Quantifying Tip60 (Kat5) stratifies breast cancer.

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Supplemental figures and legends.

Supplemental Figure 1. A. Tip60 IHC staining in Control cell line (MCF10A) and indicated Breast Cancer Cell lines. Left, full size image; Right, 5x magnification. Blue: DNA counter stain, Orange/Brown: Tip60 staining. **B.** IHC Negative control. Staining performed using Tip60 antibody PAB18305 (Abnova).

Supplemental Figure 2. Additional examples of Tip60 IHC staining in breast tumour samples (Supplementary to Figure 2B). Image Pairs: Top image: full stained TMA. Bottom image: 12x magnification of tumour. Blue: DNA stain, Orange/Brown: Tip60 staining (Tip60 antibody K-17: sc-5727, Santa Cruz).

Supplemental Figure 3. Tip60 staining of invasive breast cancer tumour with second Tip60 antibody (Abnova). Blue: DNA stain, Orange/Brown: Tip60 staining. Staining performed using Tip60 antibody PAB18305 (Abnova).

Supplemental Figure 4. A. Tip60 staining patterns observed in TMA (percent). **B.** Tip60 staining patterns by percent of Luminal A cohort.

Supplemental Figure 5. A. Tip60 staining patterns observed, by tumour grade. **B.** Tip60 staining patterns observed, by Union for International Cancer Control (UICC) tumour stage.

Supplemental Figure 6. A. Tip60 staining patterns observed by ER positivity. **B.** Percent of cytoplasmic only Tip60 staining, ER positivity.

Supplemental Figure 7. A. Tip60 staining patterns observed by PR positivity. **B.** Nuclear Tip60 staining positivity by PR status.

Supplemental Figure 8. A. Nuclear Tip60 staining positivity by menopausal status.

Supplemental Figure 9. A. Univariate complete-case Cox proportional hazard (CPH) regression modeling association with: **A.** Subtype (n=227). **B.** Tumour size. **C.** Stage (n=296). **D.** Age.

Supplemental Figure 10. A. Complete-case CPH modeling (n=185) of OS and Cyto-Only percentage. **B.** Imputed-data CPH modeling for OS using Subtype, Stage and Age (n=334). **C.** Complete-case CPH modeling for DFS (n=174) using Cyto-Only percentage, Total Cyto percentage, Nuc, Subtype, NPI, Stage, and Menopause status explain variability in DFS outcome.

Supplemental Figure 11. A. Allelic changes observed (change, % and n indicated) in Kat5 gene in breast cancer cases. Using cBioPortal with individual Breast cancer studies indicated. **B.** Tip60 mutations observed in breast cancer, in cBioPortal.

MCF10A

Α

х5







MCF7











Negative Control 2^{ndry} antibody alone

В











Tip60 Staining pattern- majority of cells in section





Majority staining pattern by Tumour Grade





Majority staining pattern by ER status









S7



Nuclear staining pattern by menopausal status





Observations 334

*p<0.1; **p<0.05; ***p<0.01 Note:

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Variable		N	Hazard ratio		р
CytoOnly.Pct		185		0.99 (0.99, 1.00)	0.005
Stage	I	41		Reference	
	II	93		1.86 (0.70, 4.93)	0.213
	Ш	41	⊢∎⊣	5.97 (2.26, 15.80)	<0.001
	IV	10	⊢ ∎1	12.59 (3.95, 40.14)	<0.001

В

Variable	•	Ν	Hazard ratio		р
Subtype	e Luminal A	179	- -	Reference	
	Luminal B	40	i ¦∎ ⊣	1.46 (0.82, 2.58)	0.2
	Her 2	31		1.49 (0.86, 2.58)	0.2
	Basal	84	⊢∎⊣	4.45 (2.86, 6.94)	<0.001
Stage	1	71		Reference	
	П	161	i∔∎-i	1.36 (0.76, 2.44)	0.3
	Ш	80	⊢∎→	3.47 (1.89, 6.36)	<0.001
	IV	22	⊢∎⊸	7.61 (3.63, 15.94)	<0.001
Age		334		1.04 (1.02, 1.05)	<0.001

С

Variable		Ν	Hazard ratio		р
CytoOnly.Pct		174		0.97 (0.95, 0.99)	< 0.001
TotalCyto.Pct		174		1.03 (1.01, 1.05)	0.003
NucStainingInd	d Nuc-	145		Reference	
	Nuc+	29		0.21 (0.05, 0.85)	0.028
Subtype	Luminal A	98		Reference	
	Luminal E	3 22	- - -	0.49 (0.19, 1.28)	0.146
	Her 2	13		0.84 (0.33, 2.14)	0.713
	Basal	41	- 	1.91 (1.04, 3.52)	0.038
NPI	Index 1	5	•	Reference	
	Index 2	26		0.68 (0.08, 5.79)	0.721
	Index 3	87		0.70 (0.08, 6.11)	0.748
	Index 4	56		1.52 (0.16, 14.44)	0.717
Tumour.Size		174		0.99 (0.97, 1.01)	0.394
Stage	1	41		Reference	
	11	92		1.37 (0.54, 3.44)	0.507
	III	41		2.74 (0.81, 9.26)	0.105
Age		174		0.99 (0.96, 1.03)	0.721
Menopause	Pre,Peri	66	, in the second se	Reference	
	Post	108		1.56 (0.63, 3.83)	0.333

Α Amplification 17.24% n=5 (of 117) 15% Mutation Amplification





3 Mutations (page 1 of 1)					0 0	Columns -		
Study	Sample ID	Protein Change	Annotation V	Mutation Type	Copy #	COSMIC	Allele Freq (T)	# Mut in Sample
The Metastatic B	MBC-MBCProject_5	S313F		Missense	Gain			694
Breast Invasive	TCGA-A2-A0D1-01	R220Q		Missense	Diploid		0.21	58
Breast Invasive	TCGA-BH-A18P-01	X442_splice		Splice	ShallowDel		0.38	271

Deep Deletion



Tip60



β-Tubulin

