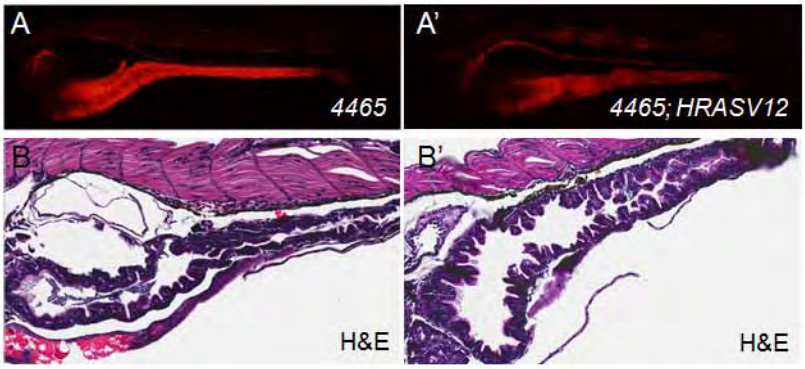
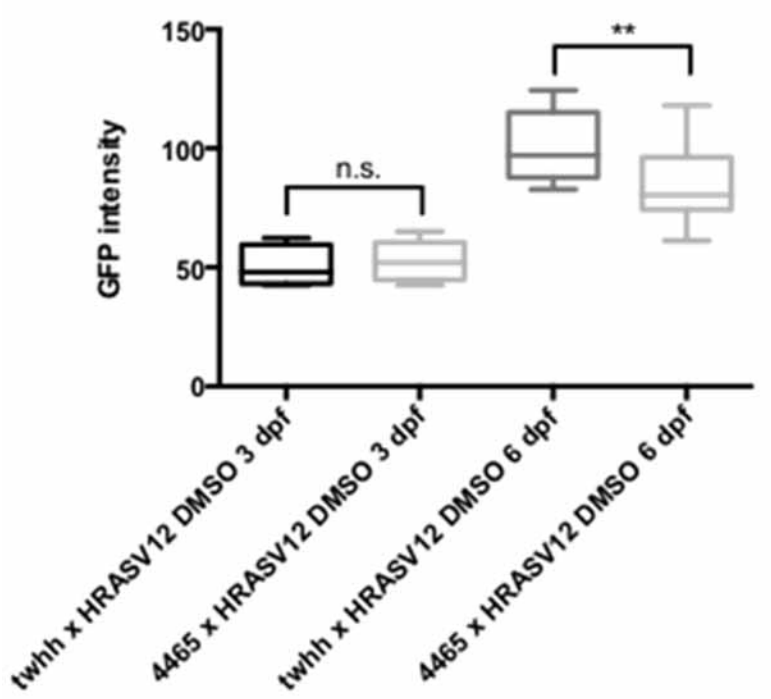
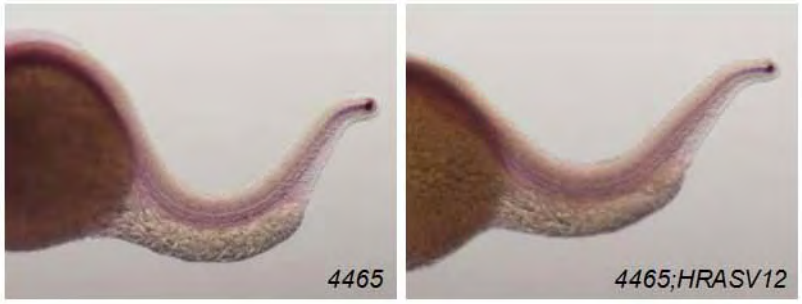


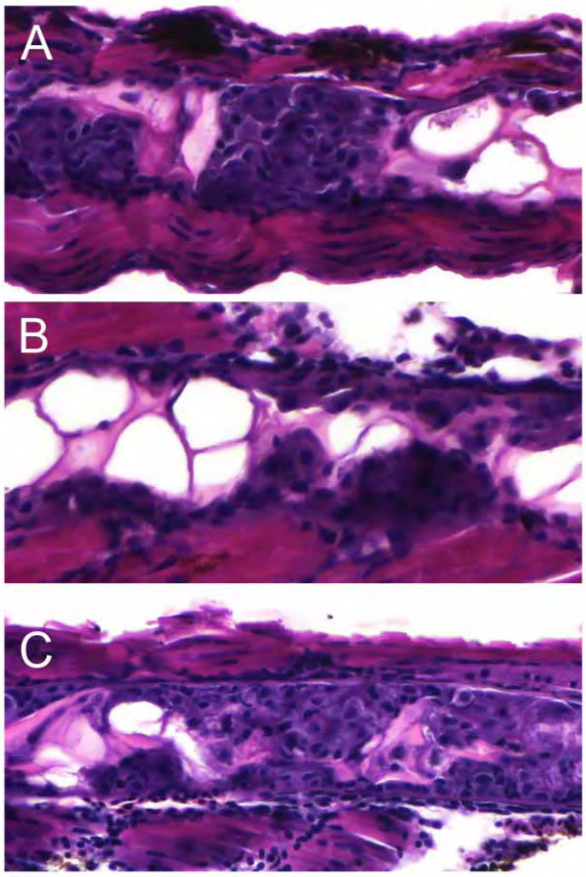
Supplemental Figure S1.
(A) Gross morphology of Tg(mü4465_13:Gal4,UAS:mCherry) using fluorescent microscopy. mCherry is expressed in the gut, kidney, and notochord. (B) H&E staining. (A', B') Same as in (A-B), but double transgenic 4465:Gal4;UAS:HRASV12.



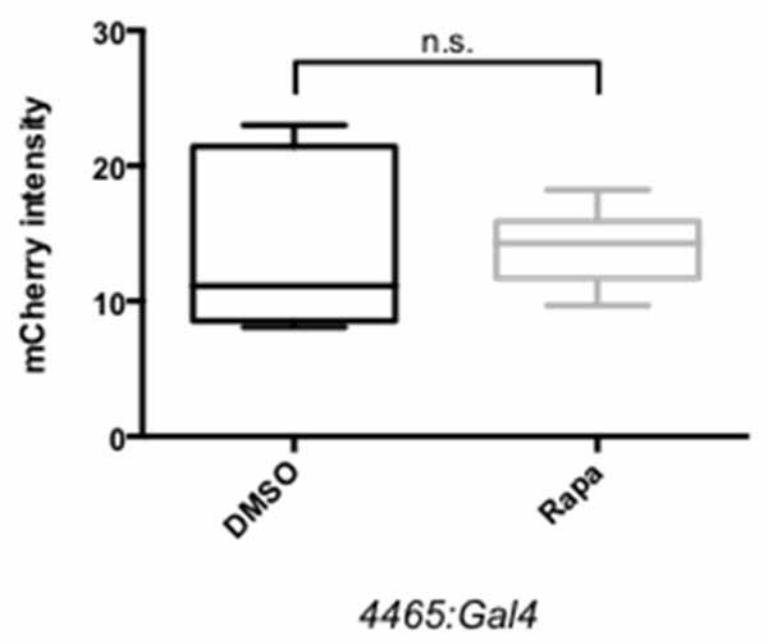
Supplemental Figure S2.
GFP intensity is similar at 3 dpf in 4465;HRASV12 and twhh;HRASV12 embryos, but increases significantly at 6 dpf in twhh;HRASV12 embryos compared to 4465;HRASV12 embryos (unpaired t-test; p=0.0025). Each time point and condition includes n>4 animals.



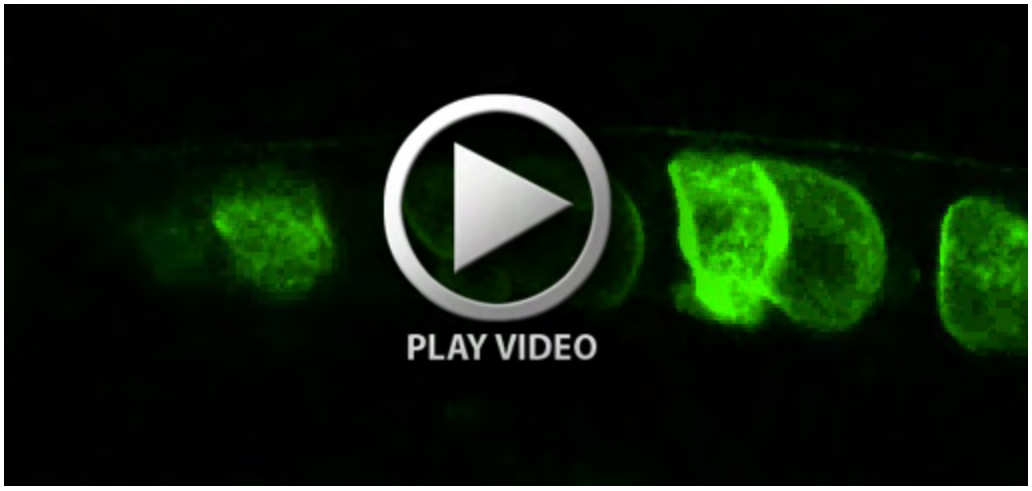
Supplemental Figure S3.
No tail (ntl; Brachyury) in situ hybridization of 30 hpf 4465;Gal4 and 4465;HRASV12 embryos. No significant change in ntl-expression upon HRASV12-overexpression, suggesting that Brachyury is independent of HRASV12. In situ hybridization was performed according to standard protocols for zebrafish embryos.



Supplemental Figure S4.
(A) 8 dpf old twwh:HRAS embryos treated with 1% DMSO (control) show the typical tumor phenotype. (B) 8 dpf old twwh:HRAS embryos treated with 15 μM LY294002 with slightly reduced tumor load; however, this effect was inconsistent and LY294002 treatment did not result in improved survival (data not shown). (C) 8 dpf old twwh:HRAS embryos treated with 10 μM bpV(HOpic). The observed tumor load was similar to that in control fish.



Supplemental Figure S5.
mCherry intensity in the anterior notochord of rapamycin- vs. DMSO-treated 4465:Gal4 larvae is not significantly different (unpaired t-test; p=0.9239). mCherry intensity was measured at 7 dpf in n=9 animals in rapamycin- and n=5 in DMSO-treated fish.



Supplemental Movie S1. Notochord cell proliferation in a twhh:Gal4;HRASV12 transgenic embryo.
Notochord cells were imaged by time lapse confocal microscopy at 36 hpf in twhh:Gal4;UAS:HRASV12 transgenic fish. Arrowheads mark cell division events. Images represent z-projection of 5 confocal slices (10um total thickness). Frame interval = 20min, number of frames = 35.

Supplemental Movie S2. Notochord cell proliferation in a 4465:Gal4;HRASV12 transgenic embryo.
Notochord cells were imaged by time lapse confocal microscopy at 3 dpf in 4465:Gal4;UAS:HRASV12 transgenic fish. Numerous cell division events are evident. Images represent z-projection of the entire thickness of the notochord. Frame interval = 20min, number of frames = 43.

Supplemental Movie S3. Lack of cell proliferation in a 4465:Gal4;UAS:YFP transgenic embryo.
Notochord cells were imaged by time lapse confocal microscopy at 3 dpf in 4465:Gal4;UAS:YFP transgenic fish. No cell division events are seen. Images represent z-projection of the entire thickness of the notochord. Frame interval = 15 min, number of frames = 49. The Tg(UAS:YFP) line will be described elsewhere.

Supplemental Table 1. Raw survival data of the Kaplan-Meier analysis.
We used Prism software to determine survival rates in rapamycin-treated vs. control embryos, in which each row represents one subject at a given time point (days elapsed = x value). The Y value stands for one individual embryo which is "1" when the embryo died at the specified time, and "0" when the embryo's data was censored at that time.

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