Figure S1

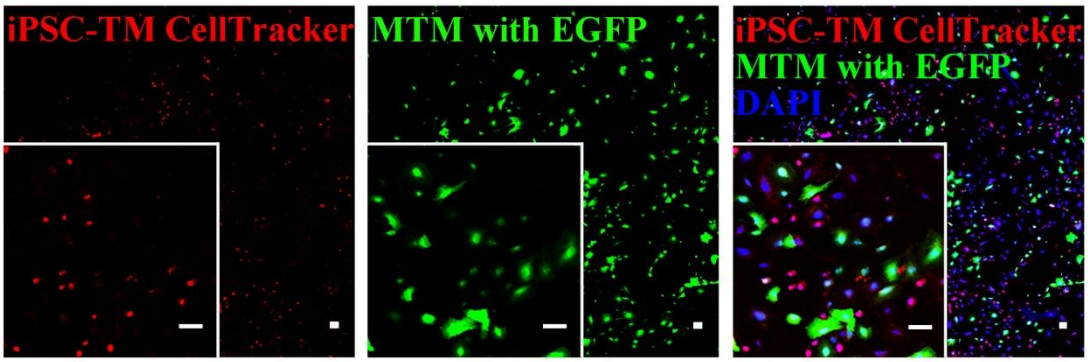

Figure S1. CellTracker transfer analysis. 40× magnification images of iPSC-TM pre-stained with CellTracker Red CMTPX and MTM infected with lentivirus carrying CMV-EGFP after co-culture for 48 hours. The region in the white frame shows a magnified view. CellTracker Red CMTPX is be detected in MTM with green fluorescence. Typical results from n=3 technical repeats are shown. Scale bar =100 μm.

Figure S2

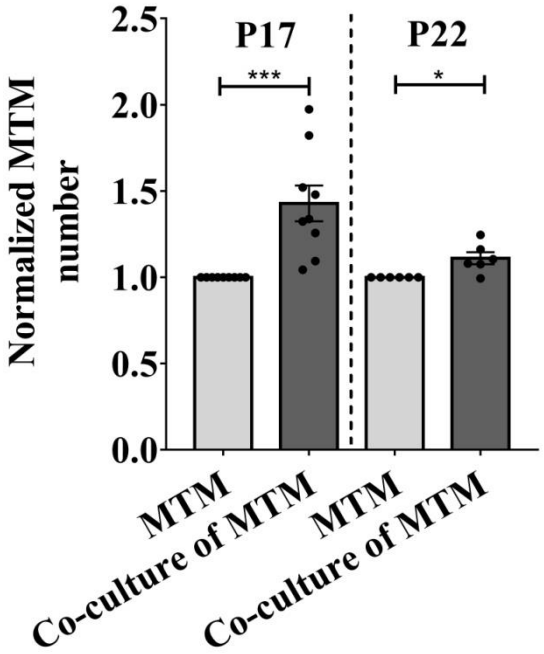

Figure S2. iPSC-TM stimulation of MTM proliferation. iPSC-TM stimulate proliferation of MTM at both passage 17 (n=9) and 22 (n=6), although the efficiency is lower at higher passage numbers. *$P < 0.05$. ***$P < 0.001$ by two-tailed t-test. Data shown are means ± SEM.

Figure S3

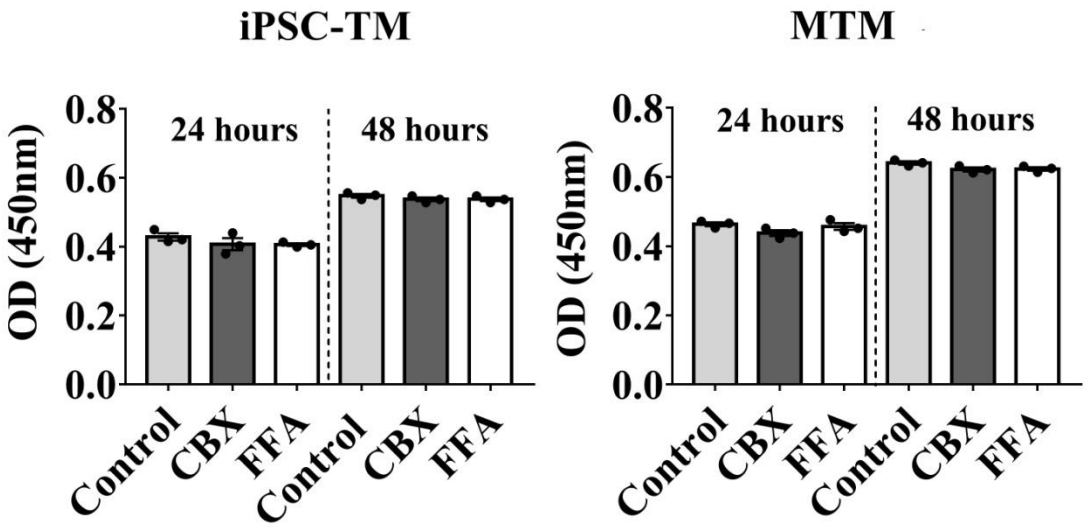

Figure S3. Cell viability of iPSC-TM and MTM treated with CBX or FFA. OD (450nm) readings of CCK8 tests revealed that CBX (50 μM) or FFA (50 μM) treatment has no toxic effect on either iPSC-TM or MTM after exposure for 24 and 48 hours. Typical results from n=3 technical repeats are shown. Statistical analysis was performed using one-way ANOVA. Data shown represent means ± SEM.

Figure S4

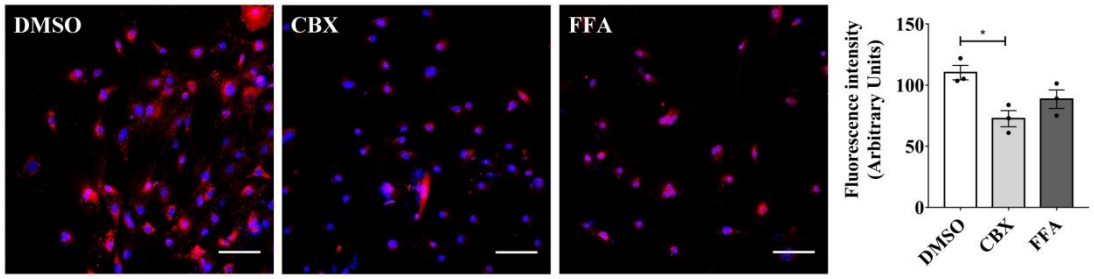

Figure S4. Treatment with gap junction inhibitors down-regulates Cx43 expression in iPSC-TM. iPSC-TM differentiated for 16 days are labeled with Cx43 antibody (Red). Typical results from n=3 technical repeats are shown. (Right) Quantification of Cx43 expression after CBX treatment (24 hours: 72.6 ± 6.6 vs. 110.3 ± 5.9, p=0.01, n=3) or FFA treatment (24 hours: 88.5 ± 7.6 vs. 110.3 ± 5.9, p=0.09, n=3). *$P < 0.05$ by two-tailed t-test. Scale bar = 100 μm. Data shown represent means ± SEM.

Figure S5

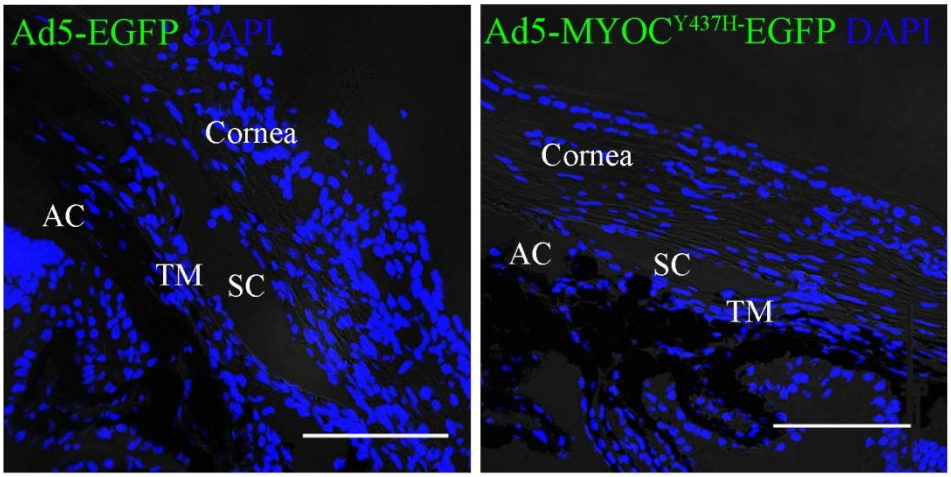

Figure S5. Immunohistochemical detection of EGFP (green) in the anterior segment of mice having received injections of Ad5-EGFP or Ad5-MYOC^{Y437H}-EGFP. EGFP fluorescence was diminished two months after Ad5 injection, similar to the previously reported findings [62]. Scale bar = 100 μm.

Figure S6

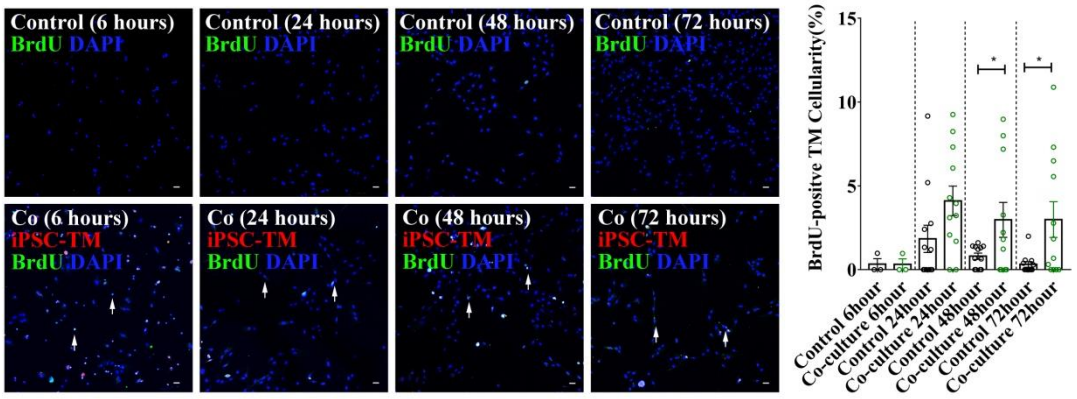

Figure S6. BrdU incorporation analysis in the co-culture system. (Left) 40× magnification images of iPSC-TM visualized by IHC staining with dsRed antibody (Red) and human TM cells (no fluorescence) in the co-culture system. BrdU is incorporated into dividing cells and appears green (white arrows). (Right) Quantification of BrdU-positive HTM cells after co-culture with mouse iPSC-TM for 6 (n=3), 24 (n=12), 48 (n=11-12), and 72 hours (n=12). $*P < 0.05$ by two-tailed t-test. Scale bar = 100 μm. Data shown are means ± SEM. Typical results from n=3-12 technical repeats are shown.

Figure S7

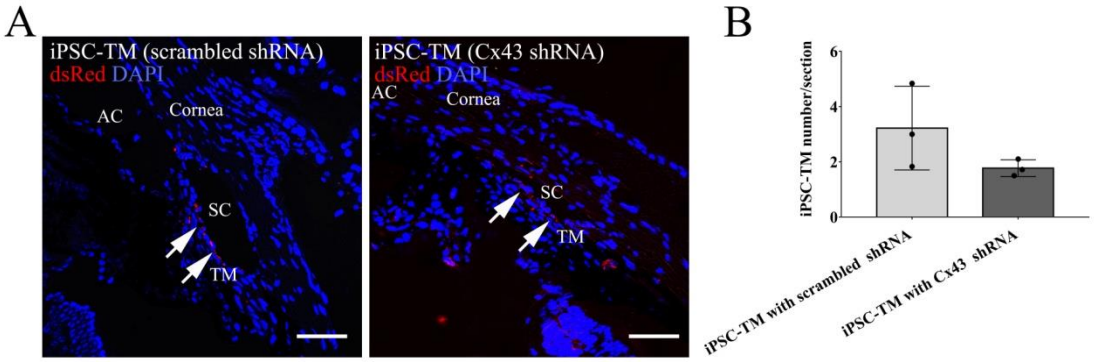

Figure S7. iPSC-TM survival in the TM after transplantation. (A) Detection of iPSC-TM (red) expressing Cx43 shRNA or scrambled shRNA one day after transplantation into 3-mon-old C57BL/6. (B) Quantification of iPSC-TM frequency in the TM one day after transplantation. scrambled shRNA expressing iPSC-TM cells integrate into the TM slightly better than those expressing Cx43shRNA (3.2 ± 0.9 cells/section, n=3 vs. 1.8 ± 0.2 cells/section, n=3) but statistical significance is not reached (p=0.2). Statistically analysis was performed by two-tailed t-test. Data shown represent means ± SEM. Scale bar= 100 μm.