Promoter proximal pausing limits tumorous growth induced by the Yki transcription factor in *Drosophila*

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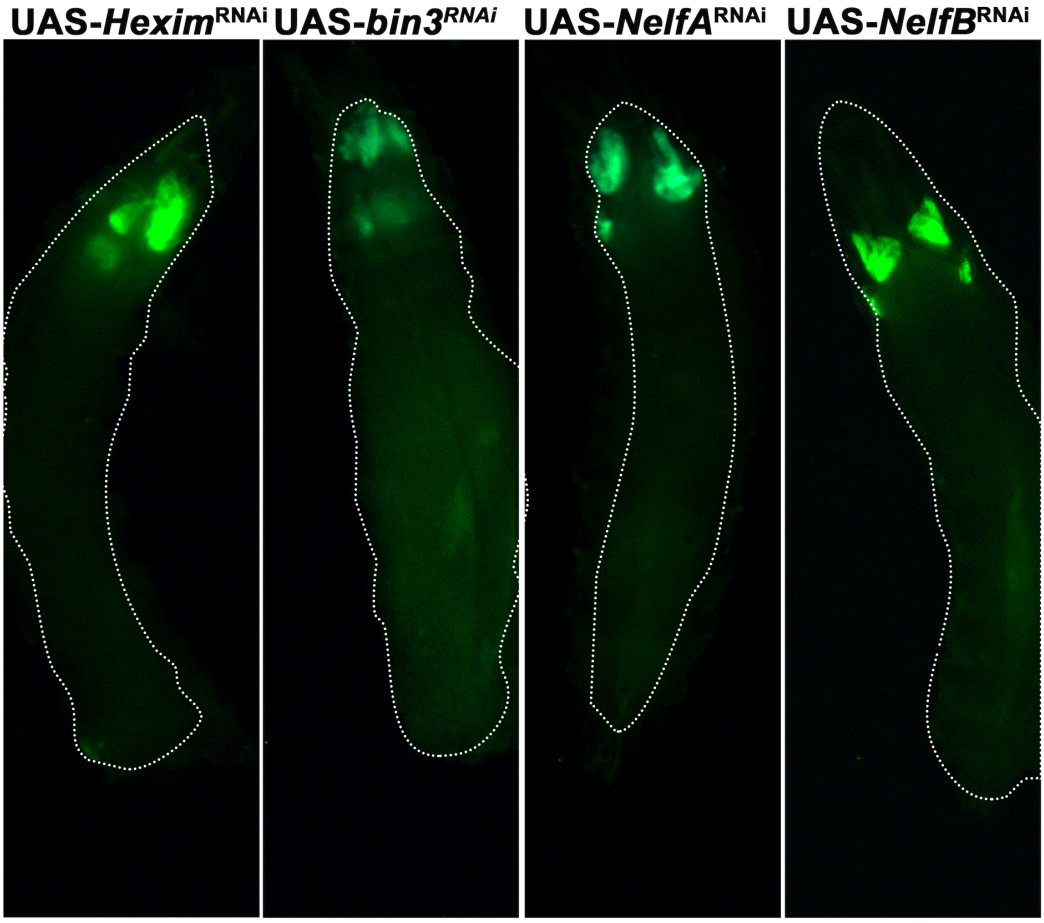
Supplementary Figures and Tables

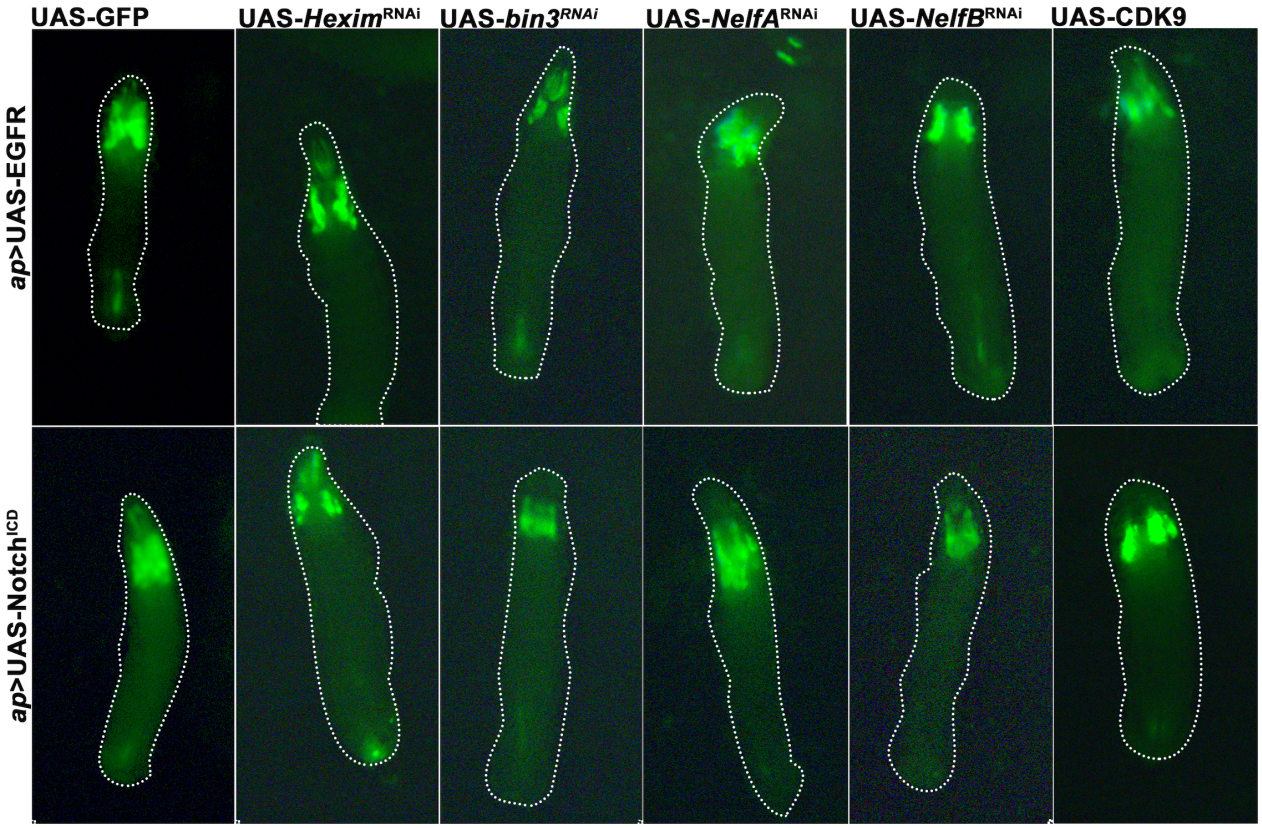
Suplementary Fig 1

The images show GFP-expressing wing discs of various genotypes as indicated. Down regulation of the any of the components of 7skRNP or NEFL complexes on their own do not cause any over-growth phenotype. All crosses were using GAL80^{TS}; *ap*-GAL4; UAS-GFP.



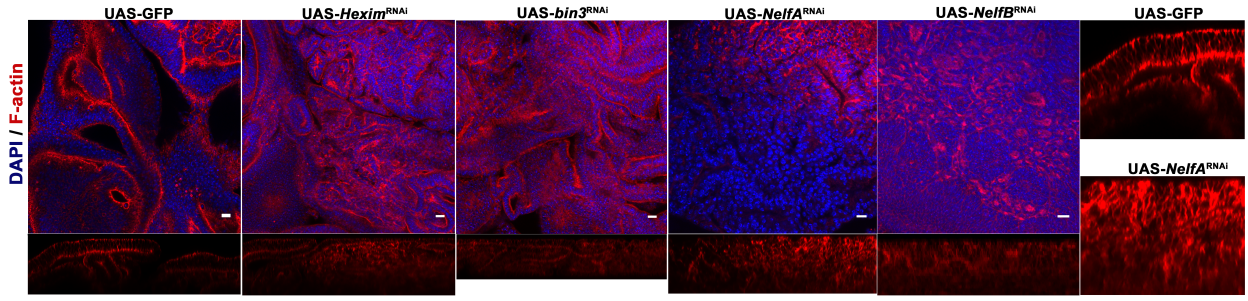
Suplementary Fig 2

The images show GFP-expressing larvae of various genotypes as indicated. Down regulation of the any of the components of 7SK snRNP or NELF or over-expression of CDK9 in combination with the over-expression of EGFR (Top panel) or the over-expression of Notch intracellual domain (Bottom panel) do not show overgrowth phenotypes (compare with the larvae over-expressing EGFR or Notch^{intra} alone). All crosses were using GAL80^{TS}; *ap*-GAL4; UAS-GFP.



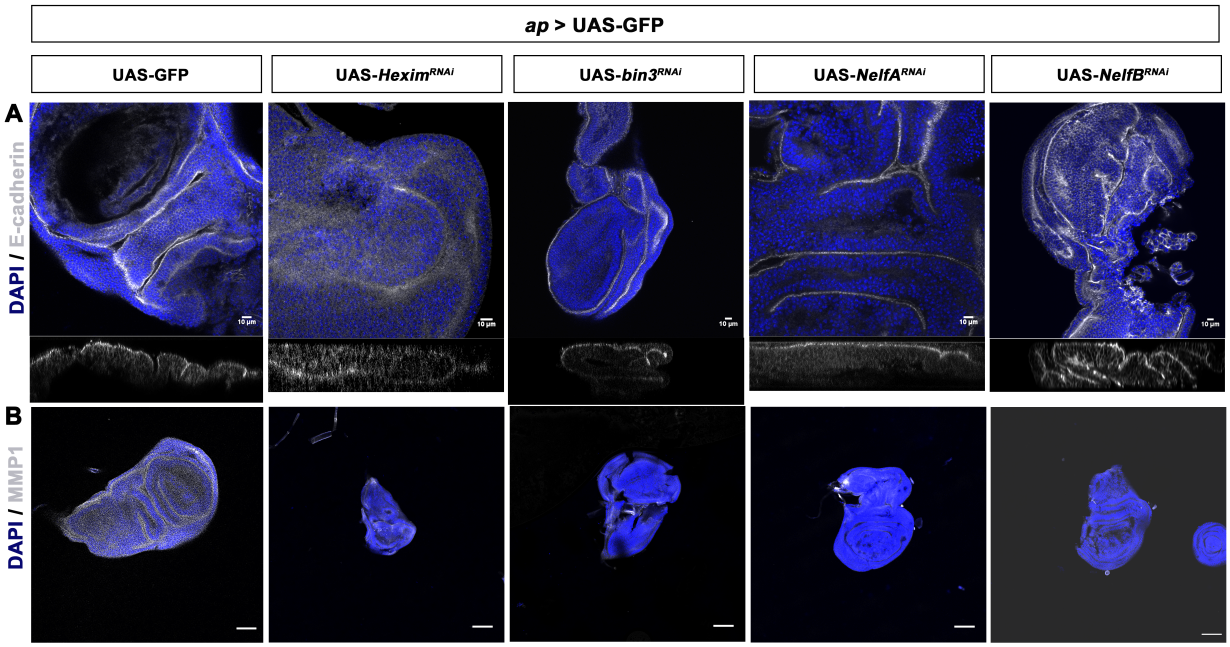
Suplementary Fig 3

Disruption of characteristic epithelial apico-basal polarity in tumor discs. Images of wing discs over-expressing Yki alone (crossed to UAS-GFP as control) or in combination RNAi-mediated knowckdown of *Hexim, bin3, NelfA* or *NelfB*. All crosses were using GAL80^{TS}; *ap*-GAL4; UAS-GFP. Discs are stained with Phalloidin (red), which reflects F-Actin expression and localization. Bottom panel of each image shows orthogonal optical section of respective genotype. Optical z-sections for control and one tumorous disc is shown on the right. Note delocalization of F-actin in tumorous tissues caused by the depletion of a component of PPP and Yki over-expression. All discs are also stained with DAPI (blue) to visualize nuclei.



Suplementary Fig 4 No neoplastic transformation is induced by the down regulation of components of PPP in wildtype background.

A. Images of wing discs expressing GFP alone or in combination with RNAi-mediated knowckdown of *Hexim, bin3, NelfA* or *NelfB* using GAL80^{TS}; *ap*-GAL4; UAS-GFP (scale bar = 10µm). Discs are stained for E-cad (white) expression and localization. All discs are also stained with DAPI (blue) to visualize nuclei. Bottom panel of each image shows orthogonal optical section of respective genotype. Note uniform sub-apical localization of E-cad in all discs.
B. Images of wing discs over-expressing GFP alone or in combination with RNAi-mediated knowckdown of *Hexim, bin3, NelfA* or *NelfB* using GAL80^{TS}; *ap*-GAL4; UAS-GFP (scale bar = 100µm). Wing discs are stained for MMP1 (white). All discs are also stained with DAPI (blue) to visualize nuclei. There is no detectable expression of MMP1 in these discs. All discs are imaged at lower magnification (10X) for better comparison with tumorous discs, which are shown in Fig. 2 of the main manuscript.



Suplementary Fig 5 Quantitative estimation of fold change in the mean intensity of MMP1 expression in various genetic backgrounds.

A. Quantitative estimation of fold change in the mean intensity of MMP1 staining in wing imaginal discs (images of corresponding discs are shown in Fig. 2B of the main manuscript) over-expressing Yki in combination RNAi-mediated knowckdown of *Hexim, bin3, NelfA* or *NelfB* using GAL80^{TS}; *ap*-GAL4; UAS-GFP and compared to wing discs over-expressing Yki alone. Mean intensity measurements are normalized with mean intesity of MMP1 in wing discs of GAL80^{TS}; *ap*-GAL4; UAS-GFP crossed to UAS GFP. Tumorous wing discs show significant increase in MMP1 staining intensity compared to non-tumorous wing discs
B. Quantitative estimation of fold change in mean intensity of MMP1 staining in wing imaginal discs of GAL80^{TS}; *ap*-GAL4; UAS-GFP crossed to UAS GFP and wing discs of RNAi-mediated knowckdown of *Hexim, bin3, NelfA* or *NelfB* using GAL80^{TS}; *ap*-GAL4; UAS-GFP (correspondng images of wing discs are shown in Suppl. Fig. S4B). Mean intensity measurements are normalized with mean intesity of MMP1 in wing discs of GAL80^{TS}; *ap*-GAL4; UAS-GFP crossed to UAS GFP. No significant change in MMP1 levels are observed in any of the wing discs

N=5 for each genotype in both A and B; **$p< 0.01$; *** $p< 0.001$; One way ANNOVA.

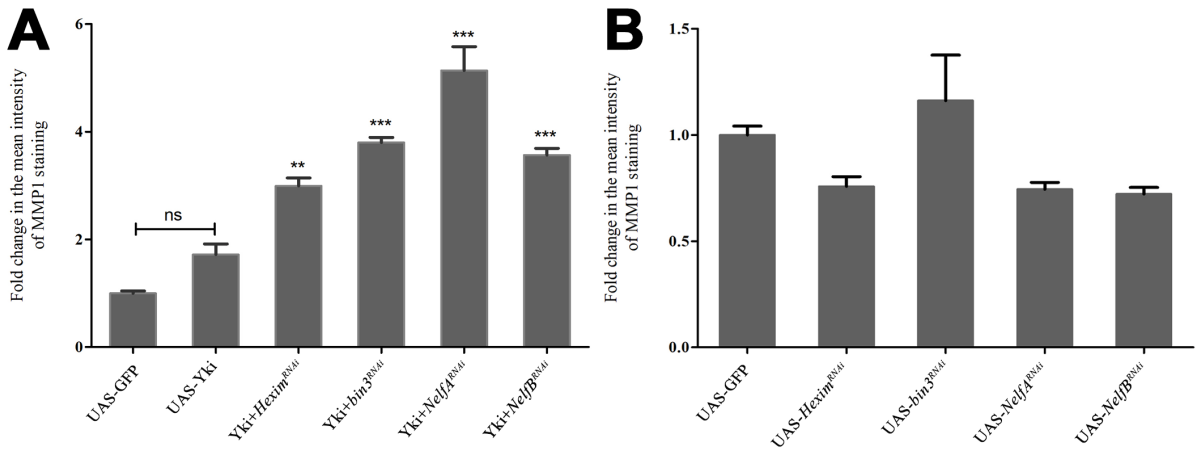


Table S1
List of direct targets of Yki whose transcripts are up/down-regulated in the wing discs of *ap*-GAL4/UAS-*NelfA*^{RNAi}; UAS-Yki

Upregulated in *ap*-GAL4/UAS-*NelfA*^{RNAi}; UAS-Yki		**Down-regulated in *ap*-GAL4/UAS- *NelfA*^{RNAi}; UAS-Yki**			
aru	***fru***	*Actn*	*CG6770*	*kek5*	*simj*
Bsg	*ft*	*Akap200*	*CG7065*	***klu***	*siz*
cbt	***ftz-f1***	***alt***	*CG7272*	***knrl***	*Sk2*
CG10075	***Gclc***	*Amun*	*CG7914*	*ko*	***smt3***
CG10462	***GlyP***	***Argk***	*CG8243*	*l(2)03659*	*sn*
CG10628	*GlyS*	*Atpalpha*	***CG8498***	***lama***	*sns*
CG10914	*l(3)02640*	***bchs***	***CG9331***	*LanB1*	*SppL*
CG11360	***Lasp***	*bowl*	***CG9650***	***LpR2***	***svp***
CG11658	***MFS17***	*brat*	*CG9663*	*Lrt*	*tio*
CG12065	*mTerf3*	***caup***	***chic***	***Lsp1alpha***	***tsh***
CG13185	*mthl1*	*CG10237*	***chm***	*mam*	*tup*
CG13398	*Mys45A*	***CG10960***	***CrebA***	*Mhcl*	*tutl*
CG13893	***neb***	*CG11050*	*cv-d*	*modSP*	*vri*
CG14322	*Nrg*	*CG12769*	*CycE*	*msi*	***wit***
CG1648	***nrv2***	***CG13875***	*Dh31-R*	*mtd*	
CG2247	***NTPase***	***CG15628***	*DOR*	*mth*	
CG2540	*Phb2*	*CG17684*	*dpr16*	*Mvl*	
CG2875	*Ptp10D*	*CG31475*	***drm***	*MYPT-75D*	
CG30069	*Ptp61F*	***CG32447***	***E(spl)m2-BFM***	*nrv1*	
CG31635	*qm*	*CG3270*	*Eip74EF*	*nub*	
CG32095	*rau*	*CG33129*	*Eip75B*	***opa***	
CG32365	*S6k*	*CG33229*	*emc*	*path*	
CG32369	***SNF4Agamma***	*CG3529*	***fax***	*Pep*	
CG33158	*Socs36E*	*CG4020*	*Fmr1*	*pk*	
CG3838	***spi***	*CG42272*	*Gale*	*Prosbeta7*	
CG5059	***Spt***	***CG42340***	*Gpdh*	*Rab5*	
CG6175	*Su(Tpl)*	***CG4374***	*grp*	***Reph***	
CG7841	*Tgt*	*CG4562*	*Gug*	*RnrS*	
CG8360	***Traf4***	*CG4615*	***hbs***	*Rtnl1*	
CG9932	*Tsp39D*	*CG5001*	*Hnf4*	*rump*	
dbe	***uif***	*CG5756*	***Hr39***	***Sb***	
dpp	*Vha26*	***CG5758***	*Hs6st*	*sbb*	
ex	***wg***	***CG5885***	*Hsp26*	*SC35*	
fng	***zormin***	*CG6163*	***Hsp83***	*Sec63*	
for		***CG6287***	*kek1*	*side*	

The genes shown in bold letters are those direct targets of Yki that are upregulated in both non-tumorous *ap*>UAS-Yki discs and tumorous *ap*>UAS-Yki; UAS-*NelfA*^{RNAi} discs, but degree of enhancement was higher in tumorous tissue.