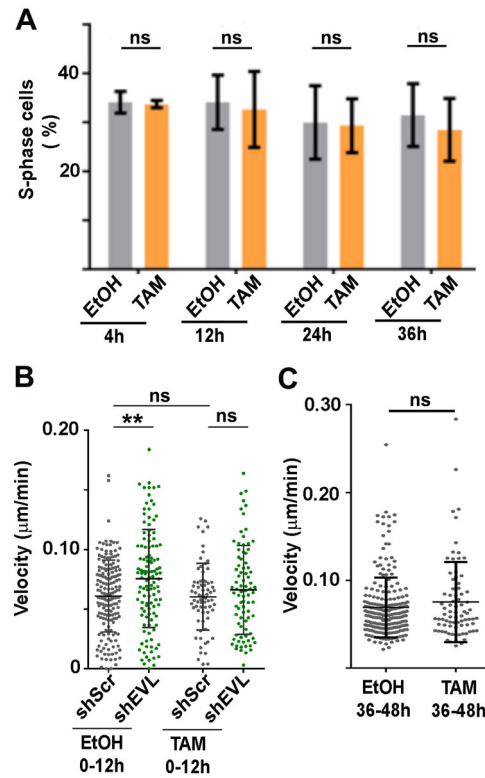
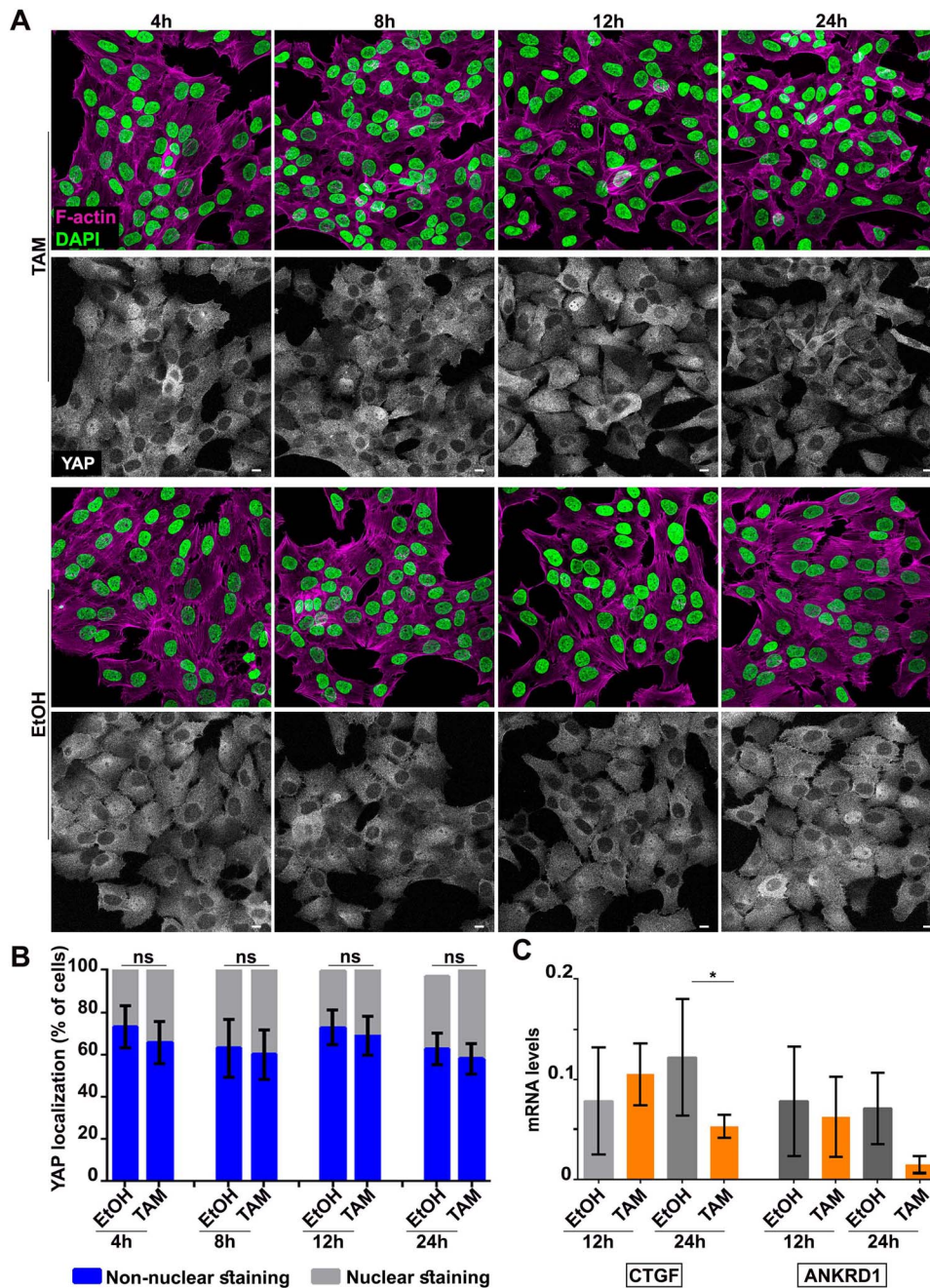


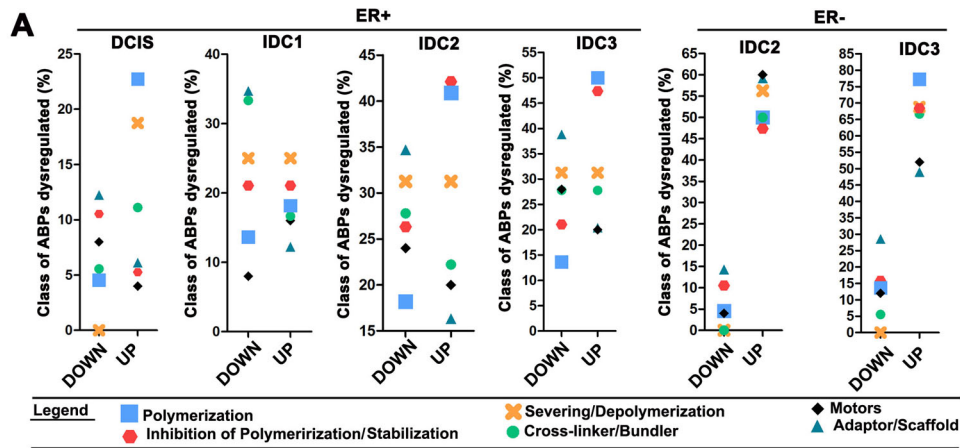
Supplementary Figure 1: Effects of TAM or EtOH on PBabe or ER-Src cells. (A) Standard confocal sections of ER-Src cells treated with EtOH for 4, 12, 24 and 36 hours, stained with Phalloidin (magenta), which marks F-actin, anti-pSrc (Cyan) and DAPI (blue). Scale bars represent 10 μm . **(B)** Images by phase contrast microscopy of PBabe cells, treated with EtOH or TAM for 4, 12, 24 and 36 hours. Scale bars represent 50 μm . **(C)** Western blot on protein extracts from untreated PBabe cells (GM) or PBabe cells treated with TAM for 4, 12, 24 or 36 hours, blotted with anti-pSrc or anti-GAPDH. The asterisk indicates non-specific bands revealed by the anti-pSrc antibody. **(D)** Standard confocal sections of PBabe cells 12 hours after EtOH or TAM treatments, stained with Phalloidin (magenta), which marks F-actin and DAPI (blue). Scale bars represent 10 μm . **(E)** Apparent Young's modulus of PBabe cells 12 hours after EtOH or TAM treatments. The data points represented as dots with horizontal line indicating median values are from one biological replicate. ns indicates non-significant. Statistical significance was calculated using simple Mann Whitney tests. Related to Figures 1 and 2.



Supplementary Figure 2: Effects of Src on proliferation and migration. (A) Percentage of ER-Src cells in S-phase after treatment with EtOH (gray bars) or TAM (orange bars) for 4, 12, 24 or 36 hours in the presence of EGF. **(B)** Quantification of cell velocity during the first 12 hours of EtOH or TAM treatments in ER-Src cells expressing *shScr* (gray dots) or *shEVL* (green dots). **(C)** Quantification of ER-Src cell velocities between 36 and 48 hours of EtOH or TAM treatments. All quantifications were from three biological replicates. Error bars indicate SD. ns indicate non-significant. ** indicates $P < 0.001$. Statistical significance was calculated using **(A)** two-way ANOVA or **(B)** one-way ANOVA or **(C)** impaired t-test. Related to Figure 1.



Supplementary Figure 3: YAP/TAZ activity in TAM-treated ER-Src cells. (A) Standard confocal sections of ER-Src cells treated with EtOH or TAM for 4, 8, 12, or 24 hours, stained with Phalloidin (magenta) to mark F-actin, anti-p-YAP (Gray) and DAPI (Blue). Scale bars represent 10 μ m. (B) Quantifications from two biological replicates of the percentage (%) of ER-Src cells in which YAP/TAZ is restricted to the cytoplasm (blue) or localizes to the nucleus (gray) after treatment with EtOH or TAM for 4, 8, 12 and 24 hours (C) Quantifications from three biological replicates of *CTGF* or *ANKRD1* mRNA levels measured by qRT-PCR on extracts from ER-Src cells treated with EtOH (gray bars) or TAM (orange bars) for 12 or 24 hours. Error bars indicate SD. ns indicates non-significant. * indicates $P < 0.05$. Statistical significance was calculated using one-way ANOVA tests. Related to Figure 1.

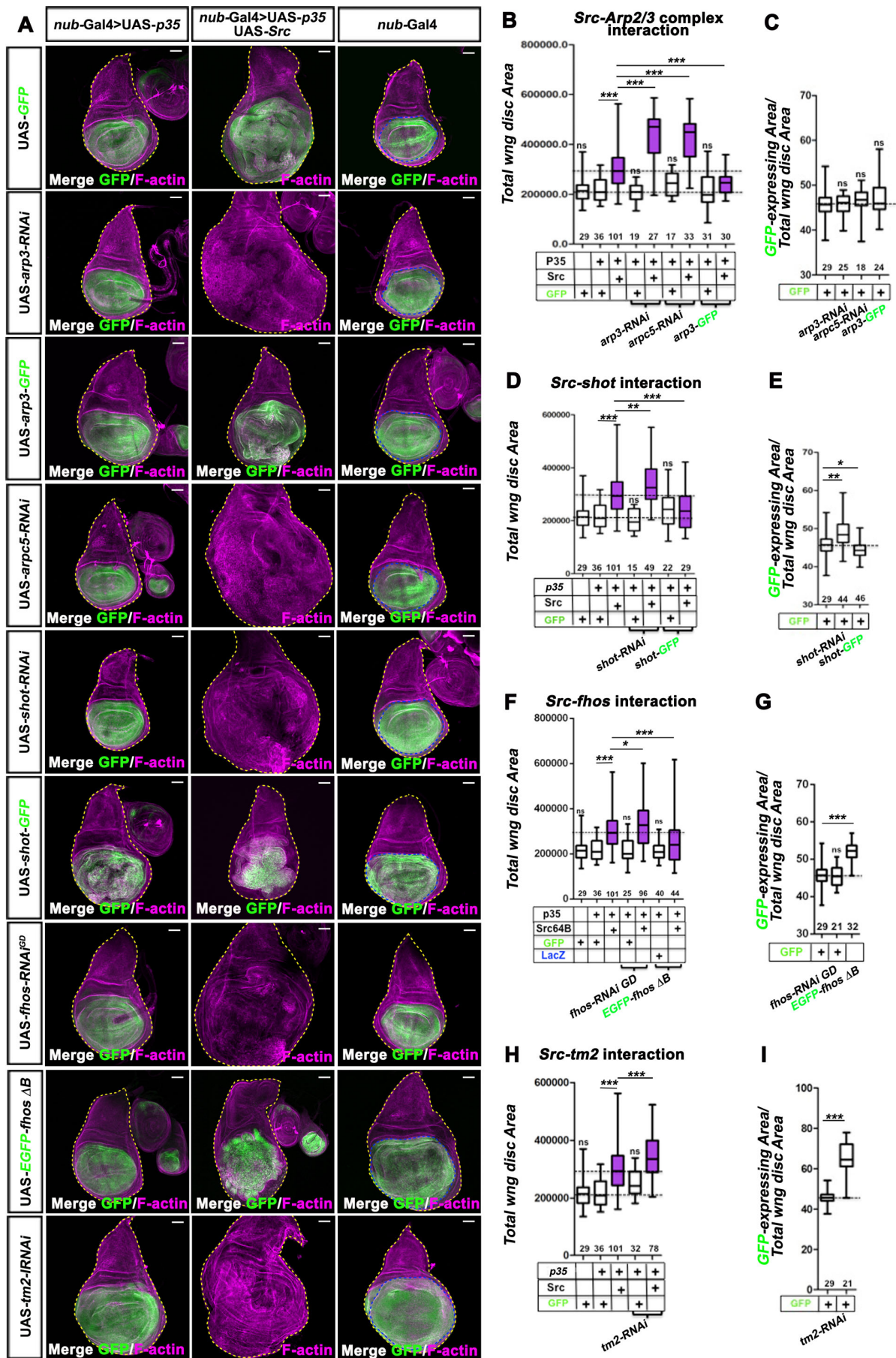


B

	Total	Down	Up
Polymerization	6	1	5
Inhibition of polymerization/ Stabilization	3	2	1
Filament severing/Depolymerization	3	0	3
Cross-linkers/bundlers	3	1	2
Motors	2	2	1
Adapter/ Scaffold	9	6	3

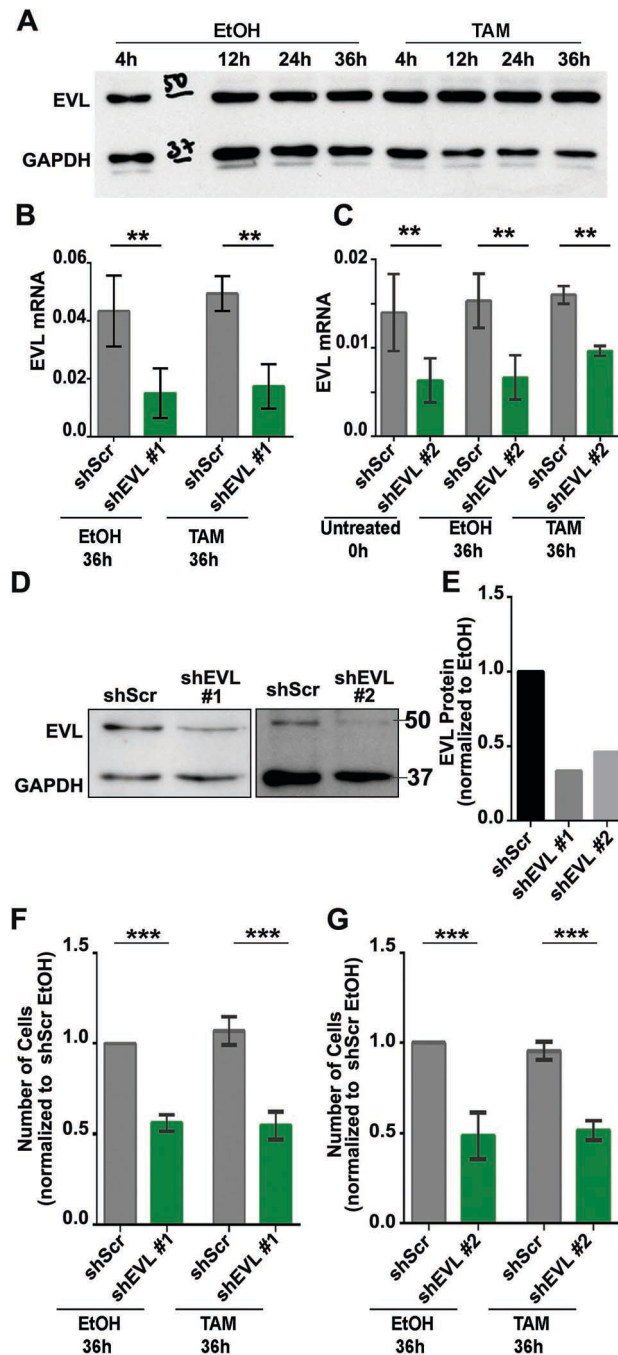
FHOD3
 ARPC5L; ARPC3; ACTR3; ENAH; EVL
 TPM2; TMOD1
 CAPZA1
 MICAL2; CFL1; GMFB
 TAGLN
 MARCKSL1; PLS1
 MYO9A; MYH3
 MYO6
 MYLK; PARVA; DST; SNTB2; CORO2B; SNTA1
 LASP1; SHROOM2; EZR

Supplementary Figure 4: Classes of ABPs deregulated in breast lesions. (A) Percentage of functional classes of ABPs up- or down-regulated in ER⁺ DCIS, ER⁺ IDC1, ER⁺ IDC2, ER⁺ IDC3, ER⁻ IDC2 and ER⁻ IDC3 in comparison with normal breast tissues. ABPs were classified using the gene-ontology vocabulary as Polymerization (Blue, Square); Inhibition of polymerization/Stabilization (Red, Hexagon); Filament severing/Depolymerization (Yellow, Cross); Cross-linkers/ bundlers (Green, Dot); Motors (Black, Lozenge) and Adapter/Scaffold (Dark Blue, Triangle), according to their effect on actin (See also Supplementary Data 2). **(B)** Classification of the 27 ABPs misregulated in ER⁺ DCIS, according to their functional role on actin and their expression levels (down- or upregulated) compared to normal breast tissues. Related to Figure 2.

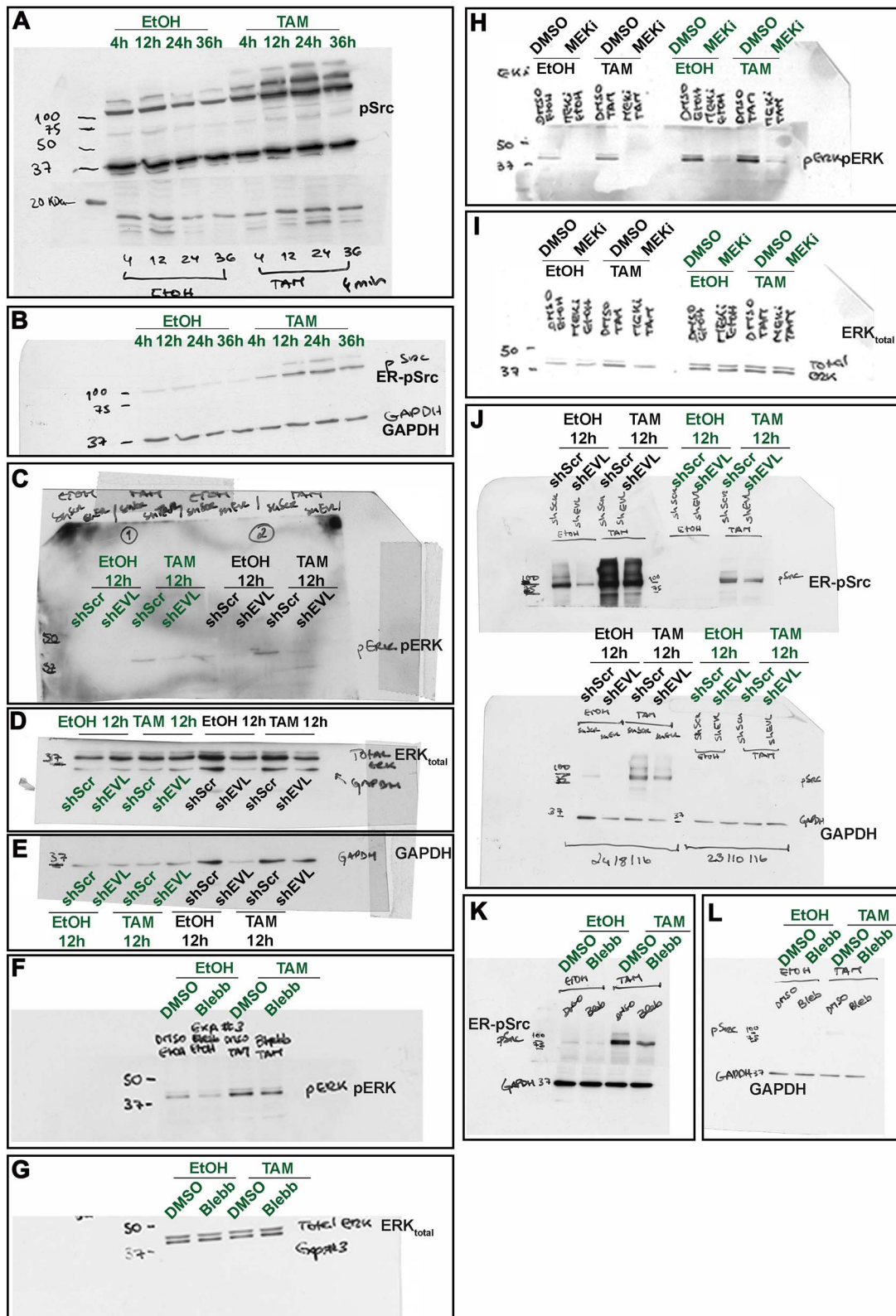


Supplementary Figure 5: Effect of *Drosophila* ABPs regulated by Src on tissue growth. (A) Standard confocal sections of third instar wing imaginal discs with dorsal side up, stained with Phalloidin to mark F-actin (magenta). *nub-Gal4; UAS-p35* or *nub-Gal4; UAS-p35, Src64B^{UY133}* or *nub-Gal4* discs, carrying UAS-*mCD8-GFP* (green) or UAS-*arp3-RNAi*

or UAS-*arp3-GFP* (green) or UAS-*arpc5-RNAi* or UAS-*shot-RNAi* or UAS-*shot-GFP* (green) or UAS-*fhos-RNAi* or UAS-*EGFP-fhos Δ B* (green) or UAS-*tm2-RNAi*. The yellow dashed lines outline the whole wing disc area. The blue lines outline the *nub>GFP*-expressing domain. Scale bars represent 30 μ m. **(B-I)** Quantifications of total wing disc area **(B,D,F,H)** or ratio of the *nub>GFP* area over the total wing disc area **(C,E,G,I)** in discs in which Arp2/3 components **(B,C)**, or *shot* **(D,E)** or *fhos* **(F,G)** or *tm2* **(H,I)** expression are affected. Genotypes are indicated below each column. Magenta bars are *nub>Src/p35*-expressing wing discs in which the expression of Arp2/3 components or *shot* or *fhos* or *tm2* is affected. Numbers of samples from two biological replicates are indicated on the top of the X axis. ns indicates non-significant. Significance on the top of each column is for comparison with *nub>GFP*, *P35* **(B,D,F,H)** or *nub>GFP* **(C,E,G,I)**. Error bars indicate SD. *** indicates $P < 0.0001$. ** indicates $P < 0.005$. * indicates $P < 0.05$. Statistical significance was calculated using impaired t-test. Related to Figure 4.



Supplementary Figure 6: EVL knocked down reduces cell growth. (A) Western blots on protein extracts from ER-Src cells treated with EtOH or TAM for 4, 12, 24 or 36 hours, blotted with anti-EVL and anti-GAPDH. (B) Quantifications from three biological replicates of *EVL* mRNA levels in extracts from ER-Src cells transfected with *shScr* (gray) or *shEVL#1* (green), and treated with EtOH or TAM for 36 hours. (C) Quantifications from three biological replicates of *EVL* mRNA levels on extracts from ER-Src cells expressing *shScr* (gray) or *shEVL#2* (green), untreated or treated with EtOH or TAM for 36 hours. (D) Western blot on protein extracts from ER-Src cells treated with *shScr* or *shEVL#1*, or *shEVL#2*, blotted with anti-EVL and anti-GAPDH. (E) Quantification from one biological replicate of total EVL levels in ER-Src cells treated with *shScr* (black bar) or *shEVL#1* (dark gray bar) or *shEVL#2* (light gray bar), normalized to GAPDH. (F) Quantifications from three biological replicates of the number of ER-Src cells expressing *shScr* (gray bars) or *shEVL#1* (green bars), treated with EtOH or TAM for 36 hours, normalized to EtOH-treated ER-Src cells expressing *shScr*. (G) Quantifications from three biological replicates of the number of ER-Src cells expressing *shScr* (gray bars) or *shEVL#2* (green bars), treated with EtOH or TAM for 36 hours, normalized to EtOH-treated ER-Src cells expressing *shScr*. Error bars indicate SD. ** indicates $P < 0.01$. *** indicates $P < 0.0001$. Statistical significance was calculated using one-way ANOVA tests. Related to Figure 5.



Supplementary Figure 7: Original images of the most relevant Western blots. (A) Western blot on protein extracts from ER-Src cells treated with EtOH or TAM for 4, 12, 24 or 36 hours, blotted with anti-pSrc, which reveals endogenous pSrc and ER-pSrc and anti-GAPDH. (B) lower exposure of the blot shown in A. See also Figure 1A. (C-E) Western blots on protein extracts from ER-Src cells expressing *shScr* or *shEVL#2* and treated with EtOH or TAM for 12 hours, blotted with (C) anti-pERK or (D) anti-ERK or (E) anti-GAPDH. See also Figure 5D. (F-G) Western blots on protein extracts from ER-Src cells treated with EtOH or TAM and DMSO or Blebbistatin for 12 hours, blotted with (F) anti-pERK or (G) anti-ERK. See also Figure 7B. (H-I) Western blots on protein extracts from ER-Src cells treated with DMSO or MEKi and EtOH or TAM for 12 hours, blotted with (H)

anti-pERK or **(I)** anti-ERK, which reveals total ERK. See also Figure 1G. **(J)** Western blots on protein extracts from ER-Src cells expressing *shScr* or *shEVL#2* and treated with EtOH or TAM for 12 hours, blotted with anti-pSrc or anti-pSrc and anti-GAPDH. See also Figure 5L. **(K-L)** Western blots on protein extracts from ER-Src cells treated with EtOH or TAM and DMSO and Blebbistatin for 12 hours, blotted with **(K)** anti-pSrc to reveal ER-pSrc or **(L)** anti-GAPDH. See also Figure 7F. Blots included in main figures are labeled in green.

Series	Platform	Samples											Reference	
		N	ADH	DCIS	IDC									
					ER status		Grade division (IDC,ER+)			Grade division (IDC,ER-)				
					ER +	ER -	I	II	III	I	II	III		
GSE15852	HG-U133A	43	0	0	22	17	4	10	8	3	10	4	1	
GSE16873	HG-U133A	12	12	12									2	
GSE7390	HG-U133A	0	0	0	102	56	21	54	27		13	43	3	
GSE20194	HG-U133A	0	0	0	115	93	7	64	44		13	80	4, 5	
GSE10810	HG-U133A PLUS 2.0	27	0	0	11	11	2	7	2		2	9	6	
GSE21422	HG-U133A PLUS 2.0	5	0	9									7	
GSE22544	HG-U133A PLUS 2.0	4	0	0									8	
GSE23593	HG-U133A PLUS 2.0	0	0	0	29	12	2	17	10			12	9	
GSE5460	HG-U133A PLUS 2.0	0	0	0	44	50	15	11	18		5	45	10	
GSE5764	HG-U133A PLUS 2.0	10	0	0									11	
GSE10780	HG-U133A PLUS 2.0	143											12	
GSE2109	HG-U133A PLUS 2.0	0	0	1	18	18	3	11	4	1	2	15	-	
GSE3744	HG-U133A PLUS 2.0	7	0	0	15	24	0	0	15			24	13, 14	
GSE17907	HG-U133A PLUS 2.0	4	0	0	13	20	1	4	8		3	17	15	
GSE19615	HG-U133A PLUS 2.0	0	0	0	45	42	13	11	21		6	36	16	
GSE23177	HG-U133A PLUS 2.0	0	0	0	116		0	0	116				17	
Total		255	12	22	530	343	68	189	273					

Supplementary Table 1: Microarray platforms used in this study. The number of samples, their histological classification and associated references are indicated for each platform. N-normal tissue; ADH – atypical ductal hyperplasia; DCIS – ductal carcinoma *in Situ*; IDC – invasive ductal carcinoma of grade 1 (I) or grade 2 (II) or grade 3 (III). ER: Estrogen receptor. Related to Figure 3.

		Tissue growth (fold changes)																
		Src+; p35+		wt														
		<i>GFP</i>	1		1													
<table border="1"> <thead> <tr> <th>Human</th> <th><i>Drosophila</i></th> </tr> </thead> <tbody> <tr> <td> <div style="background-color: orange; width: 10px; height: 10px; display: inline-block;"></div> EVL <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> ENAH <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> VASP </td> <td><i>enabled (ena)</i></td> </tr> <tr> <td> <div style="background-color: orange; width: 10px; height: 10px; display: inline-block;"></div> ACTR3 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> ACTR3B </td> <td><i>Actin-related protein 3 (Arp3)</i></td> </tr> <tr> <td> <div style="background-color: orange; width: 10px; height: 10px; display: inline-block;"></div> ARPC5L <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> ARPC5 </td> <td><i>Actin-related protein 2/3 complex, subunit 5 (Arpc5)</i></td> </tr> <tr> <td> <div style="background-color: blue; width: 10px; height: 10px; display: inline-block;"></div> DST <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> MACF1 </td> <td><i>short stop (shot)</i></td> </tr> <tr> <td> <div style="background-color: blue; width: 10px; height: 10px; display: inline-block;"></div> FHOD3 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> FHOD1 </td> <td><i>formin homology 2 domain containing (fhos)</i></td> </tr> <tr> <td> <div style="background-color: blue; width: 10px; height: 10px; display: inline-block;"></div> TPM2 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM3 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM4 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM1 </td> <td><i>tropomyosin 2 (tm2)</i></td> </tr> </tbody> </table>	Human	<i>Drosophila</i>	<div style="background-color: orange; width: 10px; height: 10px; display: inline-block;"></div> EVL <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> ENAH <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> VASP	<i>enabled (ena)</i>	<div style="background-color: orange; width: 10px; height: 10px; display: inline-block;"></div> ACTR3 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> ACTR3B	<i>Actin-related protein 3 (Arp3)</i>	<div style="background-color: orange; width: 10px; height: 10px; display: inline-block;"></div> ARPC5L <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> ARPC5	<i>Actin-related protein 2/3 complex, subunit 5 (Arpc5)</i>	<div style="background-color: blue; width: 10px; height: 10px; display: inline-block;"></div> DST <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> MACF1	<i>short stop (shot)</i>	<div style="background-color: blue; width: 10px; height: 10px; display: inline-block;"></div> FHOD3 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> FHOD1	<i>formin homology 2 domain containing (fhos)</i>	<div style="background-color: blue; width: 10px; height: 10px; display: inline-block;"></div> TPM2 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM3 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM4 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM1	<i>tropomyosin 2 (tm2)</i>	<i>ena</i>	RNAi	0.75 p<0.0001	1.03
	Human	<i>Drosophila</i>																
<div style="background-color: orange; width: 10px; height: 10px; display: inline-block;"></div> EVL <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> ENAH <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> VASP	<i>enabled (ena)</i>																	
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<div style="background-color: blue; width: 10px; height: 10px; display: inline-block;"></div> TPM2 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM3 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM4 <div style="background-color: white; width: 10px; height: 10px; display: inline-block;"></div> TPM1	<i>tropomyosin 2 (tm2)</i>																	
		Over.	1.20 p<0.0001	1.06 p<0.0009														
		<i>Arp3</i>	RNAi	1.47 p<0.0001	1													
			Over.	0.82 p<0.0004	1.03													
		<i>Arpc5</i>	RNAi	1.40 p<0.0001	1.03													
		<i>shot</i>	RNAi	1.12 p<0.005	1.06 p<0.001													
			Over.	0.80 p<0.0002	0.96 p<0.03													
		<i>fhos</i>	RNAi	1.08 p<0.03	0.99													
		<i>tm2</i>	RNAi	1.18 p<0.0001	1.41 p<0.0009													

Supplementary Table 2: Effect of ABPs deregulated by Src on tissue growth in the fly. (A) Human ABPs deregulated in TAM-treated ER-Src cells and in ADH/DCIS with their family members and their corresponding *Drosophila* counterpart. Orange and blue indicate genes up- and downregulated respectively. (B) Effect of knocking down (RNAi) or overexpressing (Over.) *ena*, *Arp3*, *Arpc5*, *shot*, *fhos* and *tm2* on growth of wing imaginal discs in which *nub*-Gal4 drives *Src64B^{UY1332}* and UAS-*p35* or UAS-*mCD8::GFP* (wt). Values represent fold changes relative to (first column) *nub>GFP*, *Src64BUY1332*, *p35* or (second column) *nub>GFP*. Blue and yellow indicate a significant suppression or enhancement relative to control, respectively. Related to Figure 4.

		EVL Classification - IDC		p-Value
		Count (%)		
		Neg	Pos	
Inflammation	0	10 (22.7)	5 (15.6)	0.037
	1	22 (50.0)	25 (78.1)	
	2	11 (25.0)	1 (3.1)	
	3	1 (2.3)	1 (3.1)	
	Total	44 (100)	32 (100)	
ER	Neg	29 (36.3)	6 (10.9)	0.037
	Pos	51 (63.8)	49 (89.1)	
	Total	80 (100)	55 (100)	
HER2	Neg	63 (78.8)	46 (83.6)	0.001
	Pos	17 (21.3)	9 (16.4)	
	Total	80 (100)	55 (100)	
CK5	Neg	75 (93.8)	54 (98.2)	0.479
	Pos	5 (6.3)	1 (1.8)	
	Total	80 (100)	55 (100)	
P-Cad	Neg	55 (68.8)	52 (94.5)	0.0002
	Pos	25 (31.3)	3 (5.5)	
	Total	80 (100)	55 (100)	
Subtype	Luminal A	50 (62.5)	46 (83.6)	0.008
	Luminal B	4 (5.0)	5 (9.1)	
	HER2 - OE	13 (16.3)	4 (7.3)	
	Basal	10 (12.5)	0 (0.0)	
	Ind.	3 (3.8)	0 (0.0)	
	Total	80 (100)	55 (100)	
Grade	Low	28 (35.0)	27 (49.1)	0.020
	Inter	22 (50.0)	25 (78.1)	
	High	11 (25.0)	9 (16.4)	
	Total	80 (100)	55 (100)	

Supplementary Table 3: Association of EVL expression in IDC. Association of EVL expression with clinicopathological features, including oestrogen receptor (ER) status, basal markers (CK5 and P-cad) expression, and breast carcinomas molecular subtypes in IDC. Related to Figure 8 and Table 1.

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