

Goto et al. Supplemental Figure 1

### Supplemental Figure 1. Time course expression of vimentin related genes

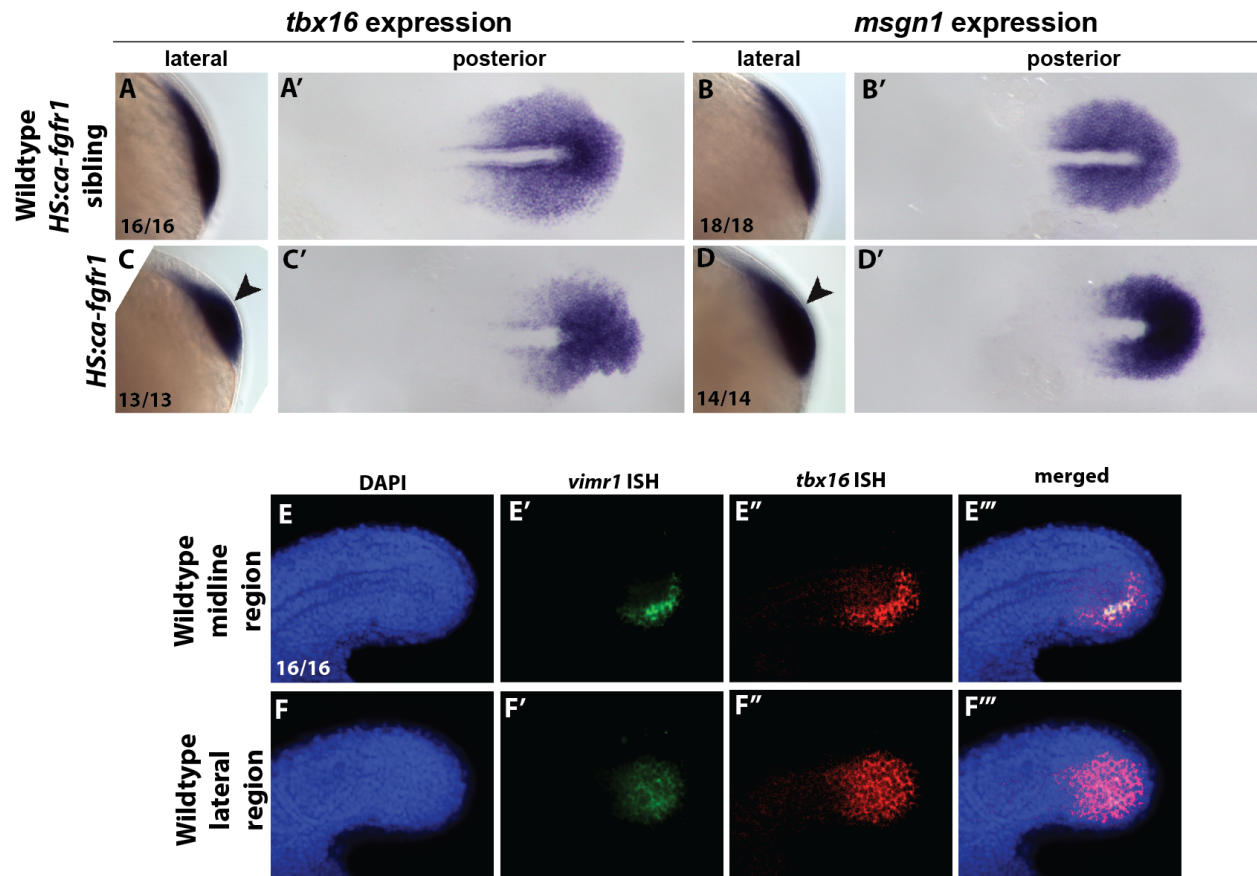
*Vimr1* (A-J) and *Vimr2* (K-T) expression in wildtype embryos fixed at different stages. The beginning of notochord expression at the 18-somite stage is indicated by arrowheads (G and Q). Shield staged embryos were taken laterally with the shield towards the right. Embryos from 8-somite stage and onward were taken laterally with anterior towards the bottom.



Goto et al. Supplemental Figure 2

## Supplemental Figure 2. Phylogenetic analysis of intermediate filaments

Bayesian phylogenetic analysis of zebrafish vimentin-like genes with other intermediate filaments with laminin genes serving as an outgroup. The numbers at the node specify Bayesian posterior probabilities. Species abbreviations with common names in parentheses: Ca, *Carassius auratus* (goldfish); Ce, *Caenorhabditis elegans* (nematode); Clf, *Canis lupus familiaris* (Dog); Dm, *Drosophila melanogaster* (fruit fly); Dr, *Danio rerio* (Zebrafish); Hs, *Homo sapiens* (human); Lf, *Lampetra fluviatilis* (Lamprey); Mm, *Mus musculus* (mouse); On, *Oreochromis niloticus* (Nile tilapia); Sp, *Stegastes partitus* (bicolor damselfish); Tr, *Takifugu rubripes* (pufferfish); Xt, *Xenopus tropicalis* (frog)



Goto et al. Supplemental Figure 3

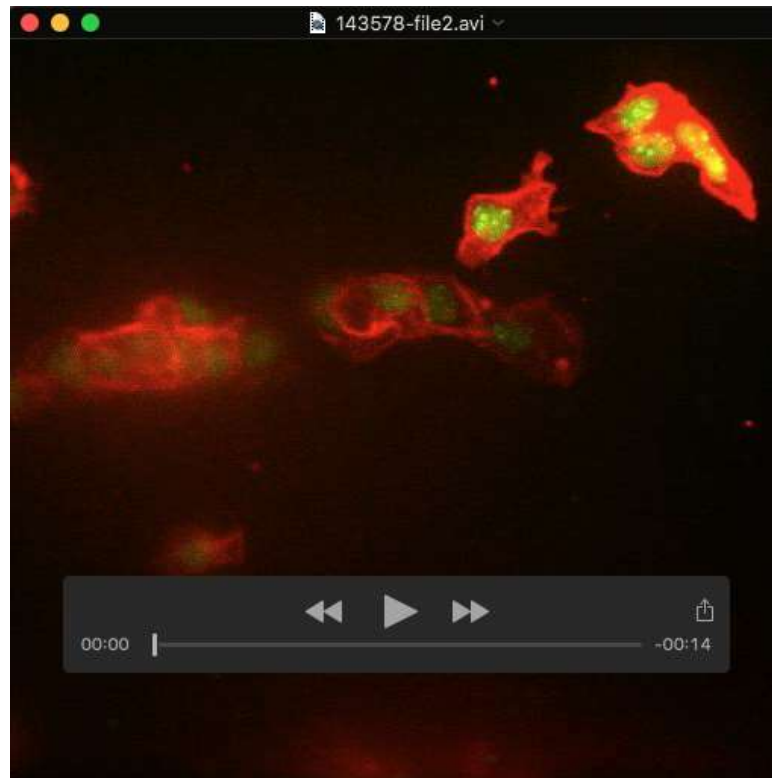
**Supplemental Figure 3. Expression of mesodermal maturation genes in FGF overactivated tailbud and expression domains of *vimr1* and *tbx16* in the wildtype tailbud.** HS:*cafgfr1* and wildtype embryos were heat-shocked at bud stage and fixed at 8-somite stage to examine the effect of FGF overactivation on maturation gene expression. Overactivation of FGF leads to strong expression of *tbx16* and *msgn1*, which expands into the NMP region (C, C', D, D') compared to the control (A, A', B, B'). Double fluorescent *in situ* hybridization of *tbx16* and *vimr1* in a wild-type 20-somite stage embryo. As the mesodermal progenitors move from the dorsal region to ventral region, the cells turn on *vimr1* and *tbx16* (E, E', E'', E'''). However, as the cells enter the maturation zone in the lateral regions, the cells turn off *vimr1* (F, F', F'', F''').

**Table S1. Statistics for time lapse tracking.** P values were calculated by Student's t-test. S indicates significant ( $p < 0.05$ ), while NS implies nonsignificant ( $p > 0.05$ ).

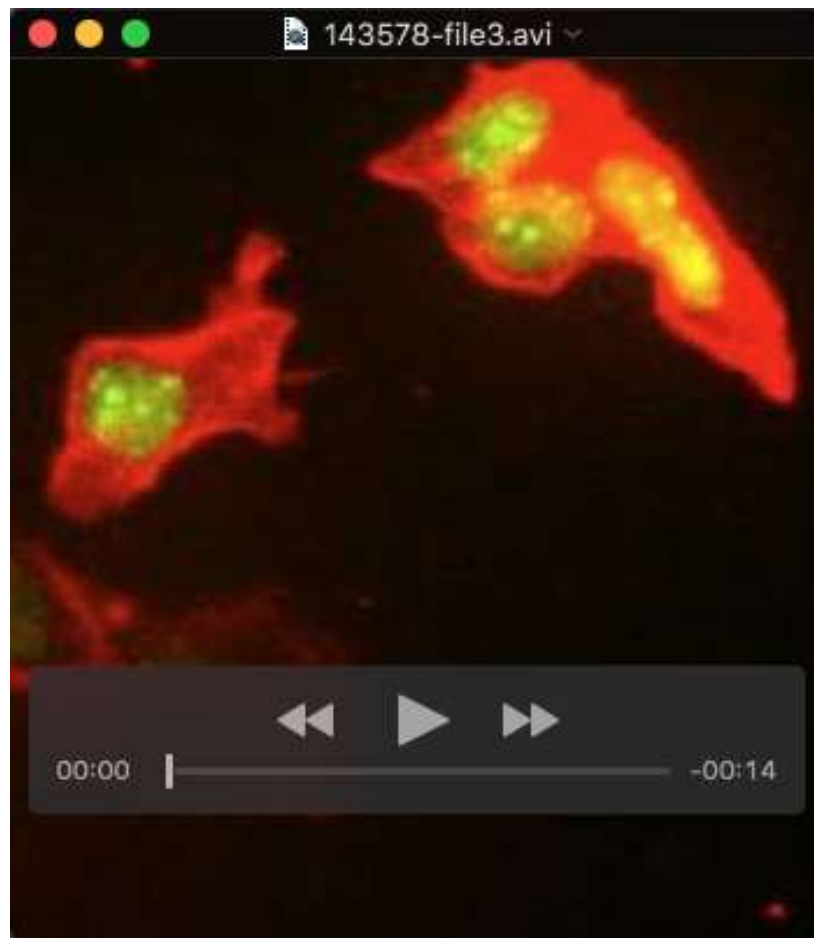
Embryo	number of embryos	Total tracks	Displacement		Mean Track Speed		Track straightness	
			P val compared to wildtype-DMSO	P val compared to HStbx16-SU5402	P val compared to wildtype-DMSO	P val compared to HStbx16-SU5402	P val compared to wildtype-DMSO	P val compared to HStbx16-SU5402
wildtype-DMSO	4	268						
HS:tbx16-DMSO	3	177	p=0.7425; NS	p=0.433; NS	p=0.0323; S	p=0.2539; NS	p=0.1601; NS	p=0.9717; NS
wildtype-SU5402	3	143	p=0.0323; S	p=0.0073; S	p=0.0073; S	p=0.0151; S	p=0.1886; NS	p=0.1116; NS
HS:tbx16-SU5402	3	223						
HS:caFGFr1-DMSO	3	169	p=0.5225; NS		p=0.0065; S		p=0.3332; NS	

**Table S2. Raw data of transplanted lineage analysis and statistics.** P values were calculated by Fisher's exact test. S indicates significant ( $p < 0.05$ ), while NS denotes nonsignificant ( $p > 0.05$ ).

Embryo	number of embryos	Total muscle cells	Total nonmuscle cells	% muscle	% nonmuscle	P val compared to wildtype	P val compared to dnfgf
wildtype	10	167	68	71.064	28.936		
HS:dnfgfr1	10	33	127	20.625	79.375	$p < 0.0001$ ; S	
pmsgn1/wildtype	9	234	88	72.671	27.329	$p = 0.7028$ ; NS	
pmsgn1/HS:dnfgfr1	10	216	191	53.071	46.929	$p < 0.0001$ ; S	$p < 0.0001$ ; S
ptbx16/wildtype	8	177	96	64.835	35.165	$p = 0.153$ ; NS	
ptbx16/HS:dnfgfr1	9	97	81	54.494	45.506	$p = 0.0006$ ; S	$p < 0.0001$ ; S

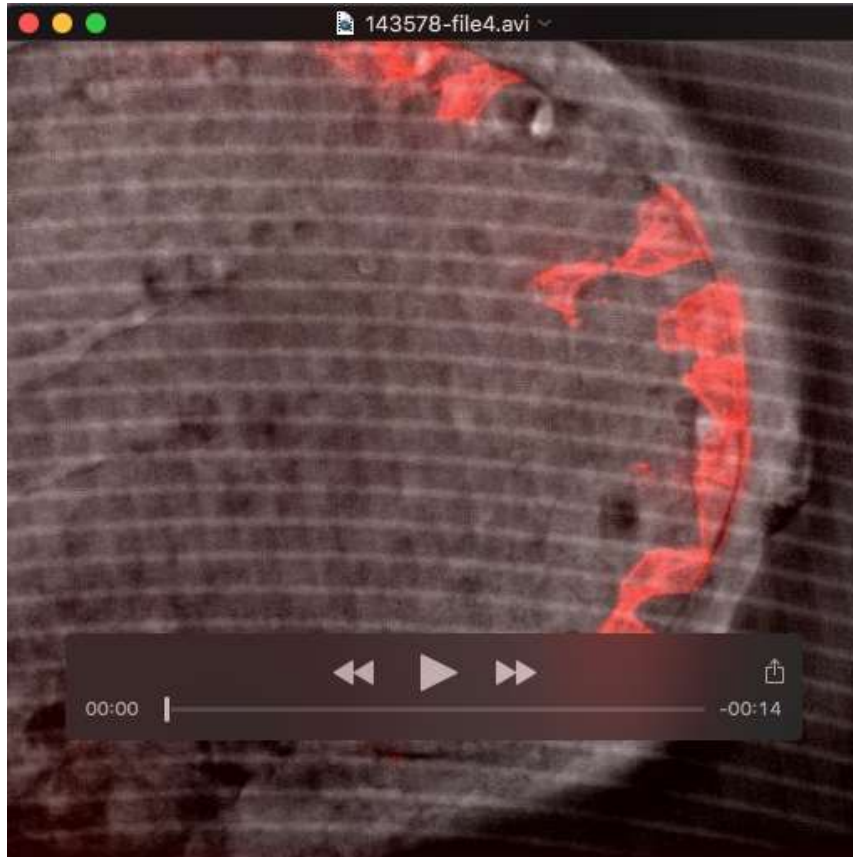


**Movie 1.** Time lapse movie of transplanted *HS:(CAAX)mCherry-p2a-(NLS)kikume* during the 1<sup>st</sup> EMT step. Cells of interest are in the upper right of the movie. Images were acquired every 5 minutes for 8 hours.

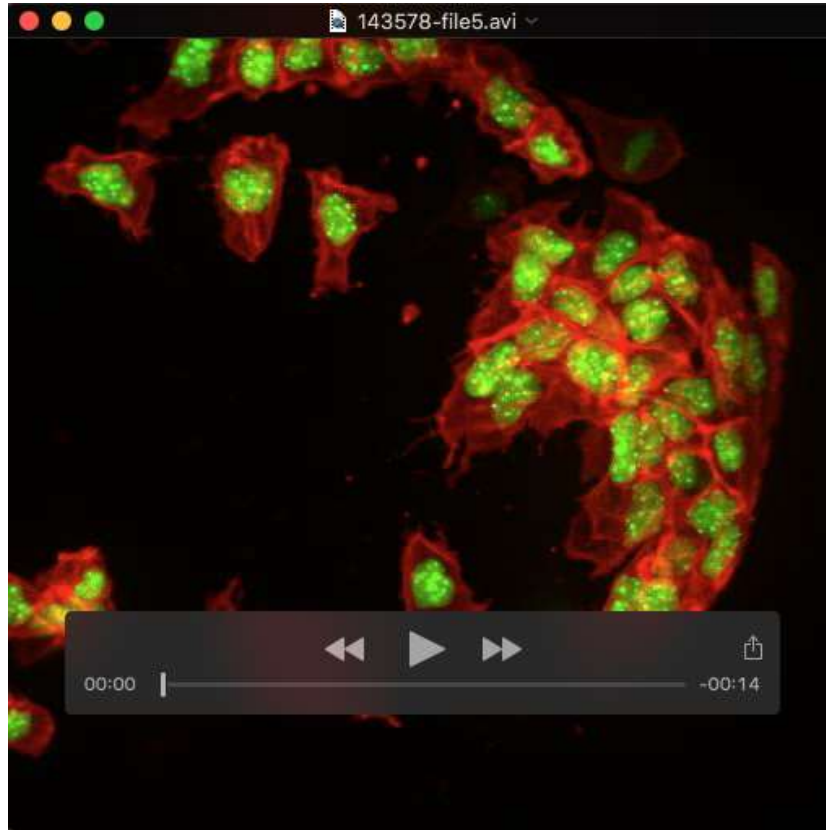


**Movie 2.** Cropped version of movie 1, focusing on cells of interest.

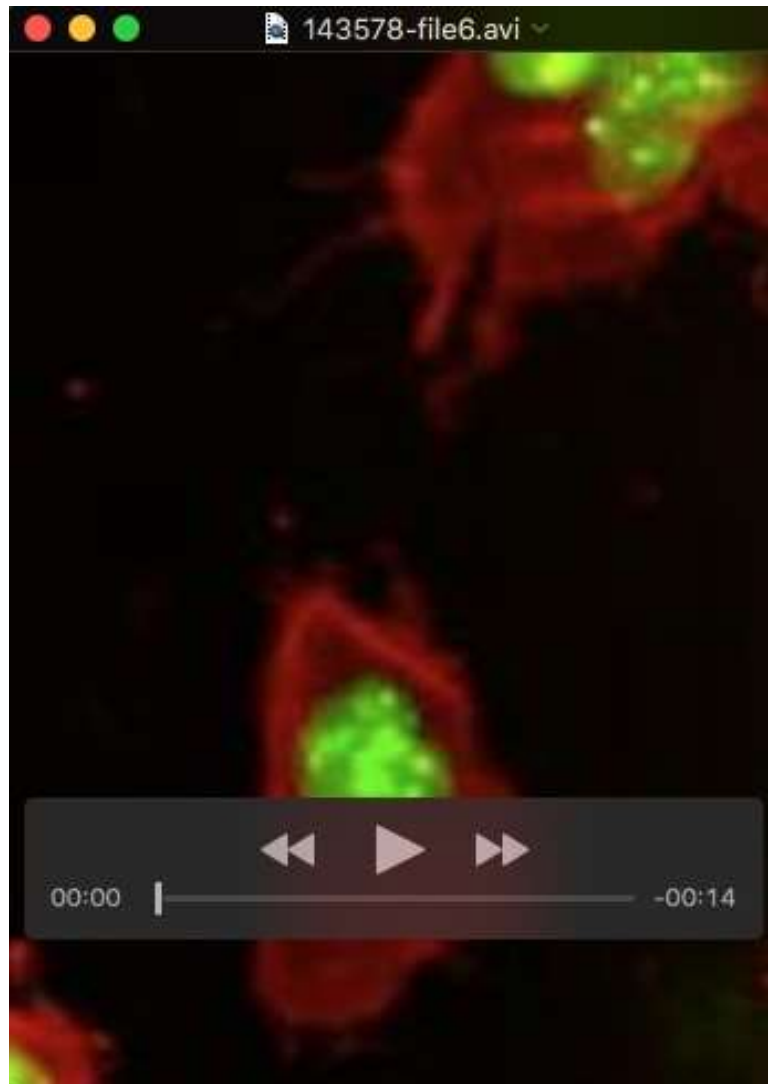




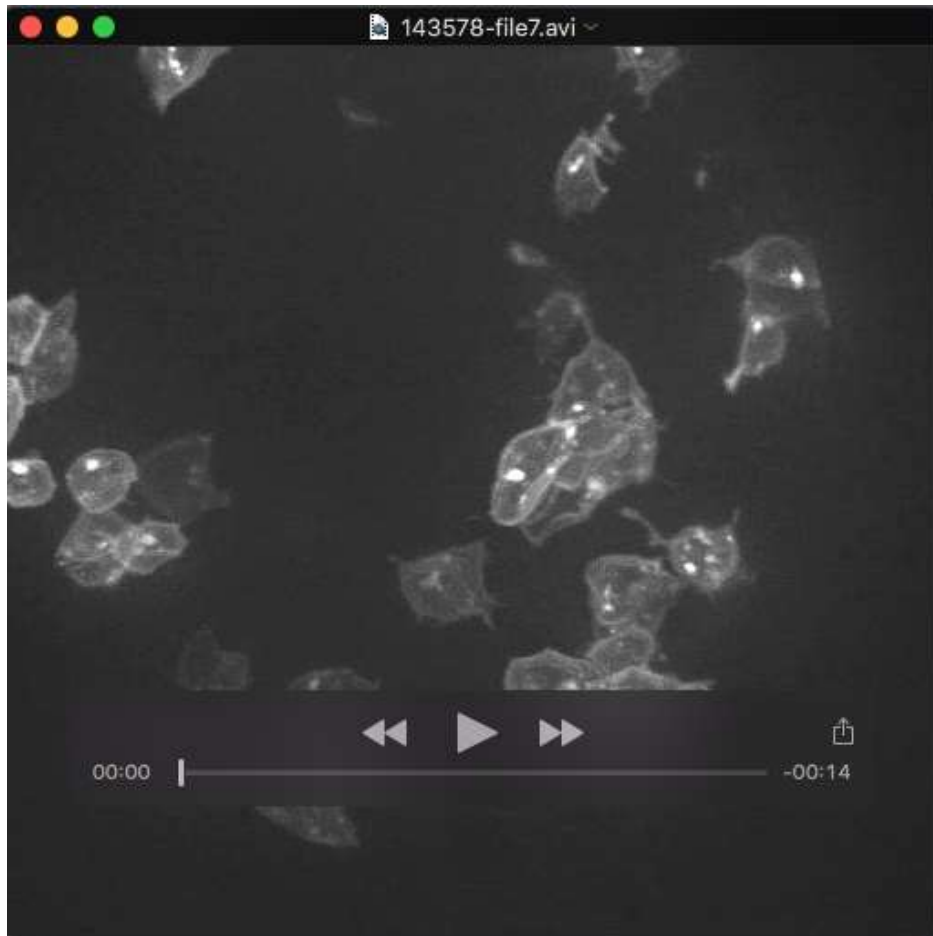
**Movie 3.** Time lapse movie of transplanted *HS:TCF $\Delta$ C* cells. . Images were acquired every 5 minutes for 8 hours.



**Movie 4.** Time lapse movie of transplanted *HS:(CAAX)mCherry-p2a-(NLS)kikume* during the 2<sup>nd</sup> EMT step. The cell of interest is in the lower left of the movie. . Images were acquired every 5 minutes for 8 hours.



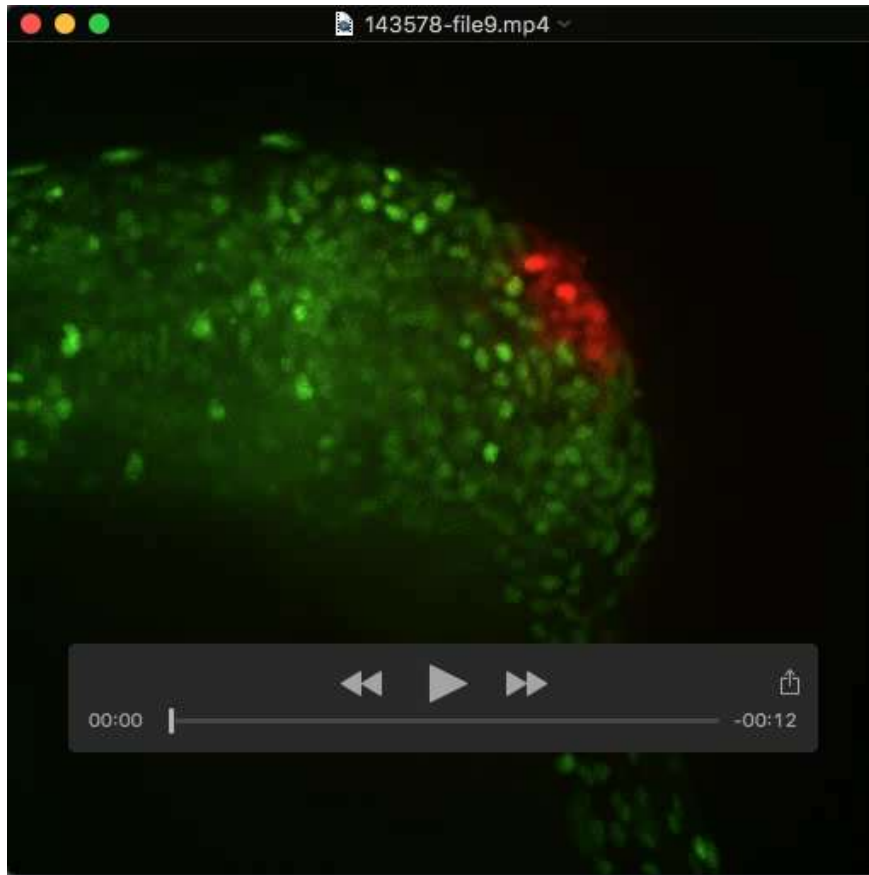
**Movie 5. Cropped version of movie 4, focusing on the cell of interest.**



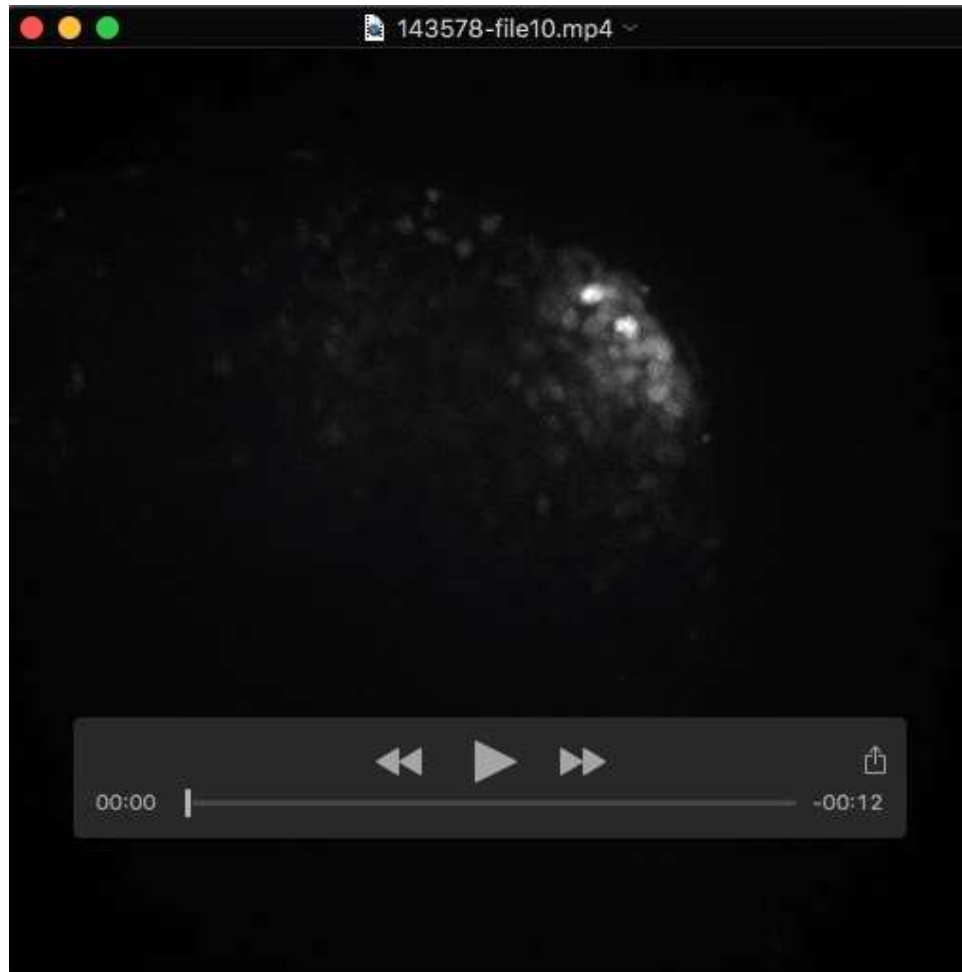
**Movie 6. Time lapse movie of transplanted *HS:dnfgfr1* cells.** The cell of interest is in the lower left of the movie. . Images were acquired every 5 minutes for 8 hours.



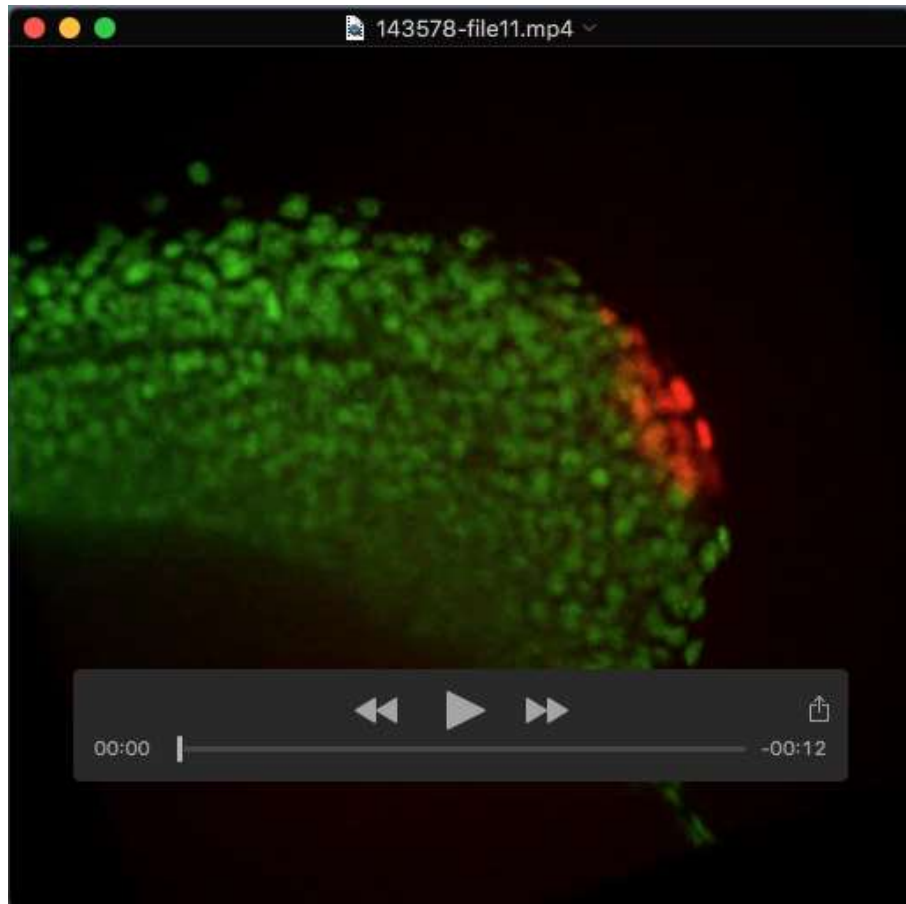
**Movie 7. Cropped version of movie 6, focusing on the cell of interest.**



**Movie 8. Time lapse movie of photoconverted cells in a wildtype tailbud (merged)** A movie of wildtype photoconverted red tailbud cell with Z projection from 5 $\mu$ m Z-stacks merged with a projection of nonconverted cells in green. The embryo was treated with DMSO (vehicle). Images were acquired every 5 minutes for 5 hours. (5 frames per second).

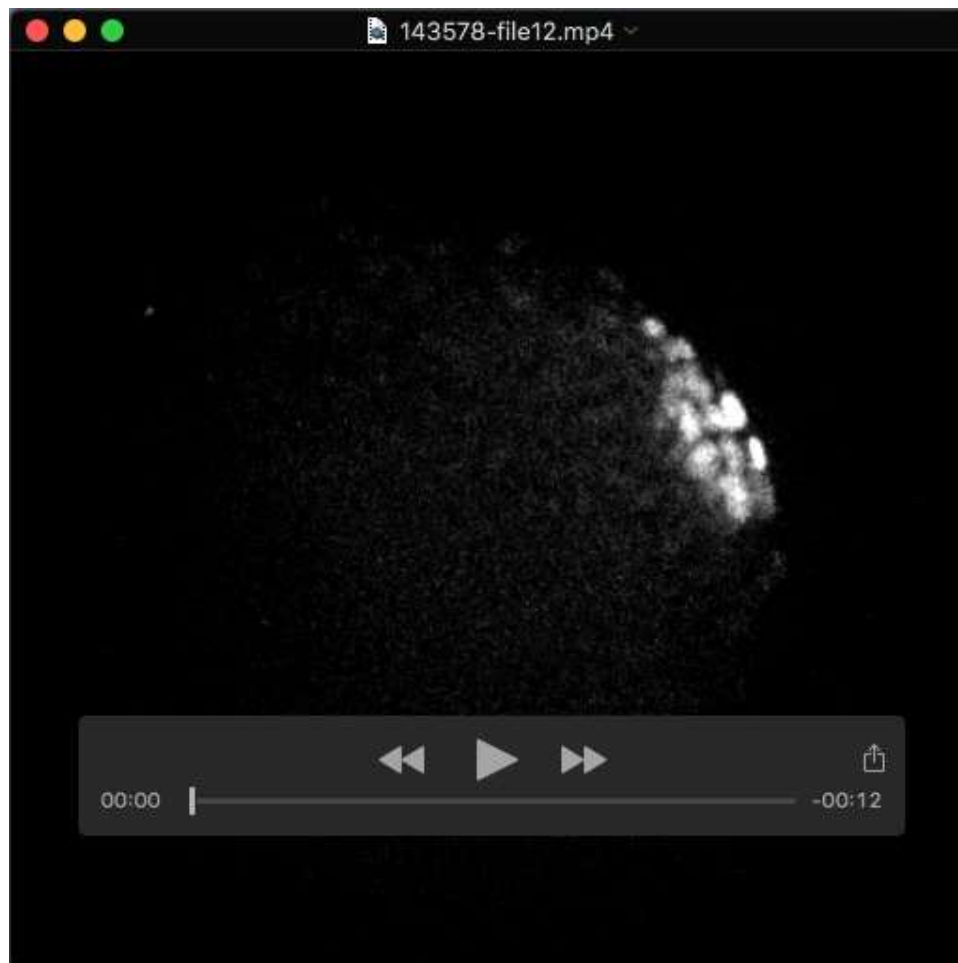


**Movie 9. Time lapse movie of photoconverted cells in a wildtype tailbud (photoconverted cells only)** A movie of wildtype photoconverted tailbud cell with Z projection from 5 $\mu$ m Z-stacks. The embryo was treated with DMSO (vehicle). Images were acquired every 5 minutes for 5 hours. (5 frames per second).

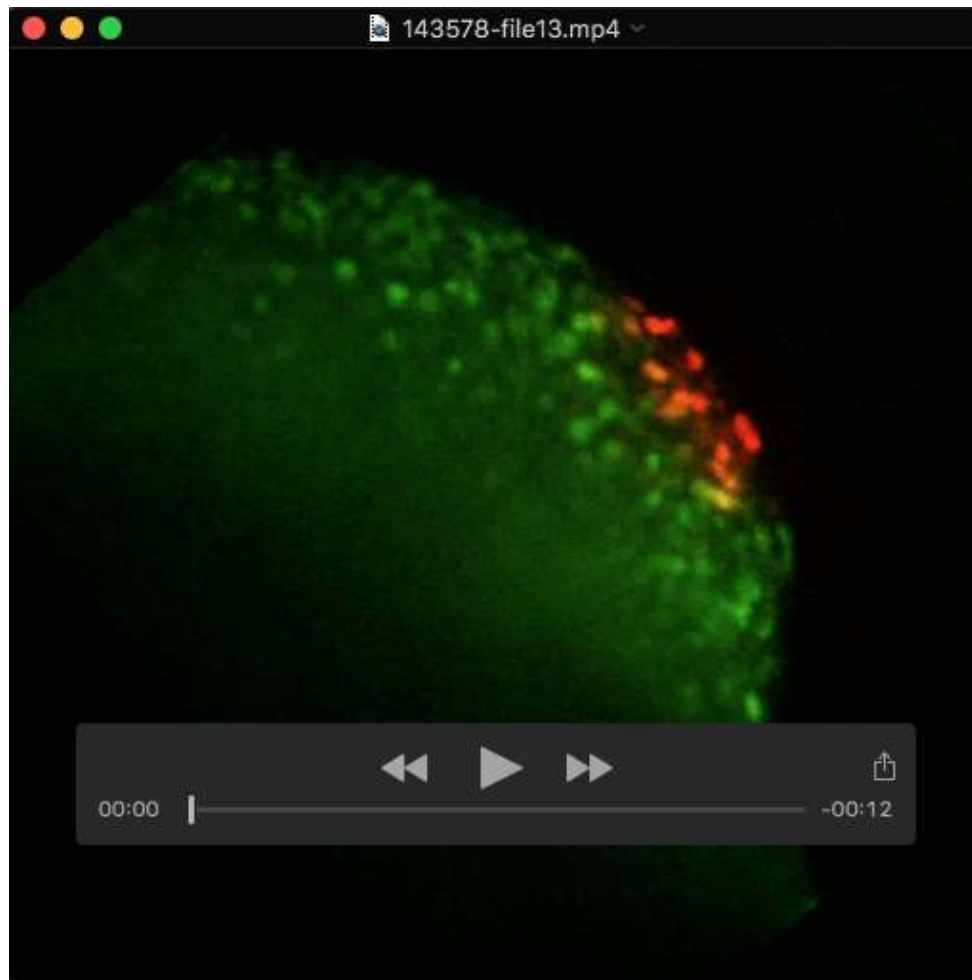


**Movie 10. Time lapse movie of photoconverted *tbx16* overexpressed progenitors in the tailbud (merged)** A movie of HS:*tbx16* photoconverted tailbud cells with Z projection from 5 $\mu$ m Z-stacks merged with unconverted green field projected plane. The embryo was treated with DMSO (vehicle). Images were acquired every 5 minutes for 5 hours. (5 frames per second).





**Movie 11. Time lapse movie of photoconverted *tbx16* overexpressed progenitors in the tailbud (photoconverted cells only)** A movie of DMSO treated HS:*tbx16* photoconverted tailbud cells with Z projection from 5 $\mu$ m Z-stacks. Images were acquired every 5 minutes for 5 hours. (5 frames per second).

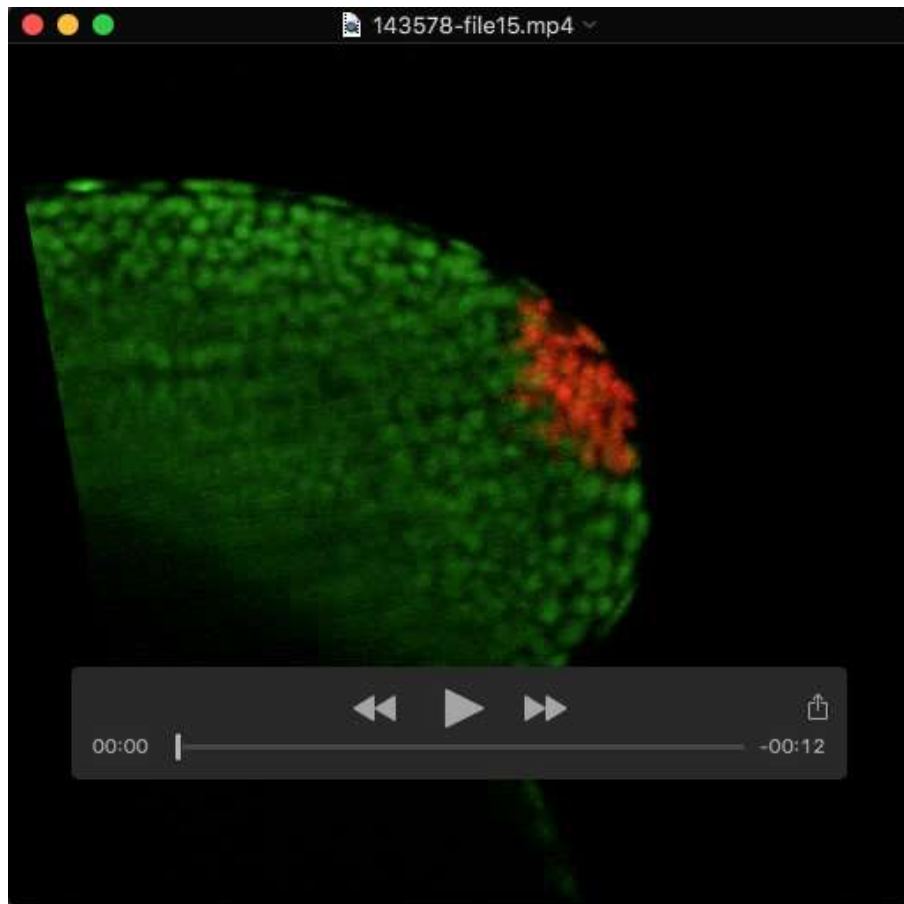


**Movie 12. Time lapse of photoconverted FGF inhibited progenitors in the tailbud (merged)**

Time lapse movie of SU5402 treated photoconverted tailbud cells with Z projection from 5um Z-stacks merged with a projection of nonconverted cells in green. Images were acquired every 5 minutes for 5 hours. (5 frames per second).



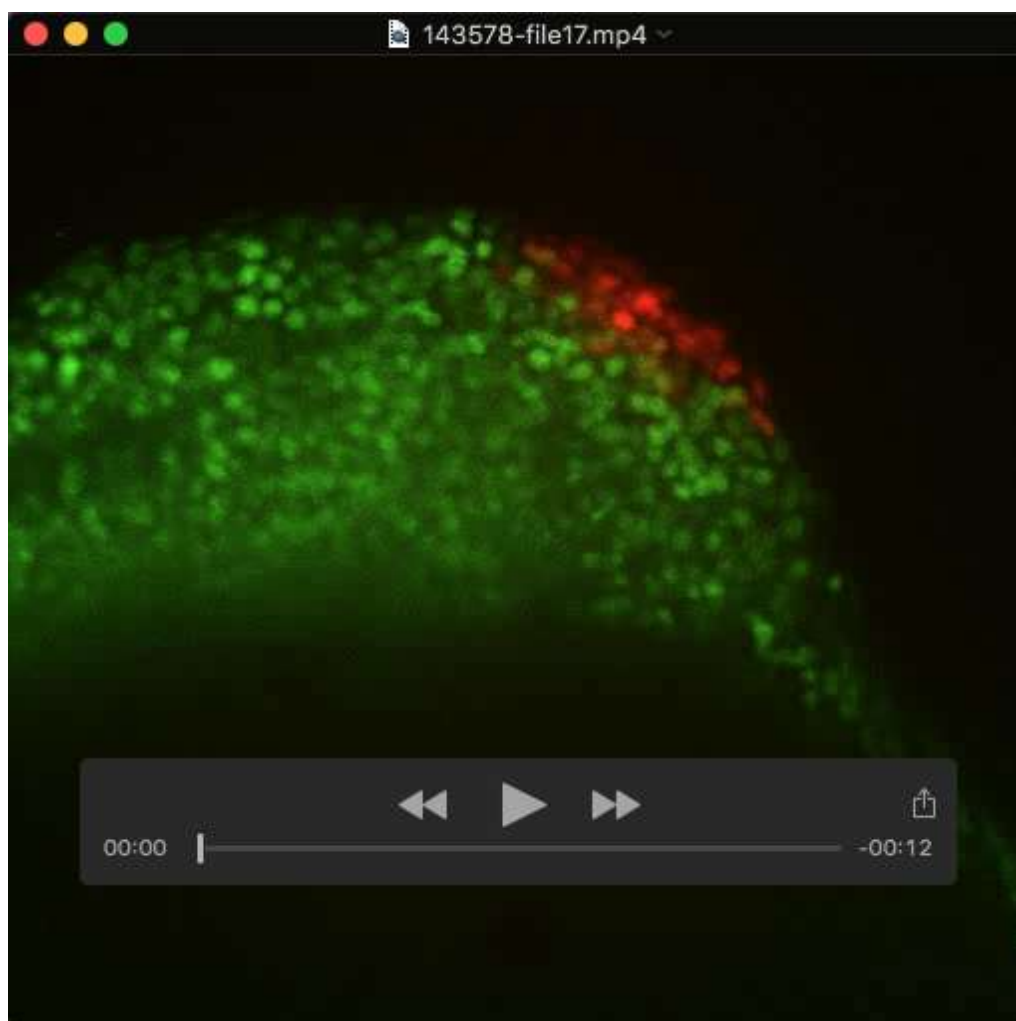
**Movie 13. Time lapse of photoconverted FGF inhibited progenitors in the tailbud (photoconverted cells only)** Time lapse movie of SU5402 treated photoconverted tailbud cells with Z projection from 5 $\mu$ m Z-stacks. Images were acquired every 5 minutes for 5 hours. (5 frames per second).



**Movie 14. Time lapse of photoconverted *tbx16* overexpressing progenitors in the SU5402 treated tailbud (merged)** Time lapse movie of SU5402 treated photoconverted *HS:tbx16* tailbud cells with Z projection from 5um Z-stacks merged with a projection of unconverted green field plane. Images were acquired every 5 minutes for 5 hours. (5 frames per second).

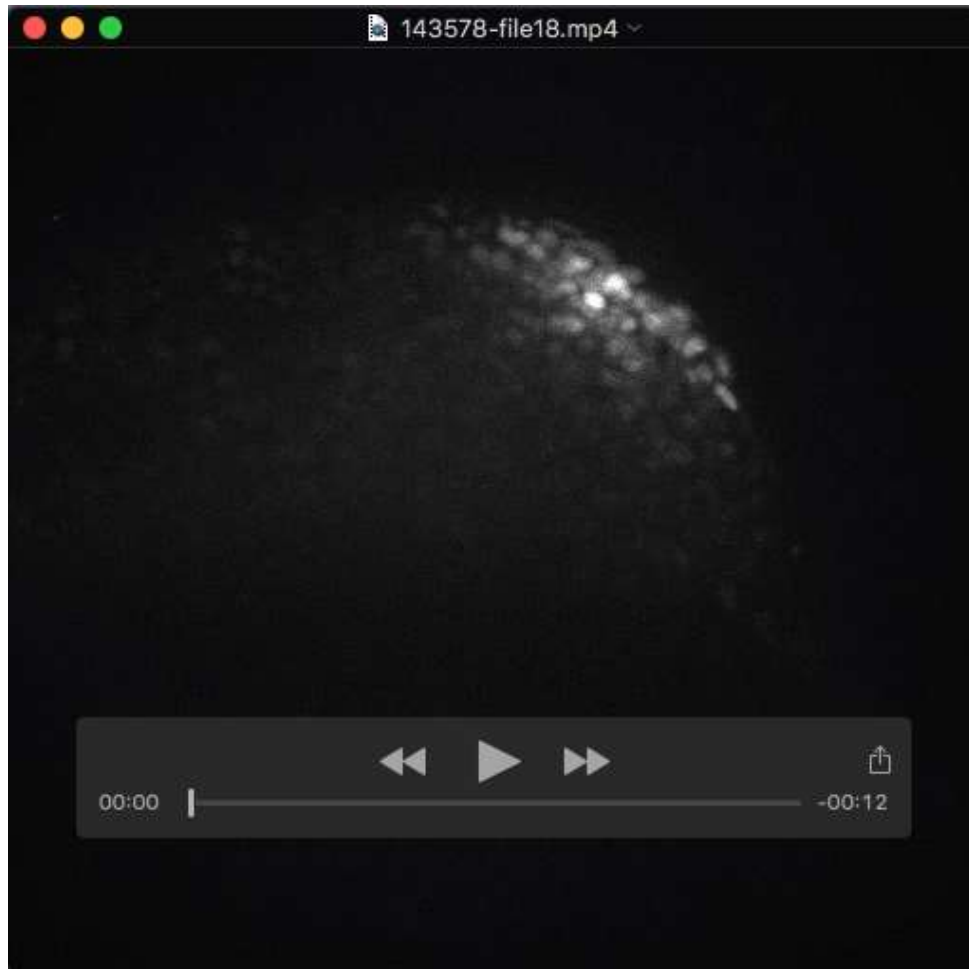


**Movie 15. Time lapse of photoconverted *tbx16* overexpressing progenitors in the SU5402 treated tailbud (photoconverted cells only)** Time lapse movie of SU5402 treated photoconverted nuclei of *HS:tbx16* tailbud cells with Z projection from 5um Z-stacks. Images were acquired every 5 minutes for 5 hours. (5 frames per second).



**Movie 16. Time lapse of photoconverted FGF overactivating progenitors in the tailbud**

**(merged)** Time lapse movie of DMSO treated photoconverted *HS:caFGFr1* tailbud cells with Z projection from 5 $\mu$ m Z-stacks merged with a projection of unconverted nuclei in green. Images were acquired every 5 minutes for 5 hours. (5 frames per second).



**Movie 17. Time lapse of photoconverted FGF overactivating progenitors in the tailbud**

**(photoconverted cells only)** Time lapse movie of DMSO treated photoconverted *HS:caFGFr1*

tailbud cells with Z projection from 5um Z-stacks. Images were acquired every 5 minutes for 5 hours. (5 frames per second).