# $LPA_3$-mediated lysophosphatidic acid signaling promotes postnatal heart regeneration in mice

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## Supplemental material:

**Supplemental Figure 1. Cardiomyocyte apoptosis in the $LPA_3$ KO and wild-type control mice.** (A) Quantification showing that no change in TUNEL signals was detected in the P4 and P7 hearts of the $LPA_3$ KO mice compared with the controls. (B) $LPA_3$ knockout did not influence the number of apoptotic cardiomyocytes after MI. n=5 mice in each group. Data are presented as the mean ± SEM; nonsignificant (N/S), $P > 0.05$.

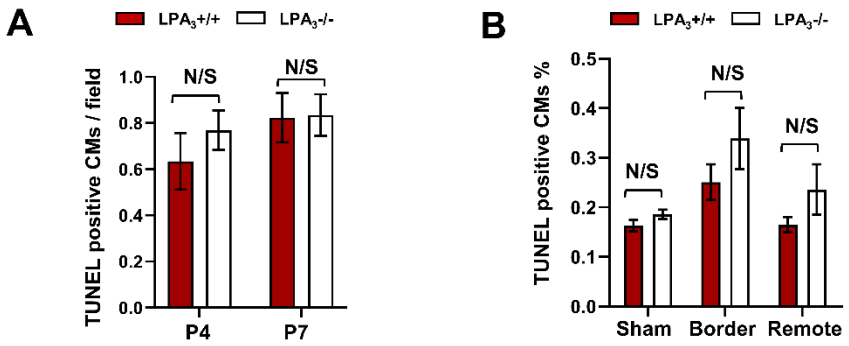
**Supplemental Figure 2. Validation of cardiac-specific $LPA_3$ expression induced by AAV9-cTNT-$LPA_3$-3Flag.** (A) AAV9:$LPA_3$ or AAV9:EGFP viruses were injected subcutaneously into the P1 mice. Seven days or 21 days later, the hearts were analyzed by western blots, qRT-PCR and immunofluorescence. (B) Western blot analysis confirmed the expression of 3Flag-$LPA_3$ in the AAV9:$LPA_3$ group but not in the AAV9:EGFP group. (C) qRT-PCR confirmed that the expression of $LPA_3$ in the AAV:$LPA_3$ group increased 3-fold at P7 and 15-fold at P28 compared with that of

AAV9:EGFP. (D) AAV9:$LPA_3$ expressed Flag-tagged $LPA_3$ in the membrane of cardiomyocytes, scale bar =200 μm. Data are the mean ± SEM; AAV-EGFP, n = 3; AAV-$LPA_3$, n = 3; "n" indicates the number of mice.

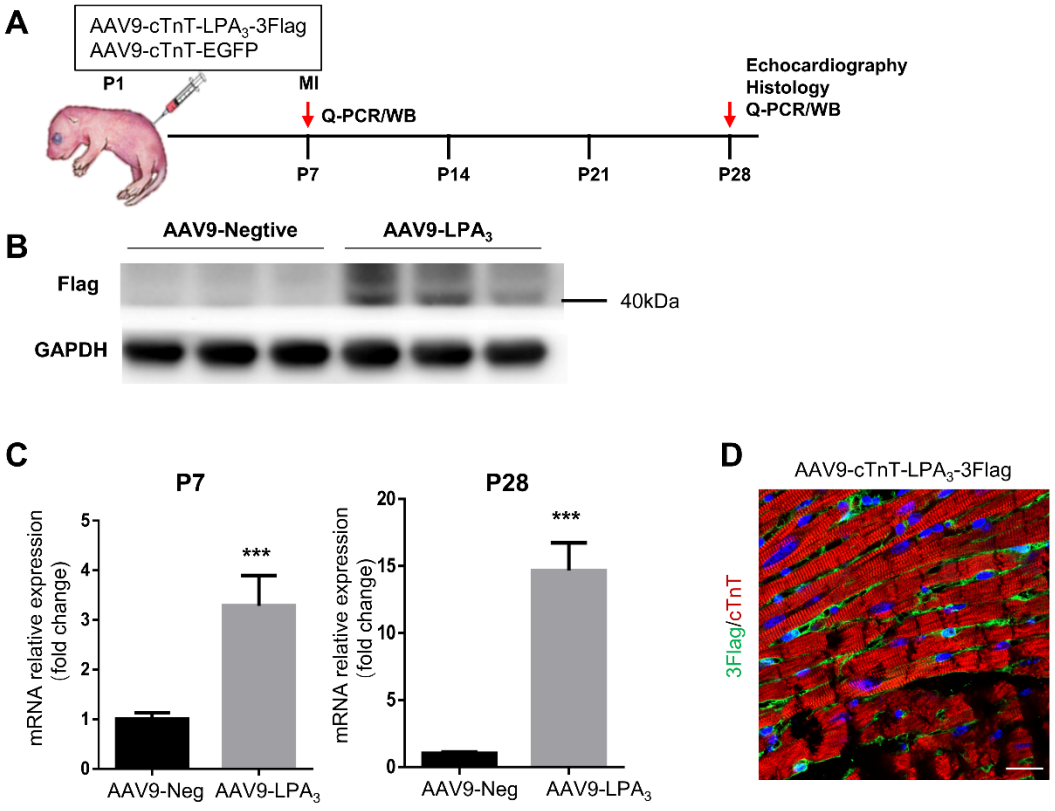
**Supplemental Figure 3. AAV9:$LPA_3$ has no effect on the healthy heart.** (A). pH3 and Ki67 immunofluorescence of cardiomyocytes in the healthy hearts of the AAV9:$LPA_3$ and control adult mice at P8. Scale bar =50 μm. (B) Representative sections of hematoxylin-eosin staining and the HW/BW ratio at P8 and P7 of AAV9. Data are presented as the mean ± SEM; nonsignificant (N/S), $P > 0.05$; n=5 mice per group.

**Supplemental Figure 4. Validation of the efficiency of siRNA.** (A) $LPA_3$ was knocked down by siRNA and verified by qRT-PCR. (B) YAP was knocked down by si-YAP and verified by Western blot. n=3 samples of each group; $^{**}P<0.01$; $^{***}P<0.001$.

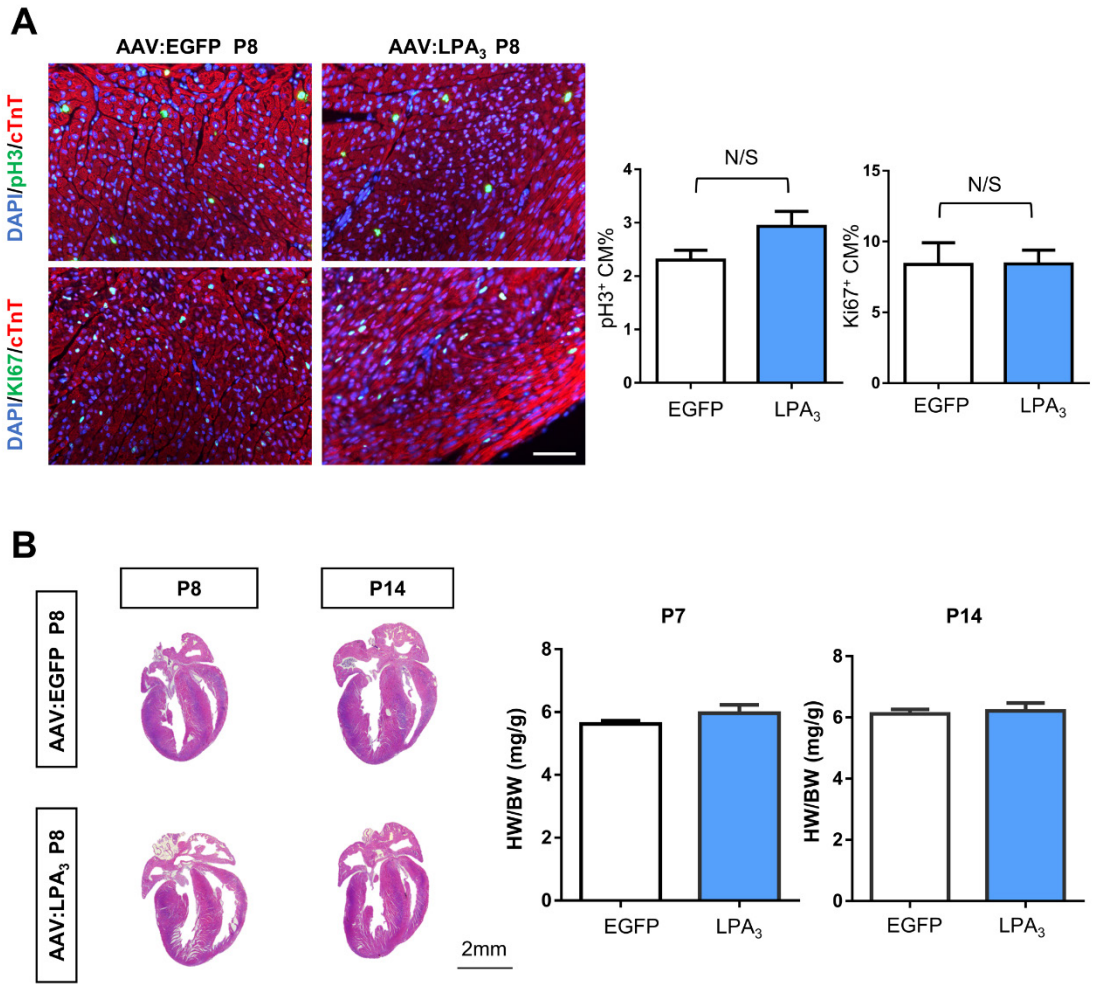
**Supplemental figure 1**



## Supplemental figure 2



**Supplemental figure 3**



**Supplemental figure 4**