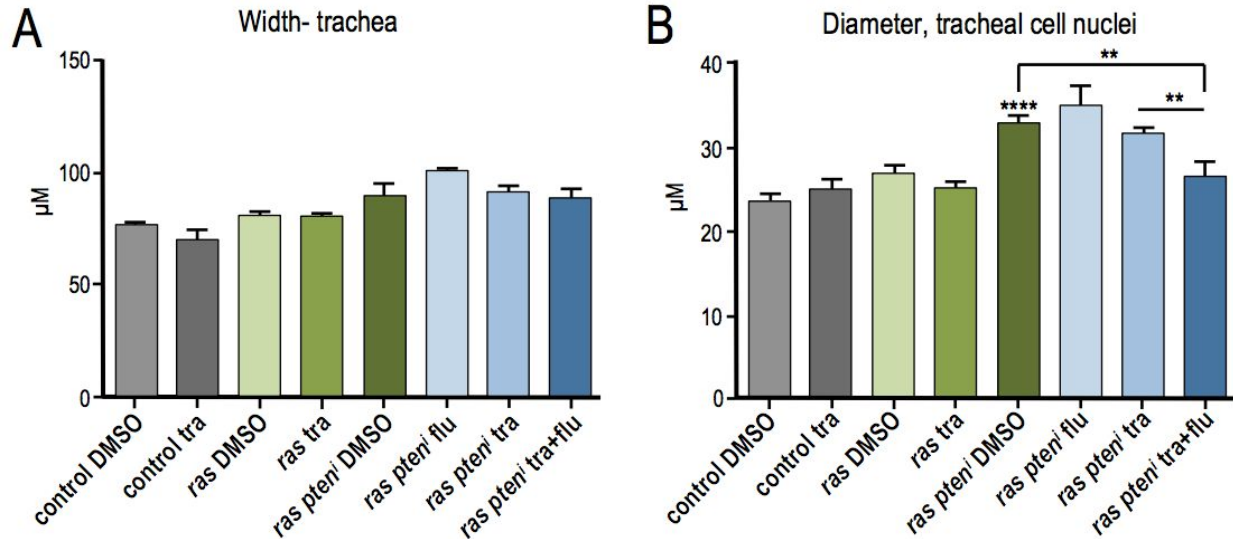


Drosophila Lung Cancer Models Identify Trametinib Plus Statin as Candidate Therapeutic

Supplemental Figures, Figure Legends

Supplemental Figure 1

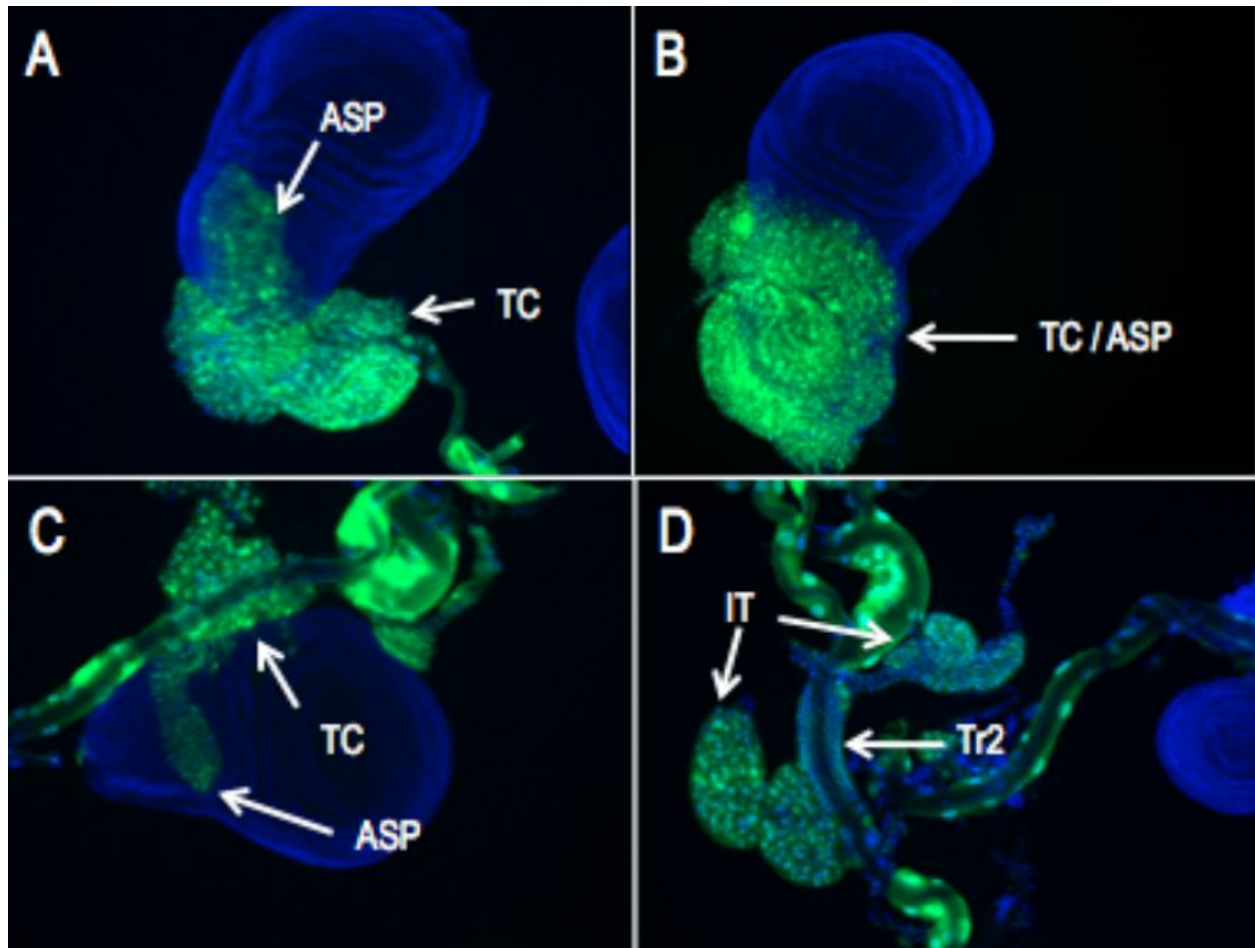


S1. Trachea width and tracheal cell nuclei diameter measurements. Related to Figure 1, Figure 3.

(A) *btl>Ras1* had little to no effect on the width of trachea; the apparent increase in *btl>Ras1,PTENi* tracheal width in DMSO controls did not reach significance. Trametinib and/or fluvastatin fed *btl>Ras1,PTENi* larvae also had larger trachea compared to controls, suggesting that there is a trend for increased trachea width in this combination that is not rescued by the drugs.

(B) *btl>Ras1* larvae exhibited slightly enlarged tracheal cell nuclei that were significantly increased upon reduction of PTEN. Trametinib had a mild, non-significant effect on the size of *btl>Ras1* and *btl>Ras1,PTENi* nuclei while fluvastatin alone had no effect. Combining fluvastatin with trametinib, however, strongly rescued *btl>Ras1,PTENi* nuclei enlargement. L3 larvae trachea were dissected as described in Materials and Methods. Trachea width and nuclei size were measured using Adobe Photoshop. To ensure consistency only the third tracheal metamere (Tr3) was measured. Drug concentrations: trametinib-1 μM , fluvastatin-100 μM .

Supplemental Figure 2

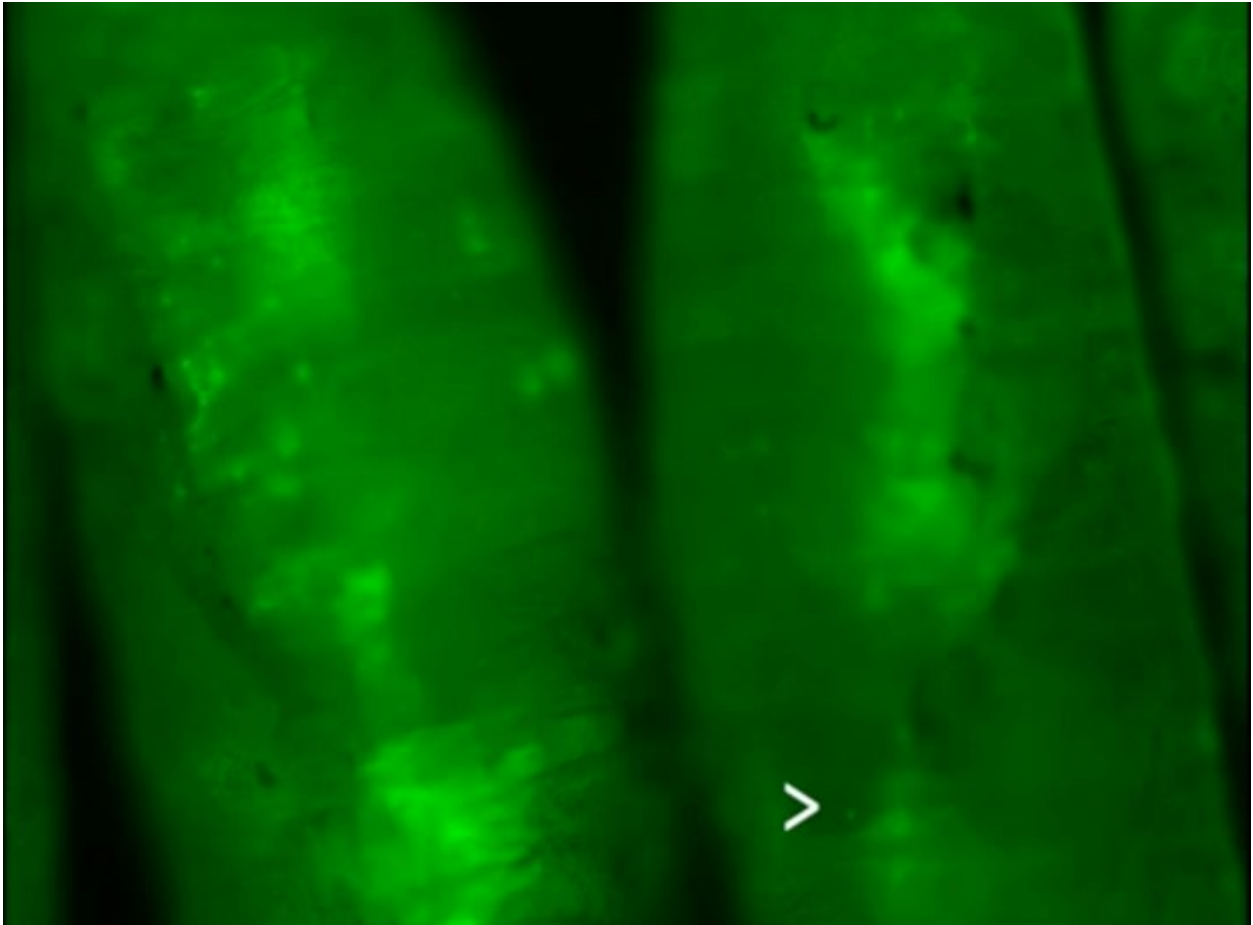


S2. Examples of cell overproliferation in the second tracheal metamere (Tr2) of *btl>Ras1,PTENi* L3 larvae. Related to Figure 3.

(A-C) *btl>Ras1,PTENi* resulted in massively over-proliferated transverse connective (TC) branches (A, C), often leading to merging of ASP and TC (B).

(D) ASP-like projections were also observed that were not associated with wing discs (no DAPI-positive [blue] wing discs). These likely derive from over-proliferating cells derived from the two known sources of imaginal tracheoblasts (IT; Guha and Kornberg 2005; Weaver and Krasnow, 2008, Denes et al., 2015).

Supplemental movie



Supplemental Movie: *btl>Ras1PTENi* cells migrating within the pupal abdomen. Related to Figure 3.

Time lapse movie visualizing migration of GFP-positive cells within the abdomen of *btl>Ras1,PTENi* pupae. Arrowheads highlight a subset of migrating cells.