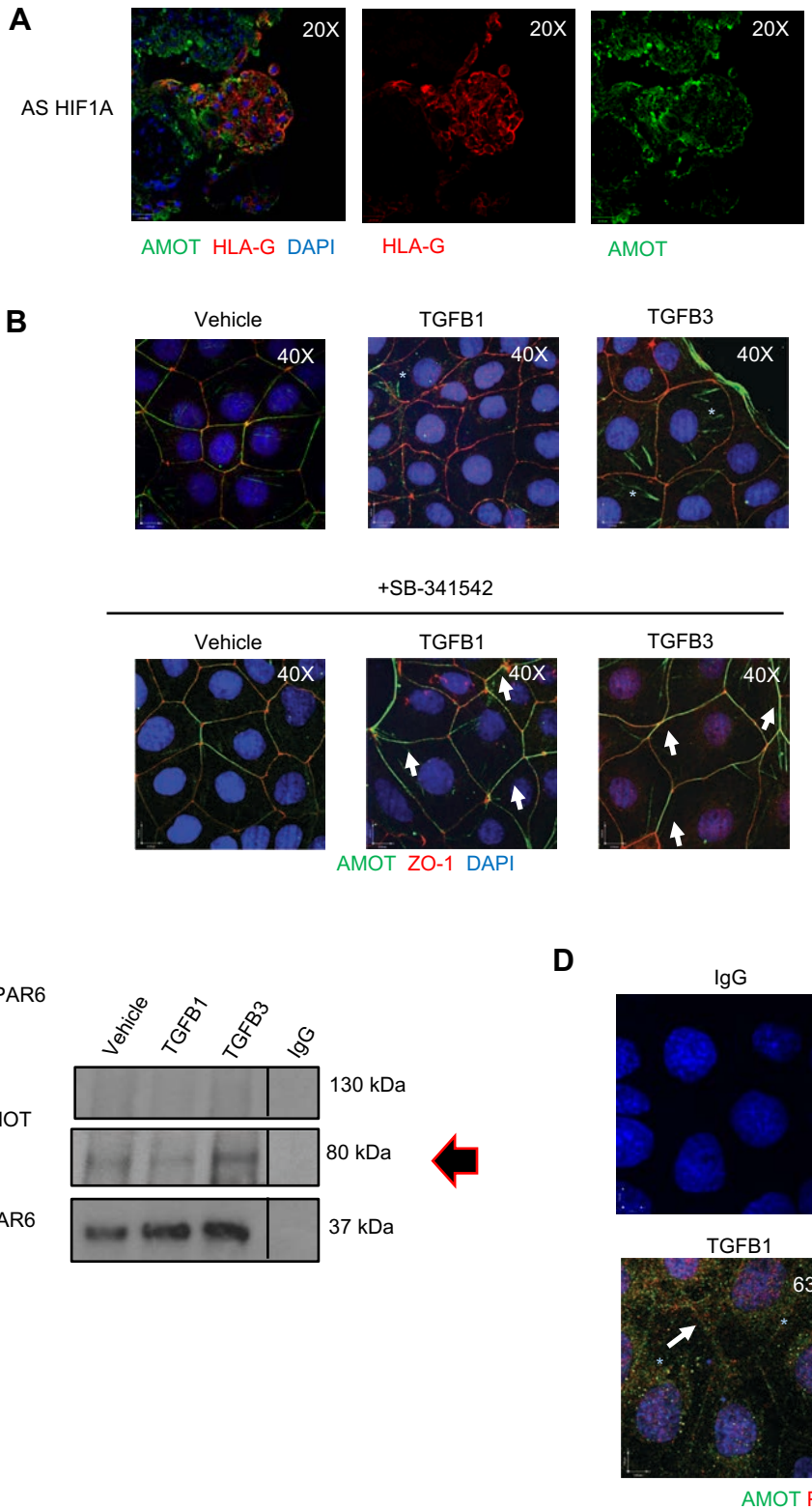
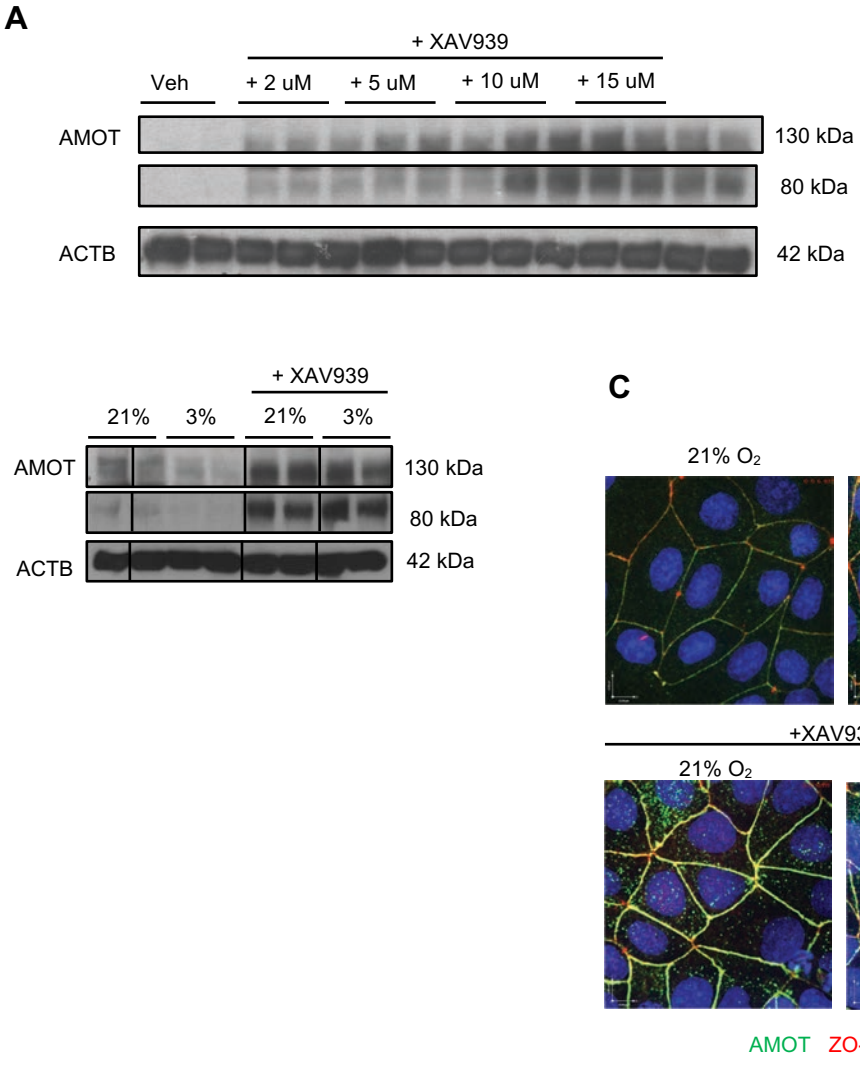
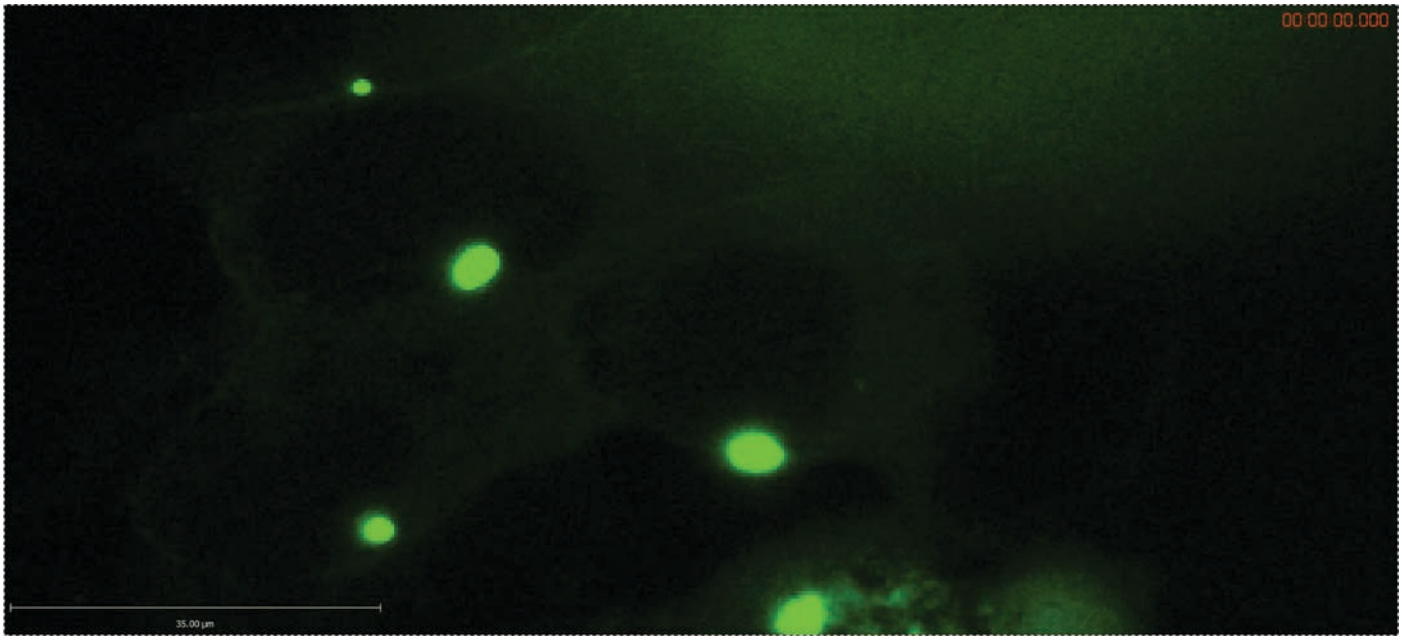


Supplementary Figure 1: TGFB mediated redistribution of AMOT occurs via Smad-dependent pathway

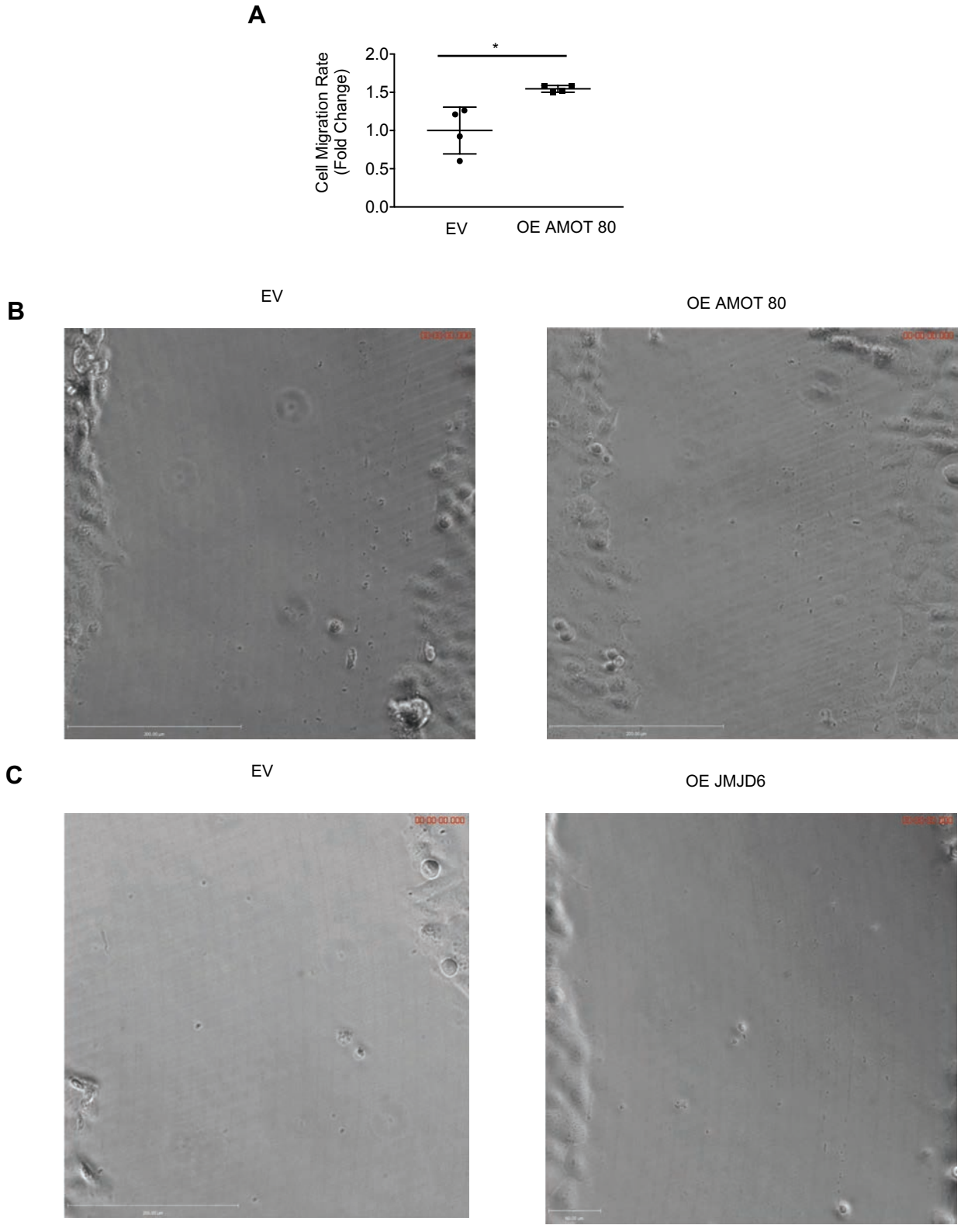
(A) Representative IF images of AMOT (green) and HLA-G (red) in villous explants of 5-8 weeks gestation treated with 10 uM anti-sense oligonucleotides to HIF1A. Explants were cultured in 3% oxygen (*n*=3 independent experiments). Original magnification, x20. **(B)** Representative IF images of AMOT (green) and tight junction protein ZO-1 (red) following treatment with vehicle, or 10 ng/mL TGFB1 or TGFB3 ligand for 24 h in the absence and presence of 5 uM ALK5 receptor inhibitor SB-431542. Nuclei were stained with DAPI (blue). Original magnification, x40. White arrows indicate AMOT retention at the tight junction (*n*=3). **(C)** IP of PAR6 followed by Western blot of AMOT in JEG3 cells after treatment with 10 ng/mL TGFB1/3 for 24 h (*n*=3 separate experiments carried out in duplicate). Lanes were run on same gel but were non-contiguous. **(D)** Representative IF images of AMOT (green) and PAR6 (red) following treatment with 10 ng/mL TGFB1 and TGFB3 ligand for 24 h. Anti-goat IgG was used as control and nuclei were stained with DAPI (blue). Original magnification, x40. White arrows indicate AMOT co-localization with PAR6 at tight junction (TJ), and asterisks indicate AMOT co-localization with PAR6 in the cytoplasm (*n*=3).



Supplementary Figure 2: Tankyrase inhibitor XAV939 restores low oxygen-mediated negative regulation of AMOT protein levels. (A) Representative WB of AMOT and ACTB in HTR-8/SVneo cells following treatment with DMSO vehicle or 2, 5, 10 or 15 uM of tankyrase inhibitor XAV939 for 24 hours (n=2). **(B)** Representative WB of AMOT and ACTB in HTR-8/SVneo cells cultured in 21% O_2 or 3% O_2 in the presence or absence of 10 uM of tankyrase inhibitor XAV939 for 24 hours (n=2). **(C)** Representative Immunofluorescence (IF) images of AMOT (green) and ZO-1 (red) in JEG3 cells cultured in 21% O_2 or 3% O_2 in the prescence or absence of 10 uM tankyrase inhibitor XAV939 for 24 hours (n=2). Original magnification, x40.



Supplementary Figure 3: AMOT dynamically redistributes during trophoblast cell migration
Representative videos depicting cell migration in JEG3 cells overexpressing AMOT 80 YFP construct. Original magnification, x20. AMOT distribution is observed to dynamically change from tight junction (TJ) (indicated by white arrows), to punctate structures, in the cytoplasm (indicated by blue asterisks) during cell migration (*n*=3).



Supplementary Figure 4: AMOT and JMJD6 promote cell migration of HTR-8/SVneo cells
(A) Linear migration rate for HTR-8/SVneo cells overexpressing AMOT 80 expressed as a fold-change relative to empty vector (EV) control. Statistical significance was assessed as **P <0.05 using non-parametric Mann Whitney U test (*n*=4). **(B)** Representative videos depicting cell migration during wound healing in HTR-8/SVneo cells overexpressing AMOT 80 and **(C)** JMJD6. Original magnification, x20.