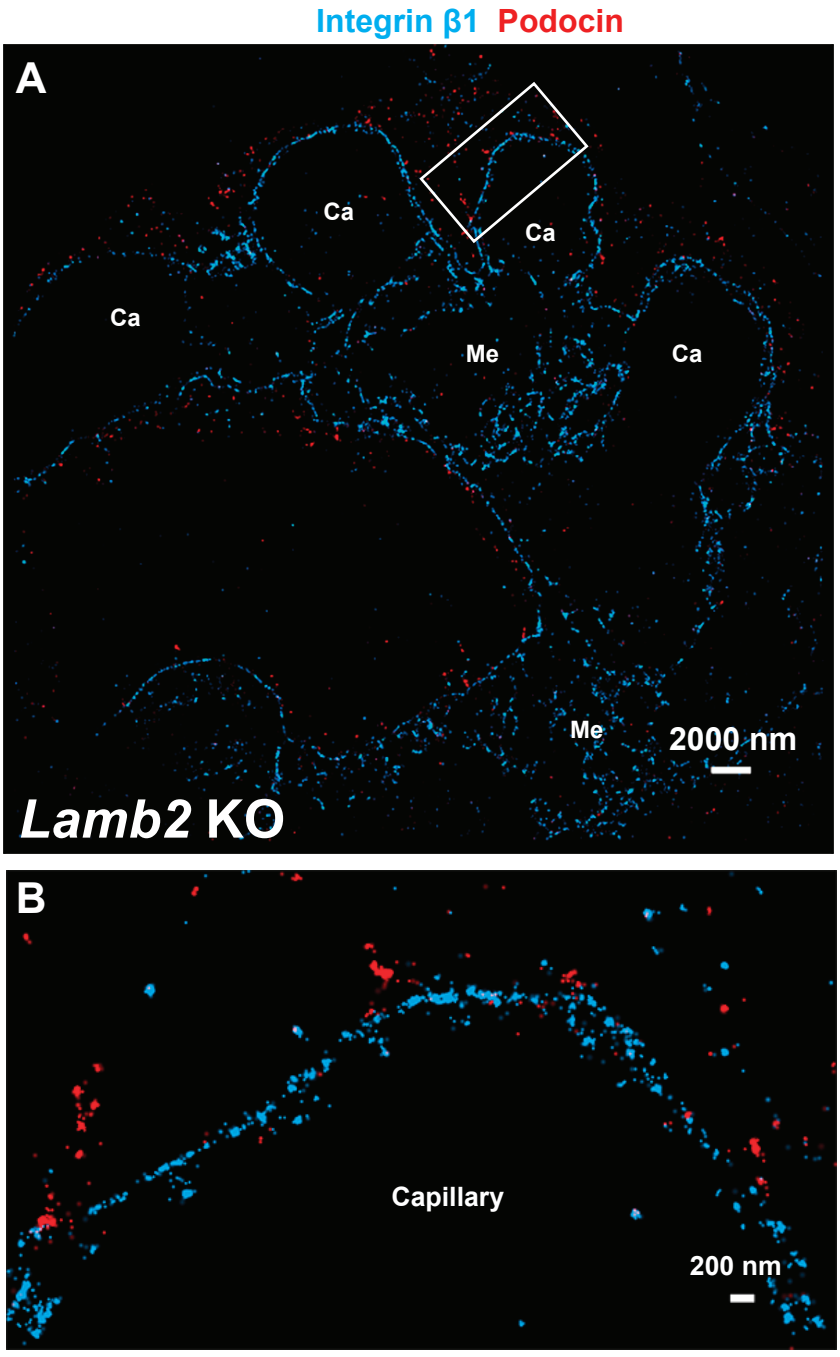
Supplemental Figure 2. Confocal sectioning of isolated glomeruli to visualize actin in podocytes.
(A and B) Confocal sections of a WT glomerulus stained with phalloidin (grey), anti-WT1 (light blue) and DAPI (dark blue) show a wide array of phalloidin-positive actin cables radiating from WT1-positive podocytes. (A', A'') and (B', B'') Confocal slices of the boxed areas in (A) and (B), respectively, demonstrate the position of WT1-positive cells on the surface of the isolated glomerulus, with radiating phalloidin-positive cables.



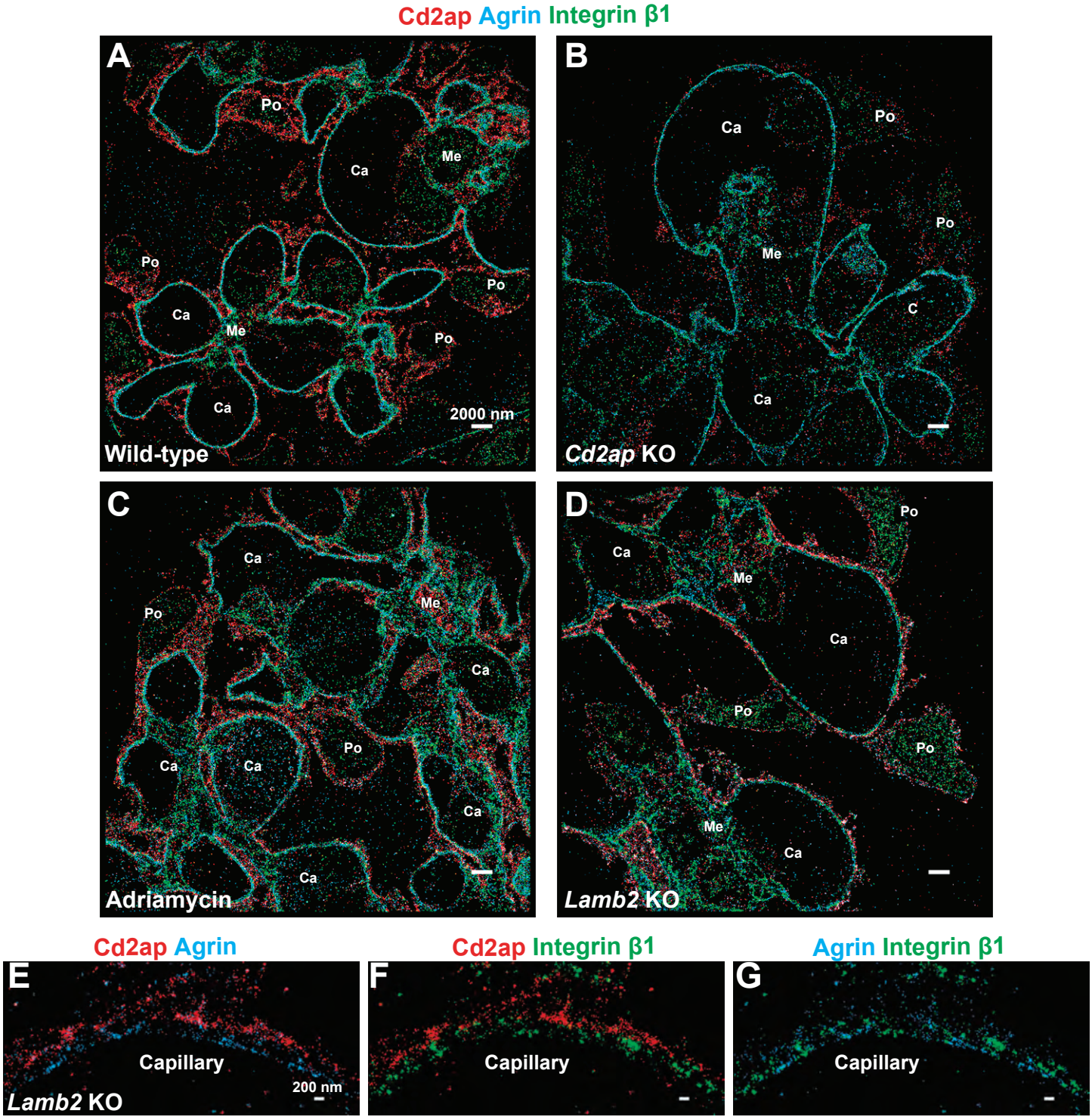
Supplemental Figure 3. Overview STORM images of nephrin localization in injury models. (A-C) STORM-EM correlation in WT (A), *Cd2ap* KO (B), and adriamycin-treated (C) glomerular sections. Images show areas with FPE (B and C) and the apical translocation of nephrin (red). Boxed areas are those chosen for high magnification images in Figure 3. (D) SDS-PAGE analysis of urine taken from 1 day and 9 days after Adriamycin injection shows massive proteinuria at 9 days.



Supplemental Figure 4. Nephrin moves apically with foot process effacement in *Lamb2* KO mice. (A) STORM-EM correlative overview images show nephrin (red) and agrin (blue) staining in a *Lamb2* KO glomerulus. Higher magnification STORM (B), EM (C) and STORM-EM correlative (D) images show an area of FPE and nephrin translocation away from the GBM towards the apical surface of the podocyte.



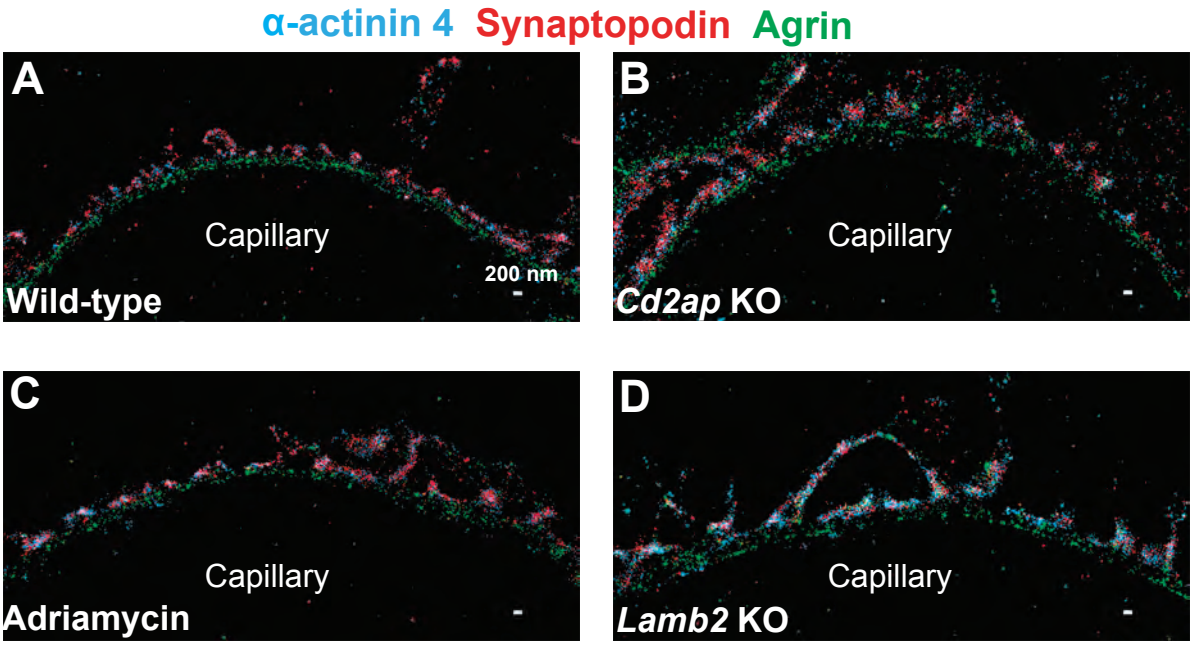
Supplemental Figure 5. Podocin moves apically with foot process effacement in *Lamb2* KO mice. STORM overview (A) and high magnification (B) images show podocin (red) and integrin β1 (blue) staining in a *Lamb2* KO glomerulus. Very little podocin staining is evident, and when present, most of it is far from the edge of the GBM (B).



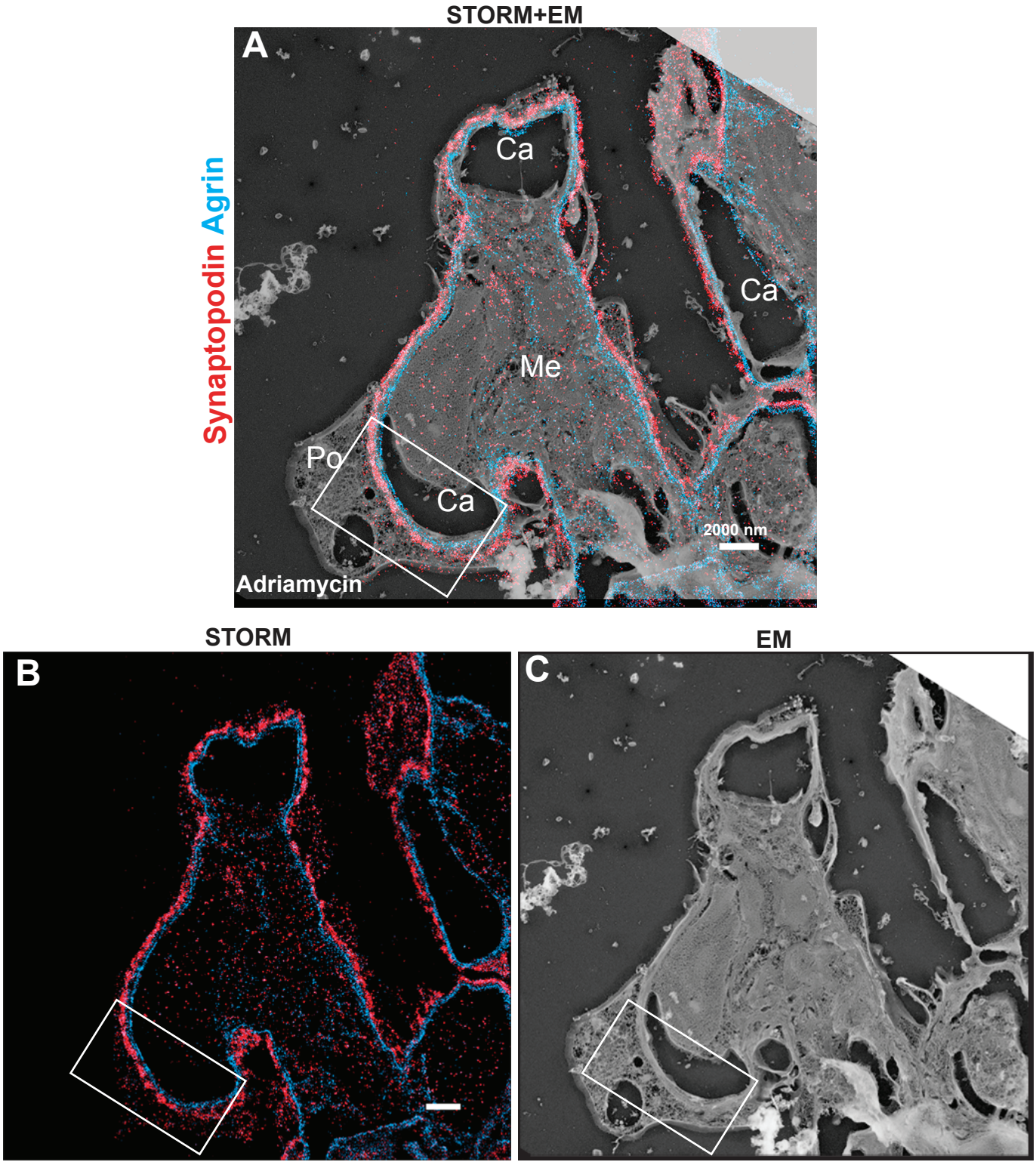
Supplemental Figure 6. Overview STORM images of CD2AP localization in WT, *Cd2ap* KO, adriamycin nephropathy, and *Lamb2* KO glomeruli.

(A-D) Overview images of WT (A), *Cd2ap* KO (B), adriamycin-injured (C), and *Lamb2* KO (D) glomeruli stained for CD2AP (red), agrin (blue), and integrin β1 (green). (E-G) High magnification STORM image of the *Lamb2* KO glomerulus in D shows CD2AP accumulation near the GBM, identified by co-staining with agrin (E) and integrin β1 (F). (E, G) Double-color images show disorganization of the subepithelial layer of agrin.

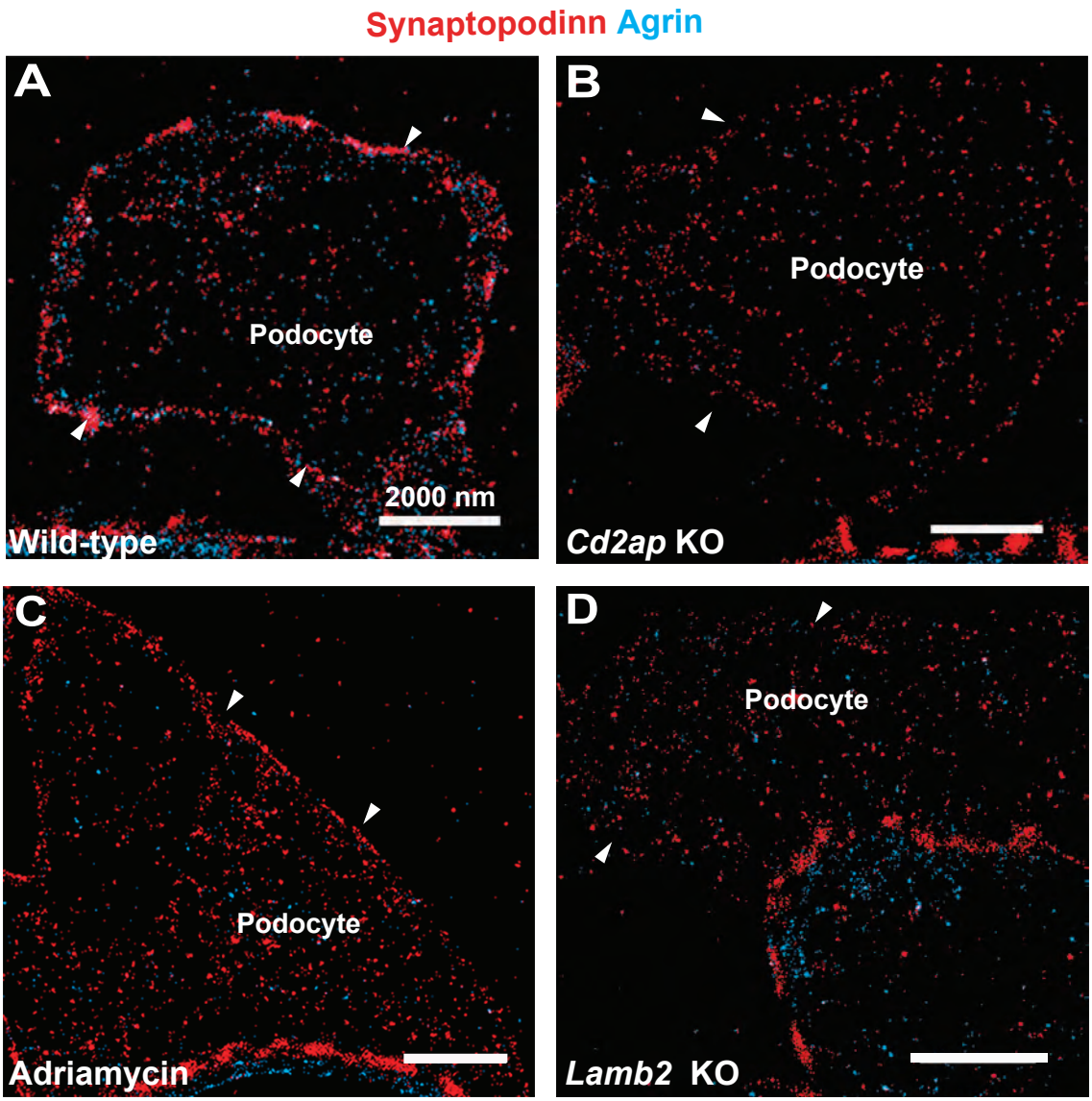
Supplemental Figure 7. Lack of major changes in agrin or integrin β1 localization in *Cd2ap* KO and Adriamycin nephropathy.
STORM images of WT (A), *Cd2ap* KO (B) and adriamycin-injured (C) glomerular capillary walls show agrin (blue) and integrin β1 (green) staining at the edges of the GBM.



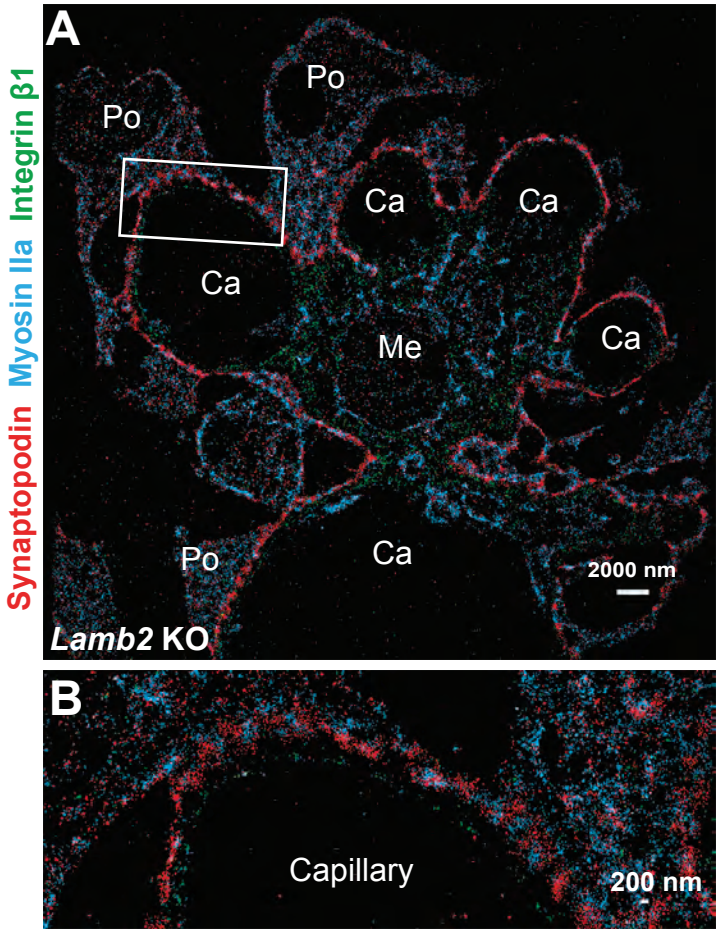
Supplemental Figure 8. Synaptopodin and α-actinin-4 overlap after podocyte injury. Triple color STORM images of WT (A), *Cd2ap* KO (B), adriamycin nephropathy (C) and *Lamb2* KO (D) capillary walls show co-localized synaptopodin (red) and α-actinin-4 (blue) clusters adjacent to the GBM stained for agrin (green).



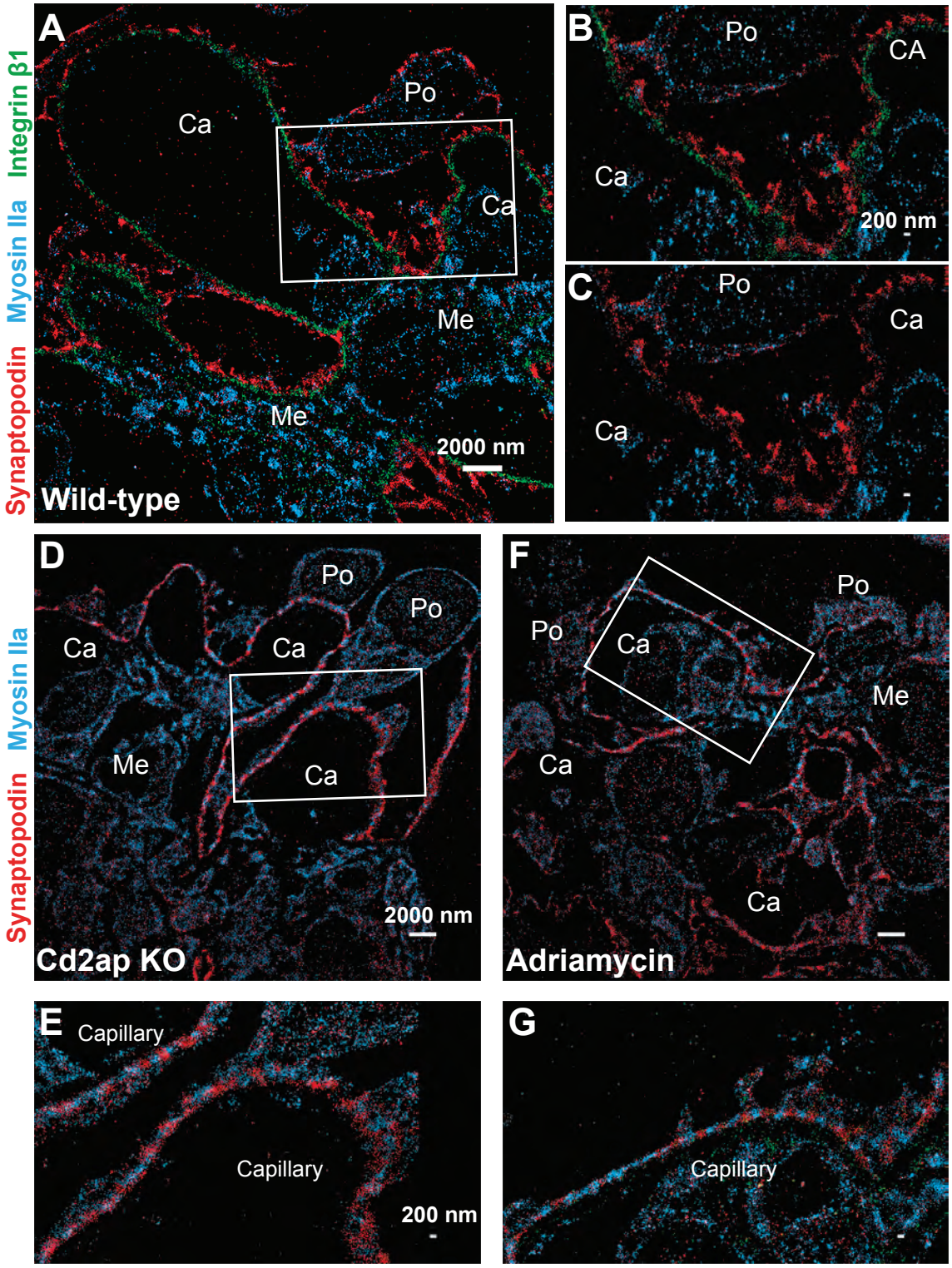
Supplemental Figure 9. Overview STORM-EM correlation of effaced areas in adriamycin nephropathy.
(A) STORM-EM overview image of the same glomerulus displayed in Fig. 4G and H show capillaries with widespread foot process effacement and large synaptopodin clusters. (B and C) Separate STORM (B) and EM (C) images of (A) are shown.



Supplemental Figure 10. Peripheral synaptopodin patches disappear after podocyte injury. Double color STORM images of WT (A), *Cd2ap* KO (B), adriamycin nephropathy (C) and *Lamb2* KO (D) show the disappearance of synaptopodin patches (red) from the edges of podocyte cell bodies after injury (B, C and D, arrowheads) when compared with WT control (A, arrowheads). The GBM is stained for agrin (blue).

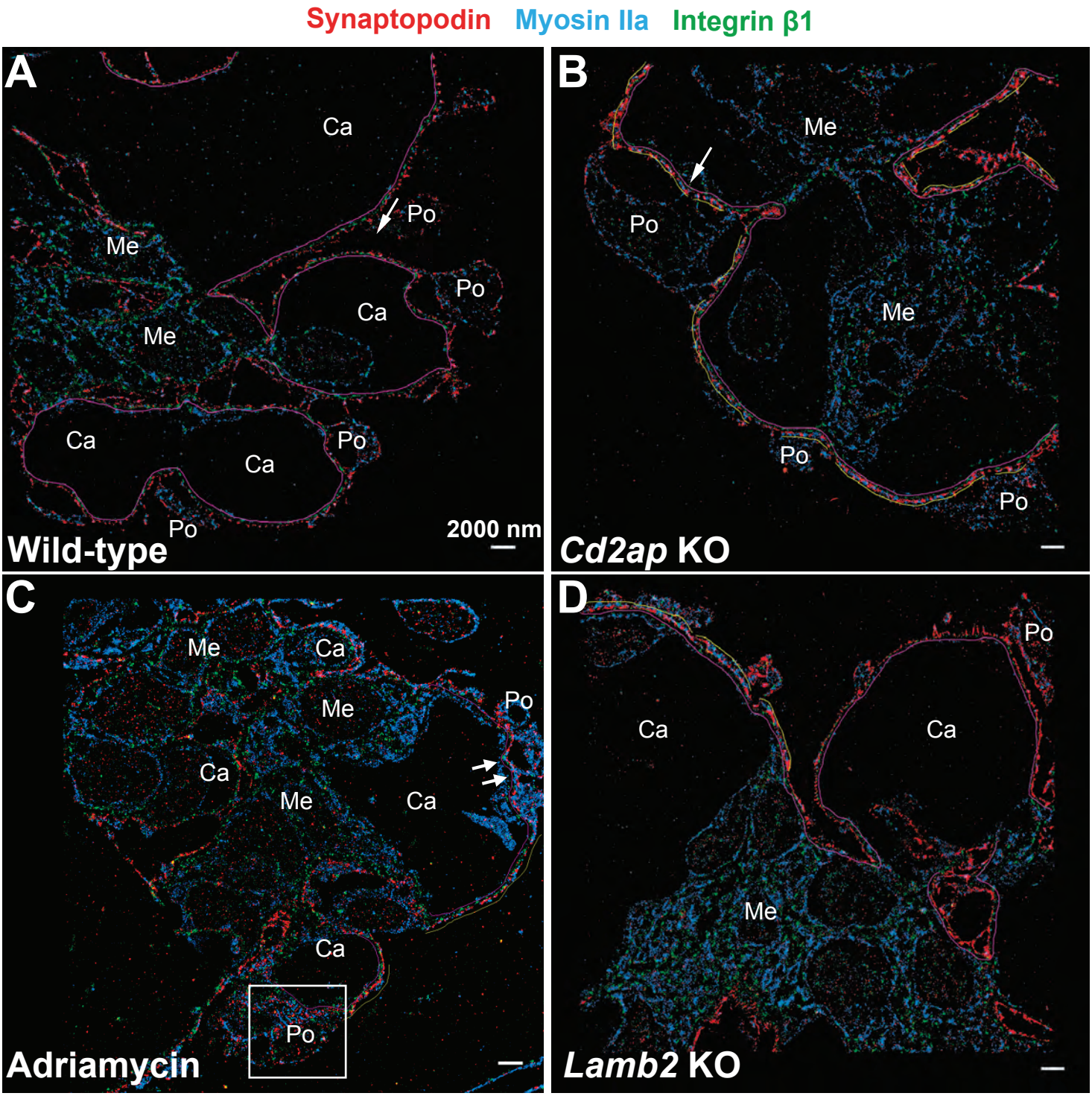


Supplemental Figure 11. The distribution of myosin IIA in the *Lamb2* KO glomerulus.
(A) Triple-color STORM overview image of a *Lamb2* KO glomerulus shows the distribution of myosin IIA. (B) High magnification STORM image of the boxed area in (A) shows myosin IIA clusters (blue) alternating with synaptopodin clusters (red) near the GBM edge identified by integrin β1 (green).

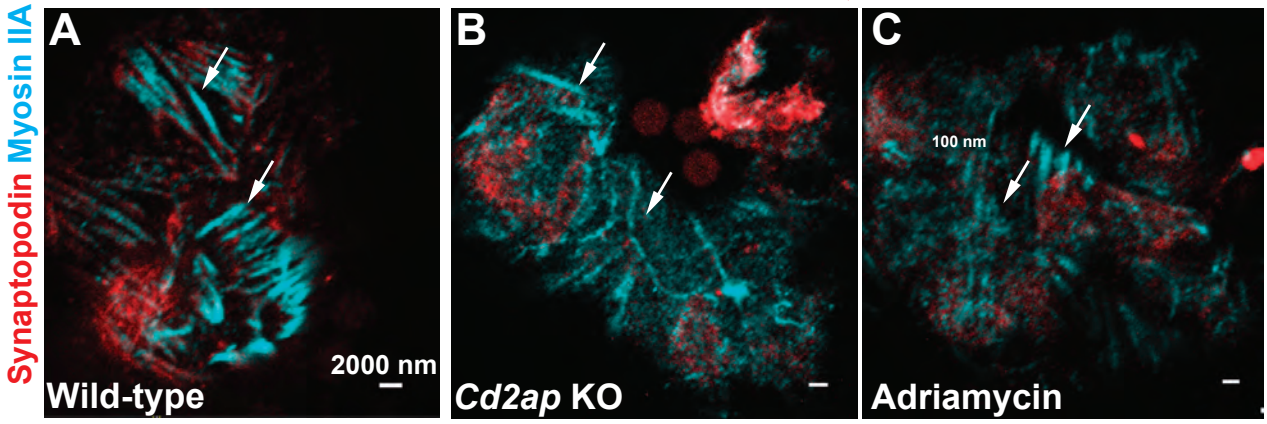


Supplemental Figure 12. Confirmation of the localization of myosin IIA in WT and injured podocytes.

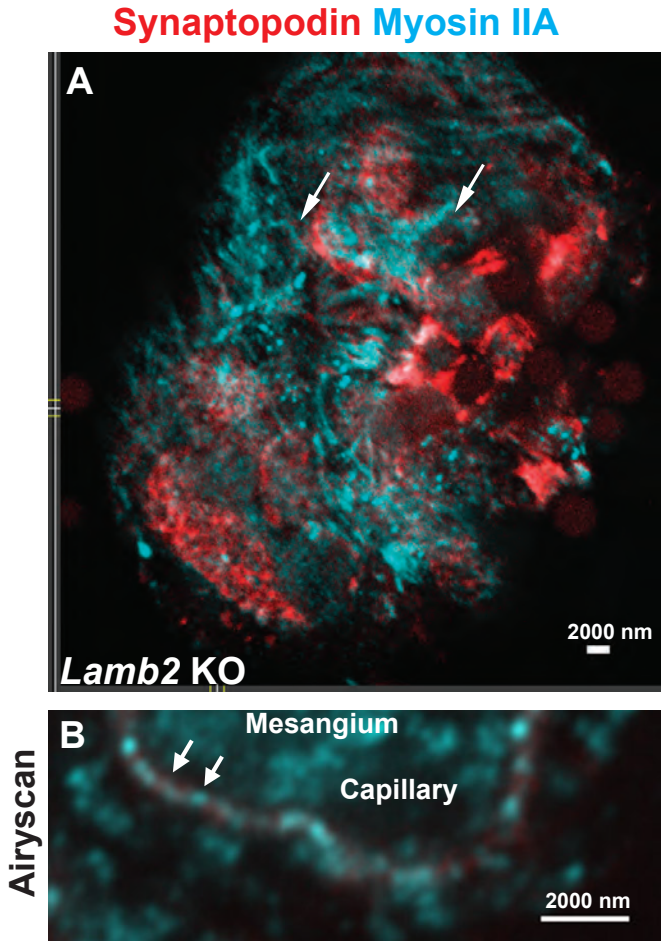
(A-C) Triple color STORM imaging of a WT glomerulus using an antibody to the N-terminus of myosin IIA (blue) demonstrates its presence in the podocyte cell body and in major processes but not in foot processes. (B, C) Triple-color and double-color higher magnification images of the boxed area in (A), respectively, shows the distribution of myosin IIA in podocytes. (D-G) Double-color STORM images of myosin IIA (blue) and synaptopodin (red) in injured glomeruli. *Cd2ap* KO (D and E) and adriamycin-induced injury (F and G) models show that synaptopodin and myosin IIA are present in alternating clusters near the GBM.



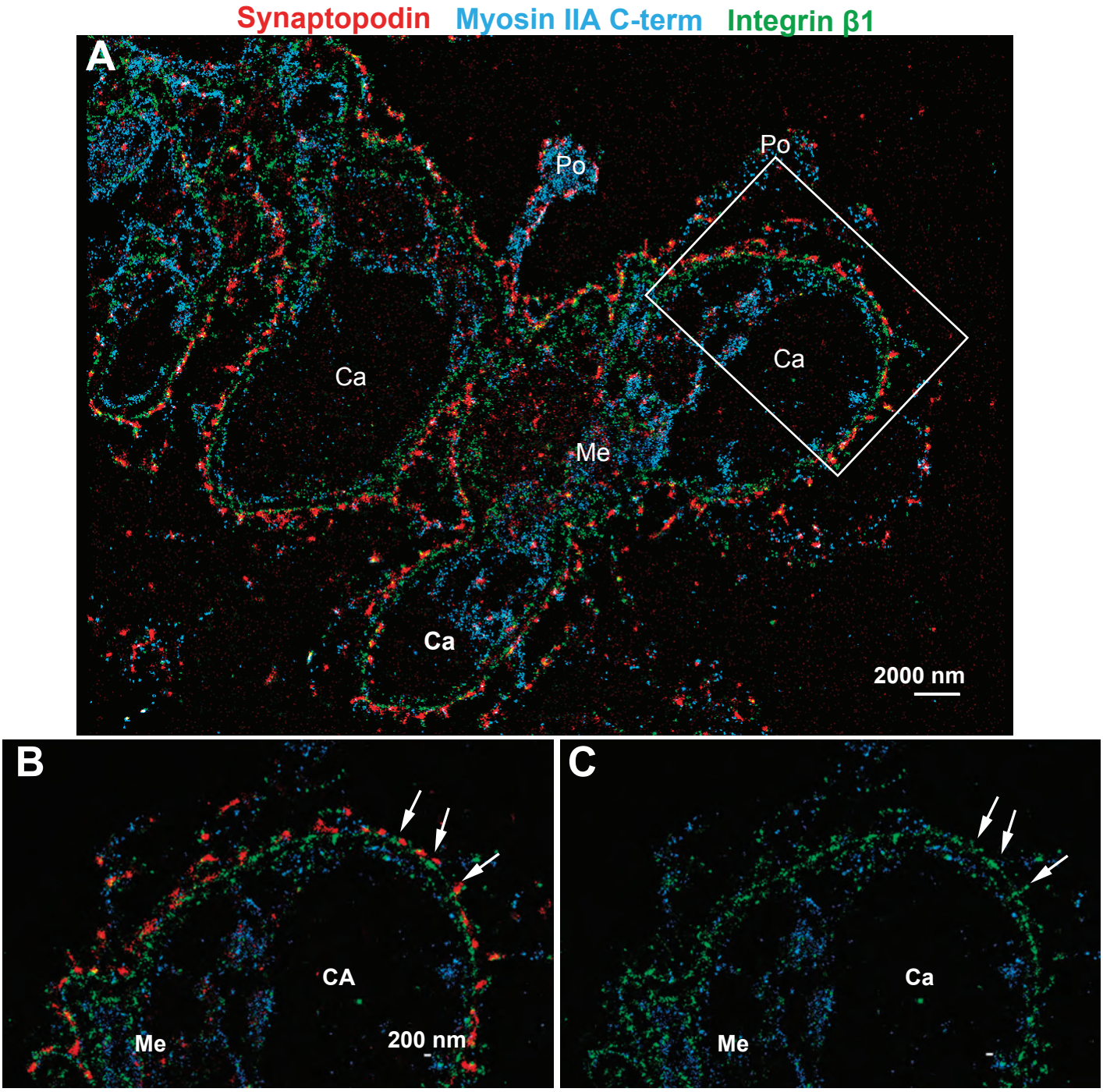
Supplemental Figure 13. Overview N-STORM imaging of the extent of sarcomere-like structures. Overview images of WT (A), *Cd2ap* KO (B), adriamycin nephropathy (C) and *Lamb2* KO (D) show the extent of the GBM (magenta lines) covered by sarcomere-like structures (yellow lines) identified by alternating clusters of synaptopodin and myosin IIA.



Supplemental Figure 14. Confocal images of actin-stabilized membrane-extracted isolated glomeruli.
(A-C) Confocal images of the glomerular surface show myosin IIA-positive actin cables in the WT (A), *Cd2ap* KO (B) and adriamycin-induced injury (C) models, indicating that myosin IIA-positive cables (arrows) are still present after injury.

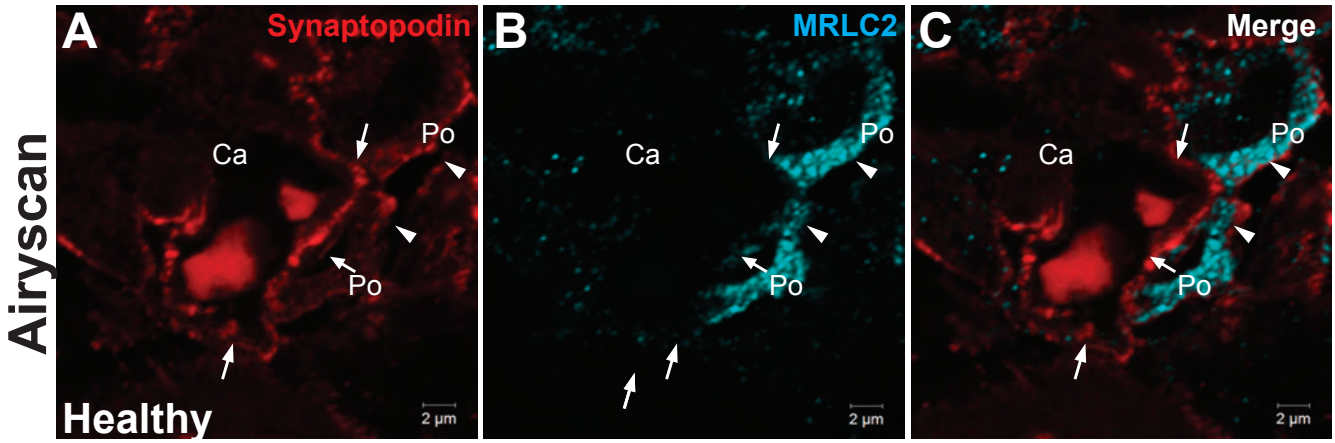


Supplemental Figure 15. Contractile actin cables in *Lamb2* KO podocytes.
(A) Confocal sectioning of a *Lamb2* KO glomerulus stained for myosin IIA (blue) and synaptopodin (red) shows the presence of a few contractile actin cables covering the glomerulus (arrows). (B) Airyscan confocal slice of a *Lamb2* KO capillary wall shows the presence of myosin IIA along the capillary wall labeled with synaptopodin in a periodic fashion (arrows).

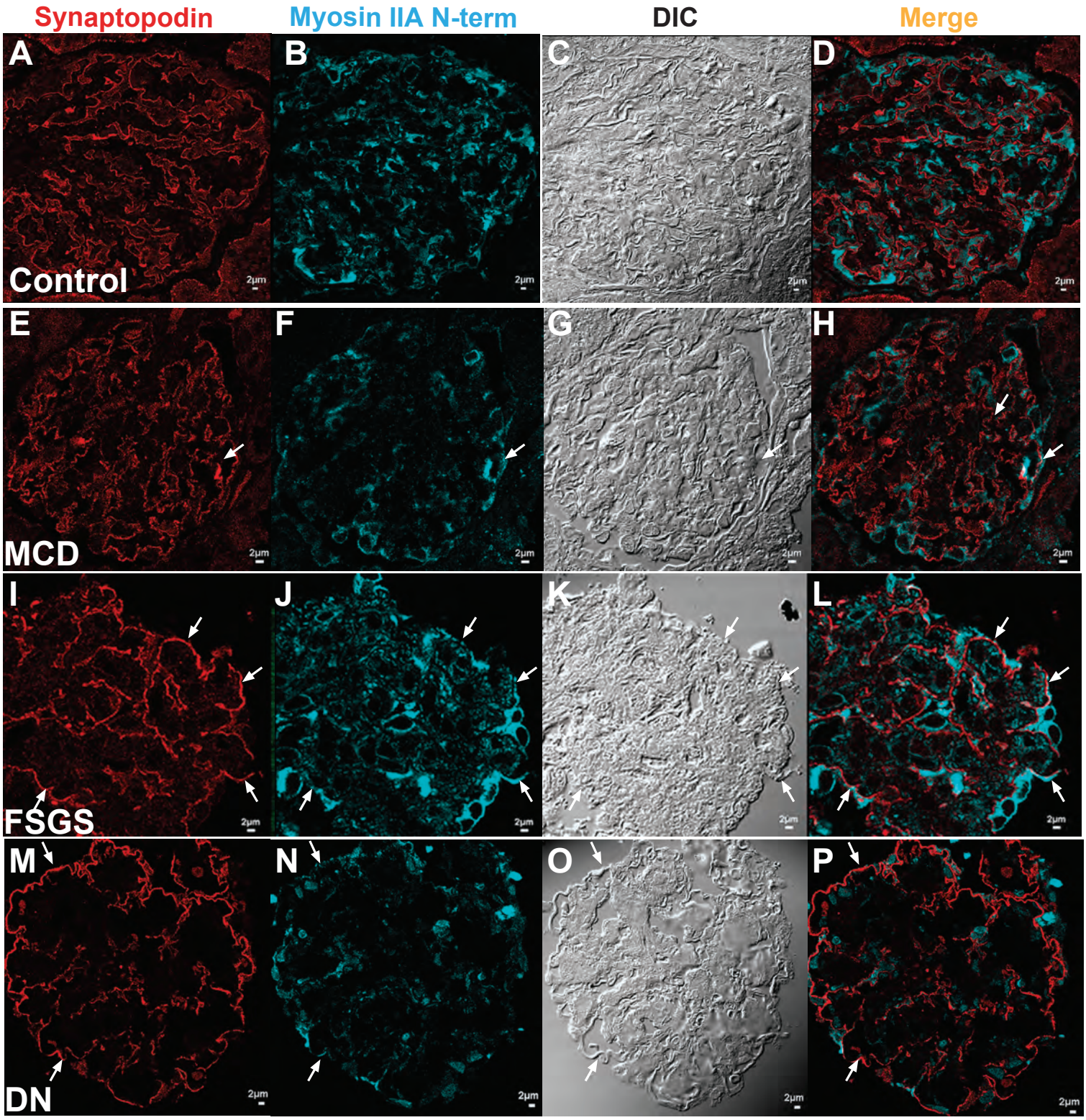


Supplemental Figure 16. Myosin IIA is present in the human podocyte cell body and major processes but not in foot processes.

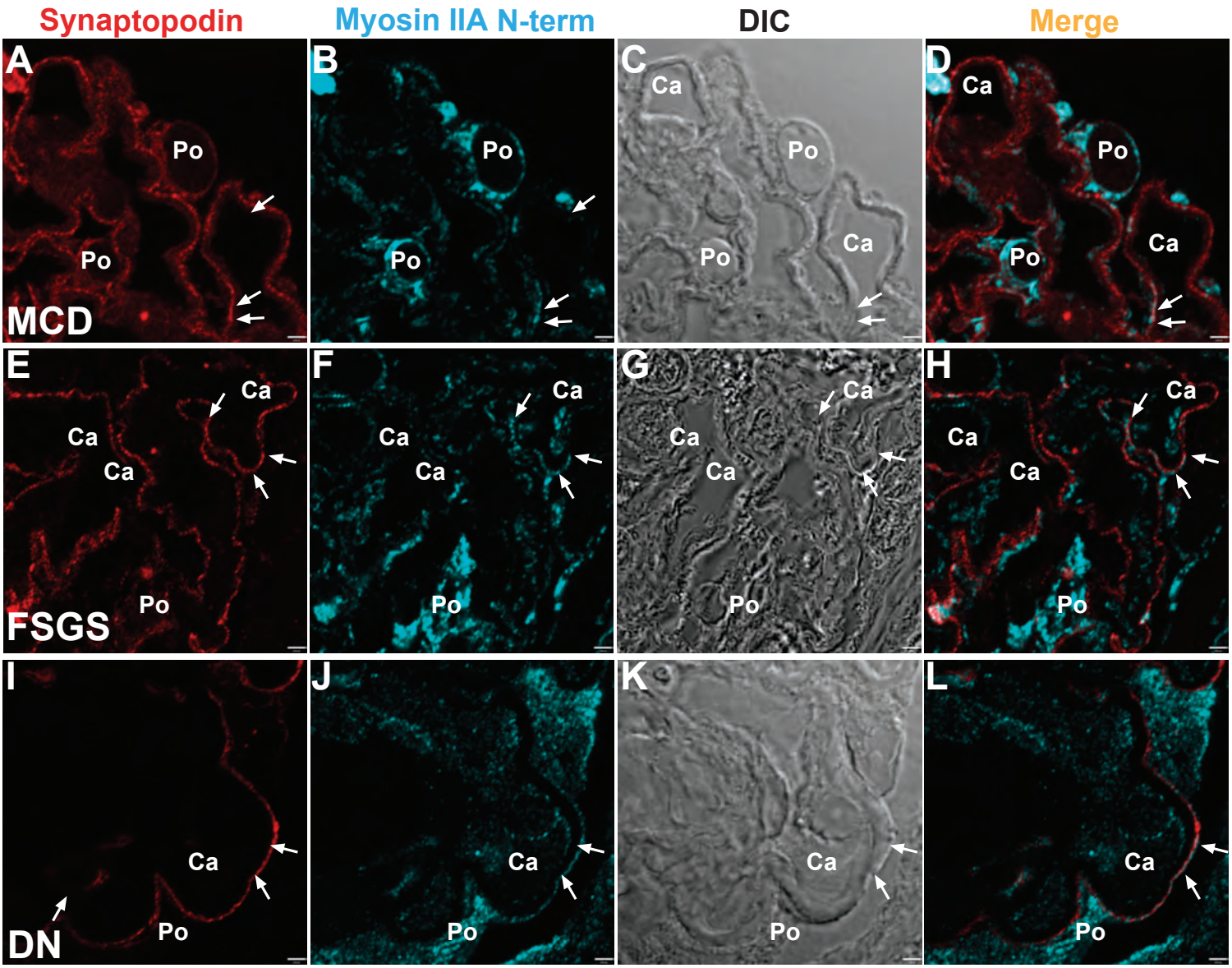
(A-C) Triple-color STORM image of a healthy human glomerulus using integrin β1 (green), synaptopodin (red) and another antibody against myosin IIA (blue) confirming that the podocyte foot processes are labeled by synaptopodin (arrows), while myosin IIA is present only in the podocyte cell body (Po) and the major processes.



Supplemental Figure 17. Myosin IIA regulatory light chain 2 (MRLC2) is present in the human podocyte cell body and major processes but not in foot processes.
(A-C) Airyscan imaging of a healthy human glomerulus stained for synaptopodin (red) and MRLC2 (blue) shows that MRLC2 is located in the cell body and the major processes (arrowheads) but not in the foot processes (arrows). Ca, capillary; Po, podocyte.



Supplemental Figure 18. Myosin IIA redistribution in human glomerular diseases.
Confocal images of human kidney biopsies from a control (A-D) and from patients with minimal change disease (E-H), focal segmental glomerulosclerosis (I-L), and diabetic nephropathy (M-P) labeled with synaptopodin (red) and myosin IIA (blue) show myosin IIA co-localization with synaptopodin at the edges of the capillary loops (arrows). Po: podocyte; Ca: capillary; Me: mesangial cell; RBCs: red blood cells; MCD: minimal change disease; FSGS: focal segmental glomerulosclerosis; DN: diabetic nephropathy.



Supplemental Figure 19. Myosin IIA as part of the sarcomere-like structures in human glomerular diseases.

Single channel Airyscan images of the capillary loop images in Fig. 7J-L labeled with synaptopodin (A, E and I), myosin IIA (B, F and J) and merged (D, H and L) show the intertwined synaptopodin (red) and myosin IIA (blue) staining at the edges of the capillary loops (arrows). DIC micrographs of the same region show the structures that were imaged (C, G and K). Note the thick GBM in the DN sample (K). Po, podocyte; Ca, capillary; Me, mesangial cell; RBCs, red blood cells; MCD, minimal change disease; FSGS, focal segmental glomerulosclerosis; DN, diabetic nephropathy.