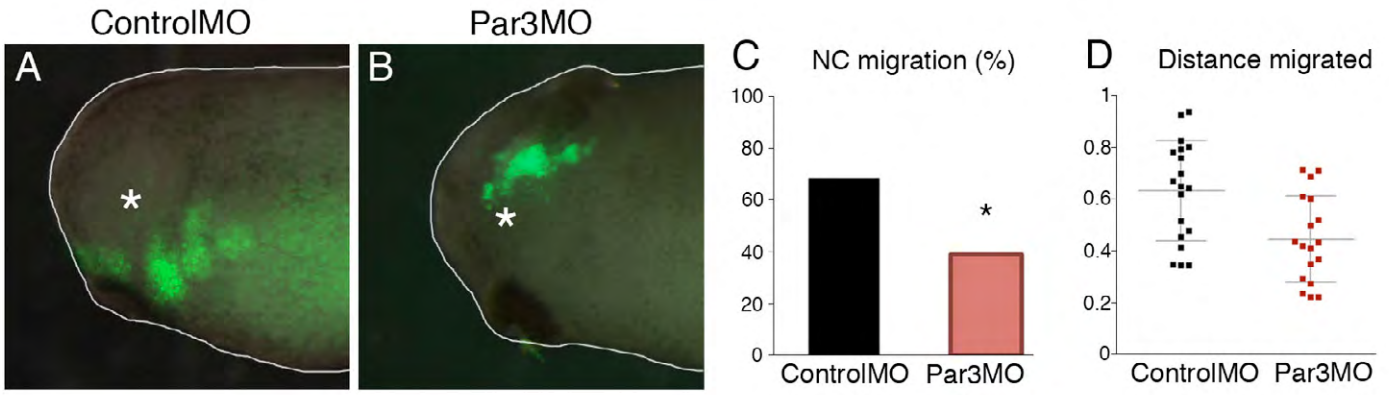


Fig. S1. Par3 is required in neural crest cells. (A-B) Grafts of fluorescently labelled NC cells into control host embryos. NC were injected with controlMO (A) or Par3MO (B). (C) Percentage of embryos with migratory grafted NC cells. Par3MO, n=18; ControlMO, n=19; $p<0.05$. (D) Average distance of NC migration for each grafted embryo. ControlMO, n=19; Par3MO, n=18; $p=0.0035$, error bars show mean and s.d.

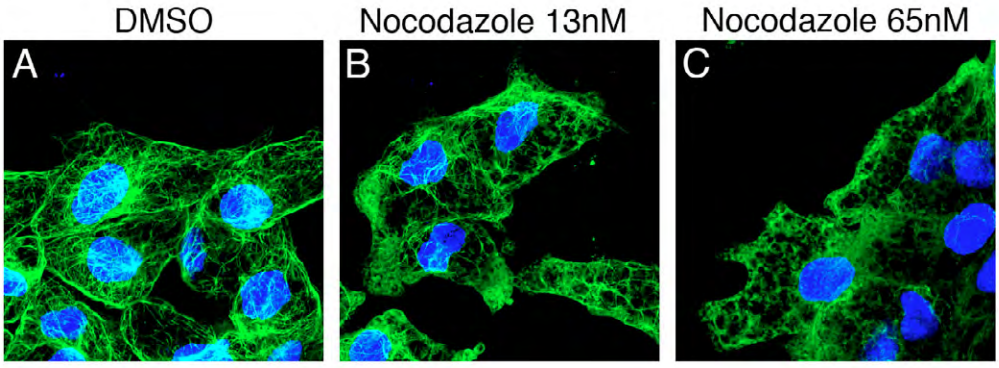


Fig. S2. Effect of nocodazole on microtubule array in NC cells. (A-C) Immunostaining against αtubulin shows the microtubule array in control NC cells (A) or NC cells treated with 13nM (B) or 65nM (C) of nocodazole. Note that in cells treated with the nocodazole concentration used to rescue Par3MO (13nM, panel B) microtubules are not completely depolymerized.

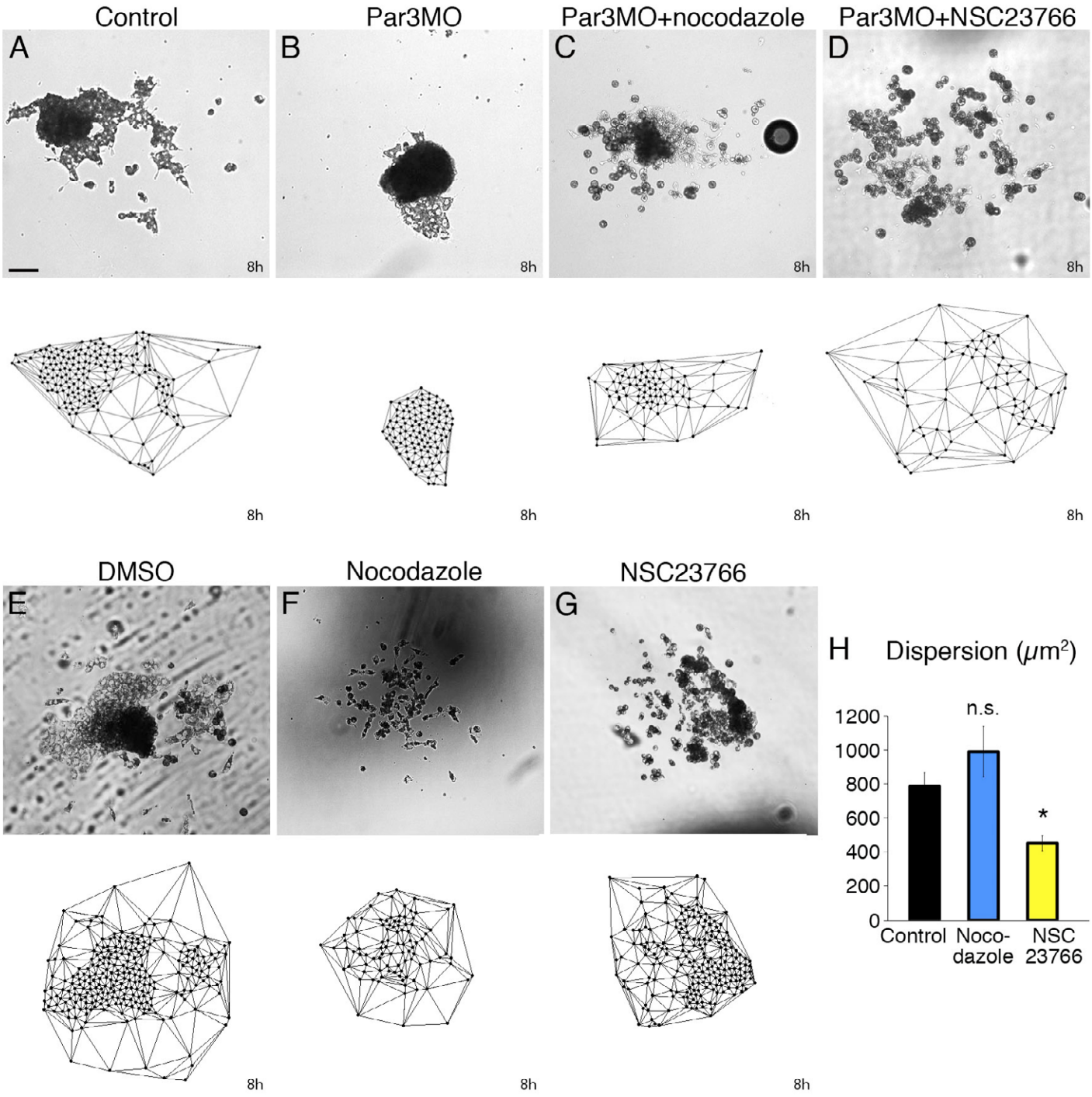


Fig. S3. Nocodazole and NSC23766 can restore normal dispersion in Par3MO NC cells. (A-D) Dispersion assay in control NC cells (A), Par3MO NC cells (B), Par3MO NC cells treated with nocodazole (C) and Par3MO NC cells treated with Rac inhibitor NSC23766 (D). (E-G) Dispersion assay in control NC cells (E), control NC cells treated with nocodazole (F) and control NC cells treated with Rac inhibitor NSC23766 (G). Note that nocodazole alone does not have an effect on NC dispersion. (H) Quantification of NC dispersion from the experiment depicted in E-G.

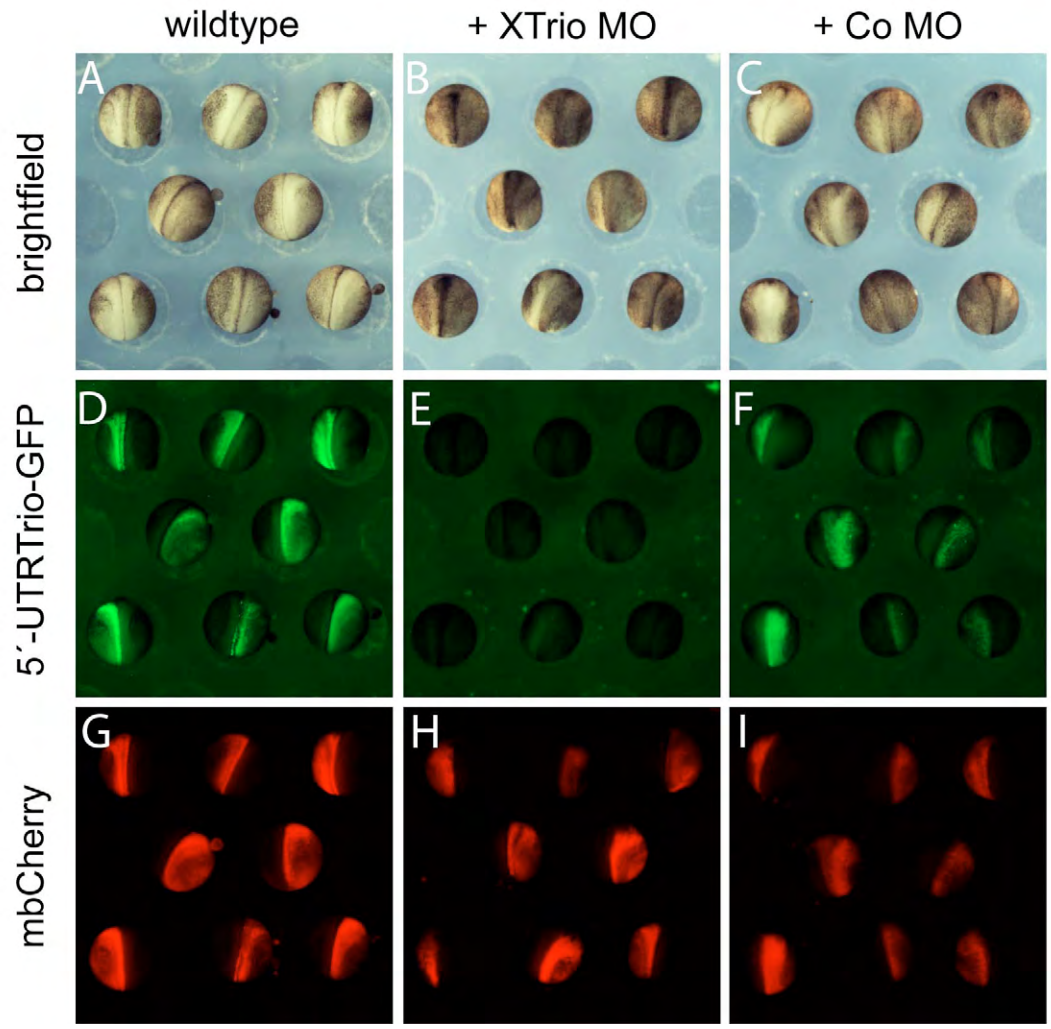


Fig. S4. Efficiency of Trio MO. Embryos were injected unilaterally with 5'UTRTrio-GFP and membraneCherry mRNA (A, D, G), together with TrioMO (B, E, H) or a control MO (C, F, I). Pictures are shown for bright field (A-C), GFP (D-F) or RFP (G-I) fluorescence. Note that injection of Trio MO leads to a strong inhibition of GFP fluorescence, indicating an efficient decrease of Trio-GFP protein.

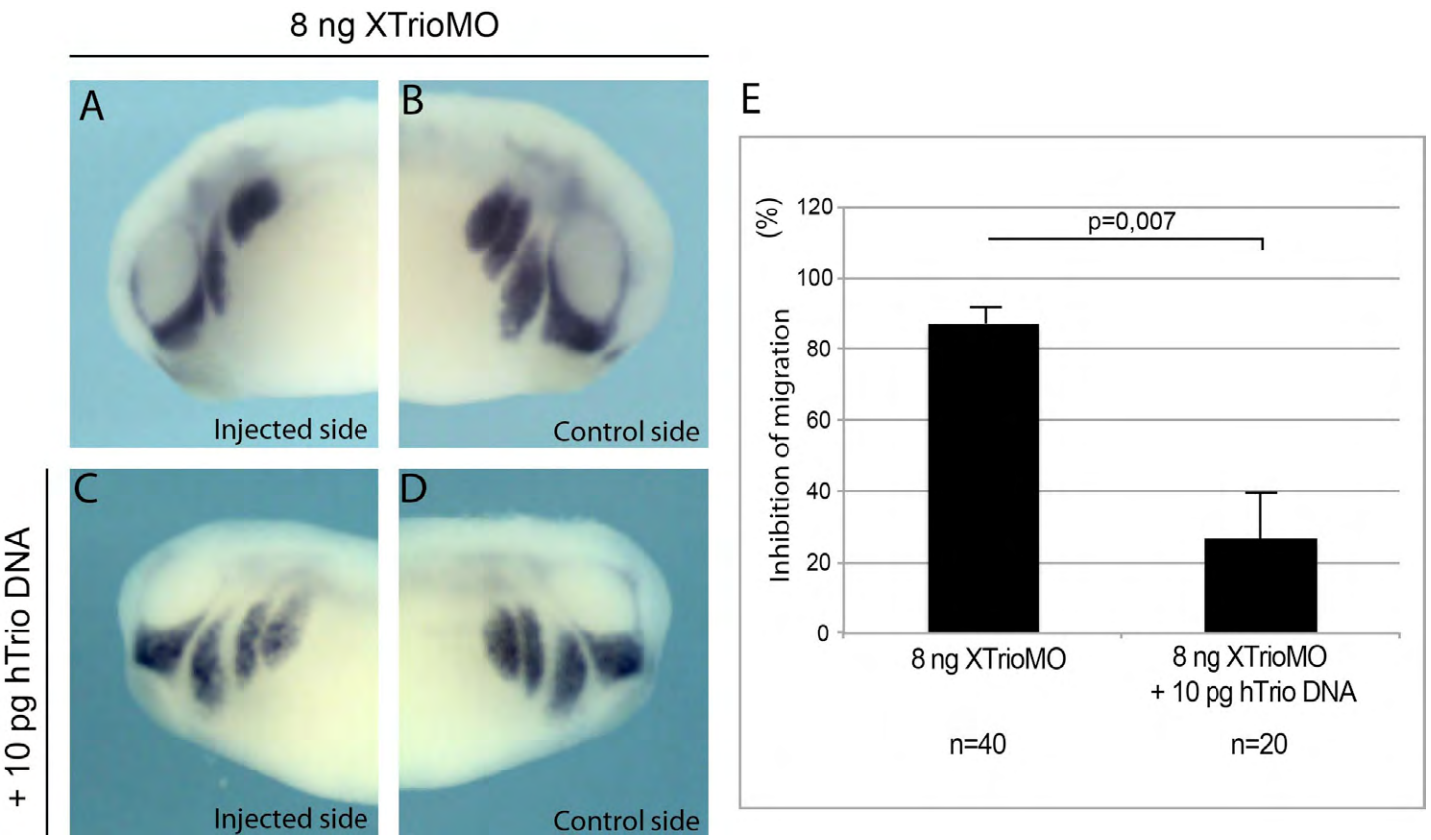


Fig. S5. Specificity of Trio MO. Embryos were injected at the two cell stage as indicated, fixed at stage 25 and migration of neural crest cells was analysed by in situ hybridization against Xtwist. (A, B) Injection of TrioMO leads to a clear inhibition of neural crest migration at the injected side. (C, D) This inhibition of neural crest migration was rescued by co-injection of human Trio that is not recognized by Trio MO. (E) Quantification of inhibition in neural crest migration produced by TrioMO and its rescue by co-injection of human Trio DNA. This efficient rescue of TrioMO by Trio DNA shows the specificity of Xenopus TrioMO used in this work.

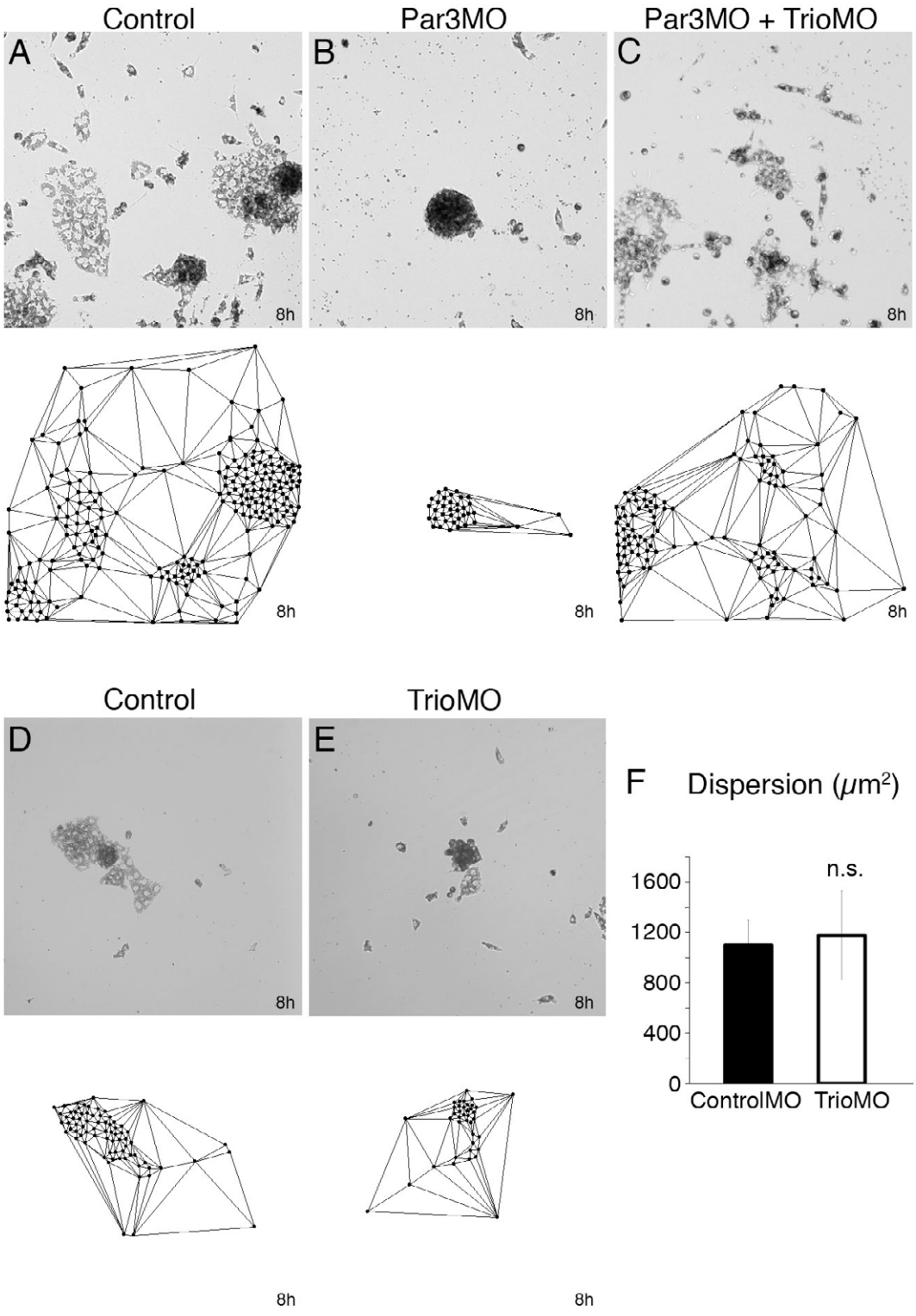
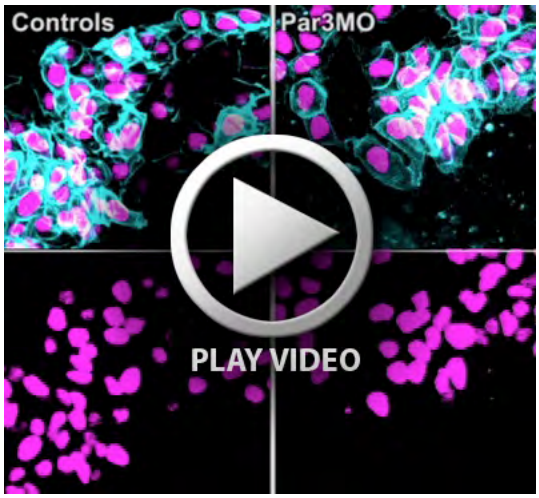


Fig. S6. Trio inhibition can restore normal dispersion in Par3MO cells. (A-D) Dispersion assay in control NC cells (A), Par3MO NC cells (B), Par3MO NC cells co-injected with TrioMO (C). (D-E) Dispersion assay in control NC cells (D) and TrioMO NC cells (E). Note that inhibition of Trio alone does not affect NC dispersion. (F) Quantification of NC dispersion from the experiment depicted in D-E.

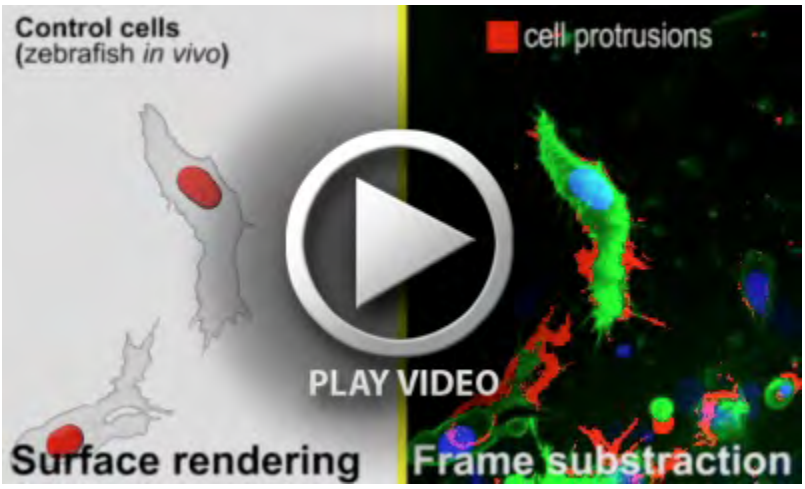
Movie 1. Neural crest cell migration assay with control cells (left panel) and cells injected with Par3MO. Note that Par3MO-injected cells fail to disperse. 10X Objective, 1 picture / 5 minutes, 8 hours.



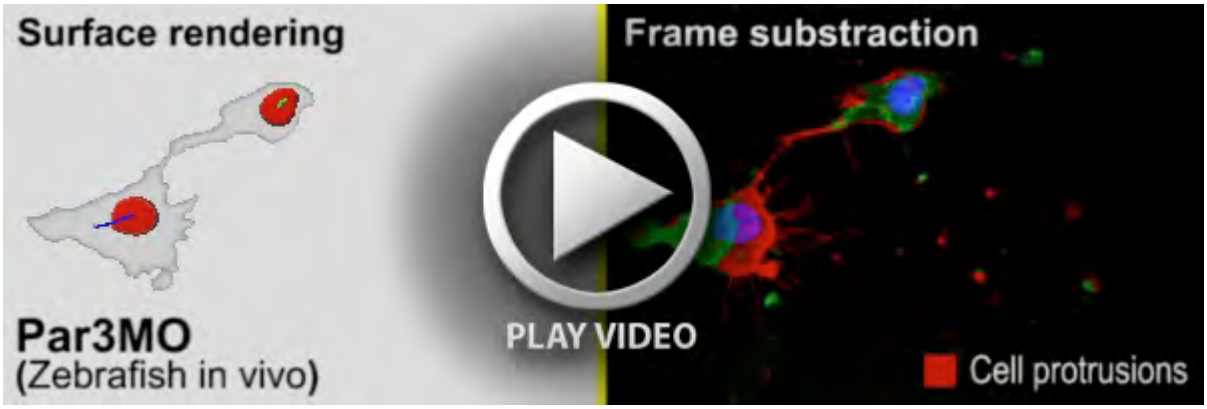
Movie 2. In vivo time-lapse movie of zebrafish neural crest cells. Control cells (left panels), Par3MO-injected cells (right panels). Note that Par3MO cells fail to disperse. Confocal spinning disk microscope 40 objective, 1 picture / X minutes, X hours.

Movie 3. Control (upper panel) and Par3MO-injected (lower panel) neural crest cells expressing membrane-GFP. Note that Par3MO cells tend to overlap with one another. 63X Objective, 1 picture / 30 seconds, 30 minutes.

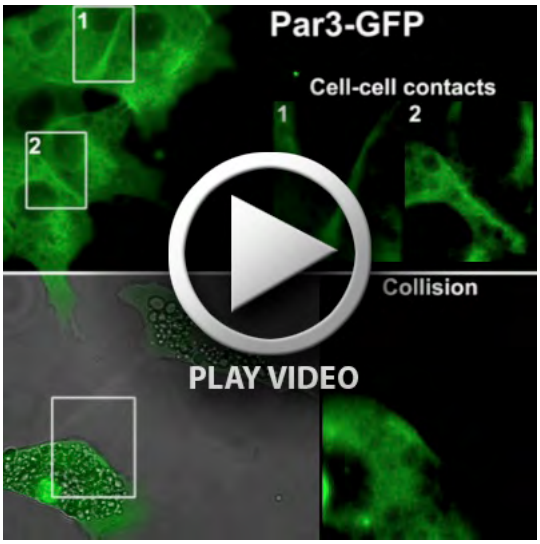
Movie 4. Collision assay with control (left panel) and Par3MO-injected (right panel) neural crest cells. 10X Objective, 1 picture / 5 minutes, 45 minutes.



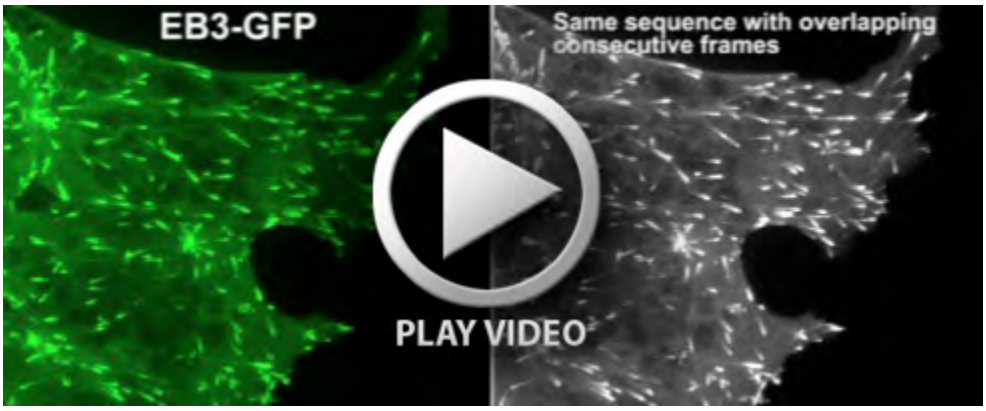
Movie 5. In vivo time-lapse movie of zebrafish neural crest cells. Collision between two control neural crest cells. Left panel shows surface rendering and tracks of the colliding cells. Right panel shows the substraction of consecutive frames in which growing membranes (protrusions) are colour-coded in red. 40 objective, 1 picture / 10 minutes, X minutes.



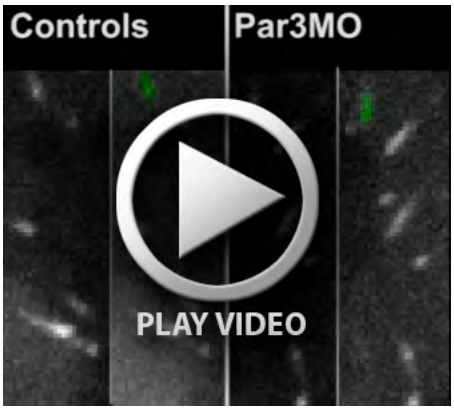
Movie 6. In vivo time-lapse movie of zebrafish neural crest cells. Collision between two Par3MO-injected neural crest cells. Left panel shows surface rendering and tracks of the colliding cells. Right panel shows the substraction of consecutive frames in which growing membranes (protrusions) are colour-coded in red. 40 objective, 1 picture / 5 minutes, X minutes.



Movie 7. In vitro time-lapse movie with control neural crest cells overexpressing Par3-GFP. Par3 is seen at cell-cell junctions in cells that are in direct apposition (upper panel) and accumulating at the cell-cell contact in colliding cells (lower panel). 63X Objective, 1 picture / 1 minute, 15 minutes.



Movie 8. In vitro time-lapse movie with control neural crest cells overexpressing EB3-GFP. Left panel shows EB3-GFP. Right panel shows the same sequence with overlapping consecutive frames to make microtubule trajectories more evident. 63X Objective, 1 picture / 6 seconds, 7.8 minutes.



Movie 9. Examples of microtubule tips labelled with EB3-GFP arriving at the region of cell-cell contact in control Neural crest cells (left panel) or Par3MO-injected neural crest cells. 63X Objective, 1 picture / 2 seconds, 36 seconds.

Movie 10. Neural crest cell migration assay with control cells (top left panel), cells injected with Par3MO (top right panel) or cells injected with Par3MO and treated with nocodazole (bottom left panel) or Rac1 inhibitor (NSC 23766, bottom right panel). 10X Objective, 1 picture / 7 minutes, 8 hours.

Movie 11. Collision assay with control cells (top left panel), cells injected with Par3MO (top right panel) or cells injected with Par3MO and treated with nocodazole (bottom left panel) or Rac1 inhibitor (NSC 23766, bottom right panel). 10X Objective, 1 picture / 5 minutes, 50 minutes.

Movie 12. Neural crest cell migration assay with control cells (left panel), cells injected with Par3MO (middle panel) or cells co-injected with Par3MO and TrioMO (right panel). 10X Objective, 1 picture / 5 minutes, 8 hours.

Movie 13. Collision assay with control neural crest cells (left panel), cells injected with Par3MO (middle panel) or cells co-injected with Par3MO and TrioMO (right panel). 10X Objective, 1 picture / 7 minutes, 49 minutes.