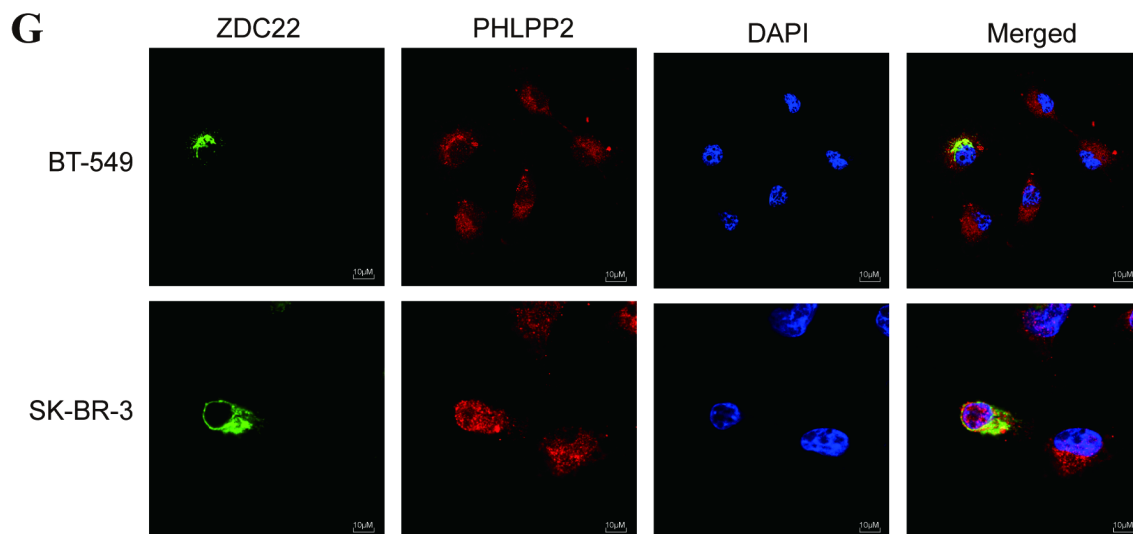
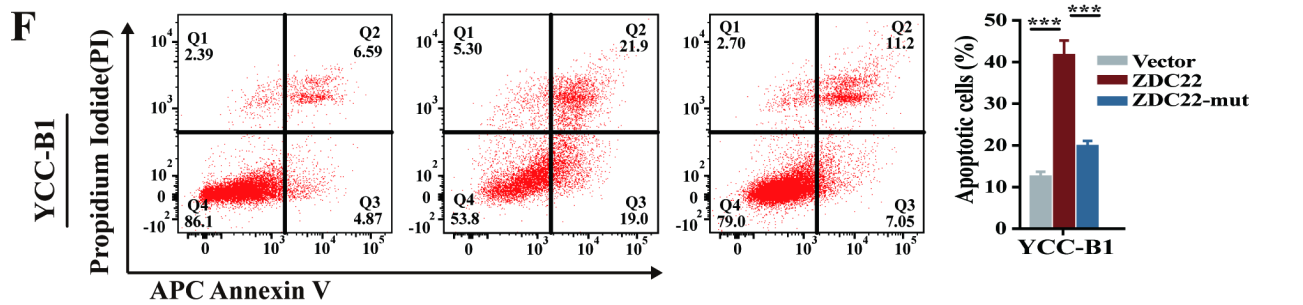
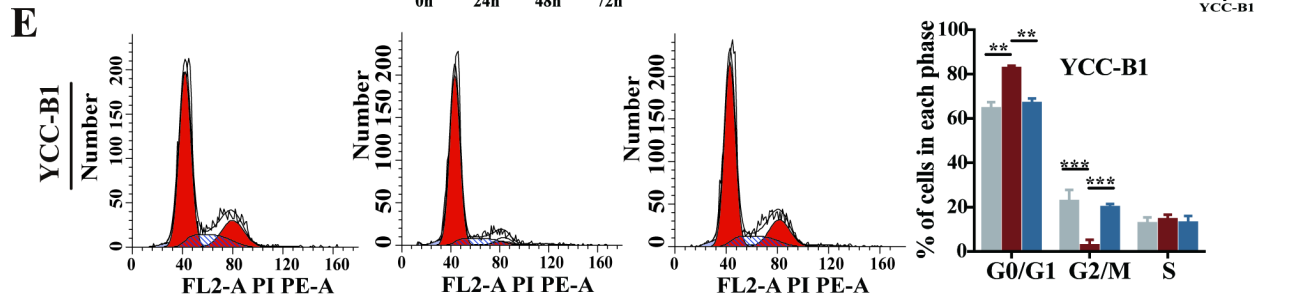
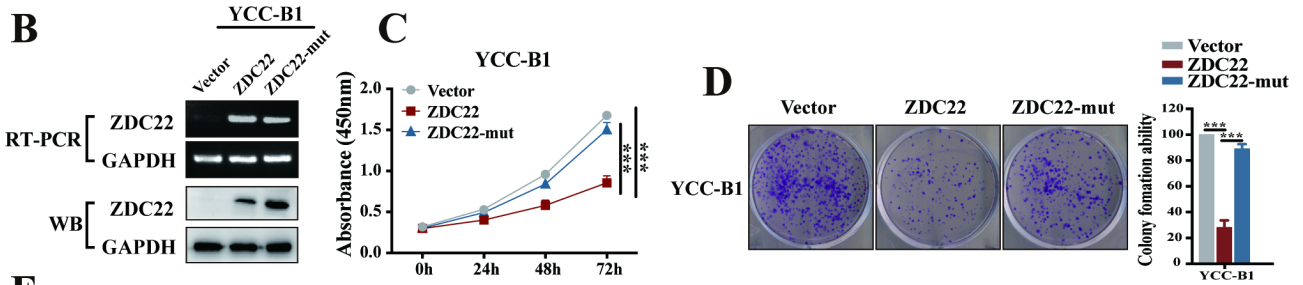
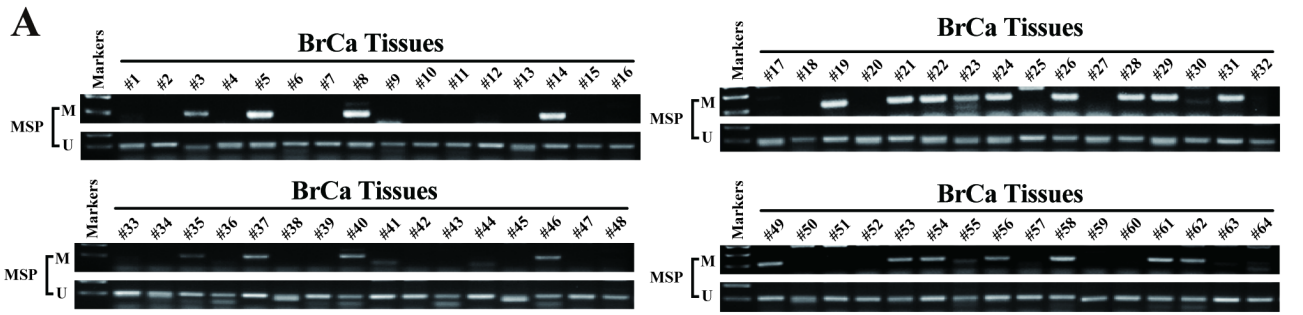


1 **SUPPLEMENTAL FIGURES :**

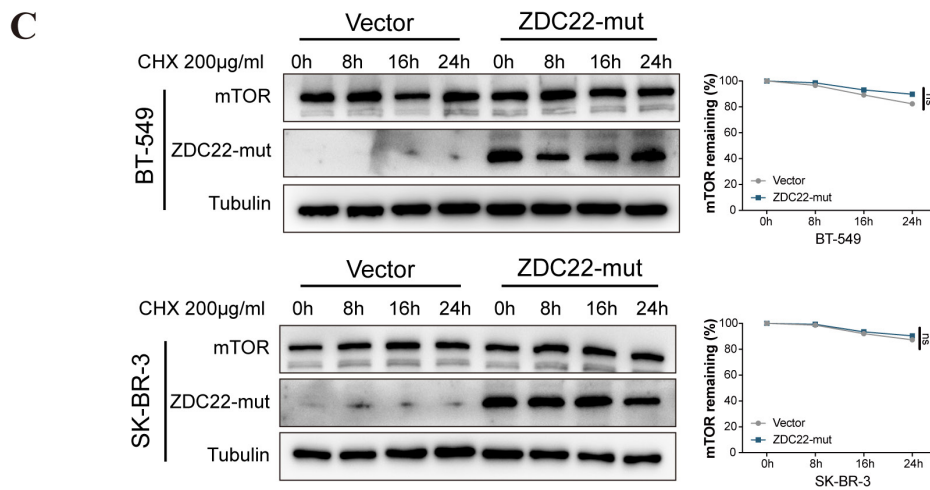
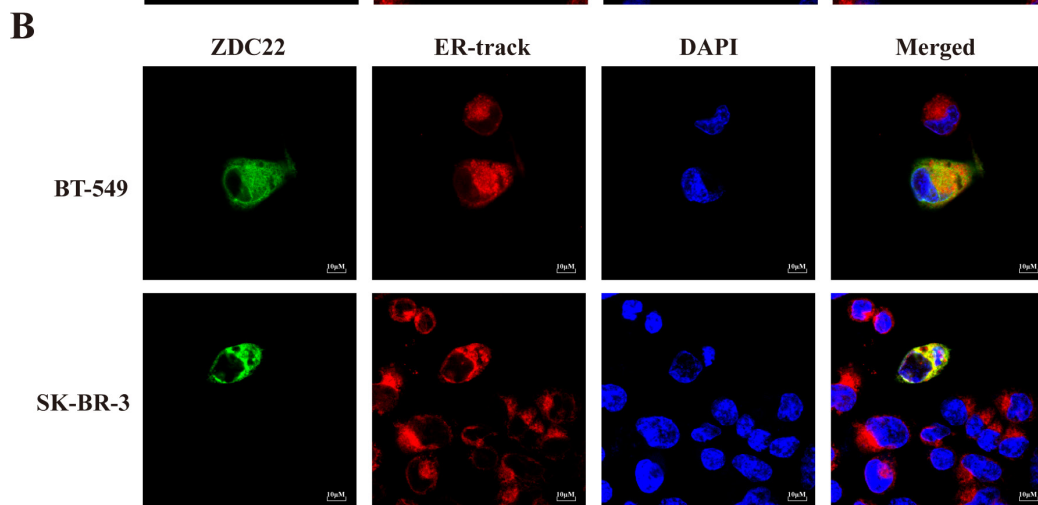
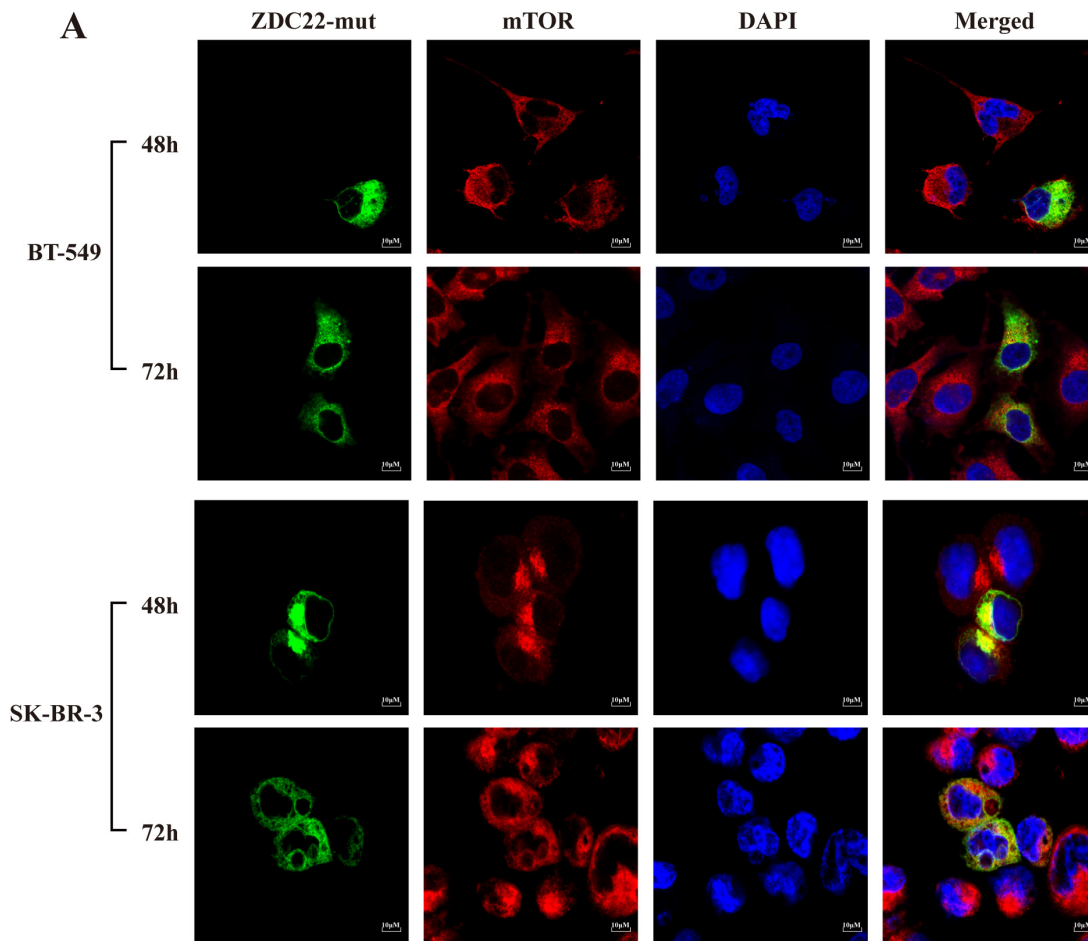
2 **FIGURE S1.** A. Promoter methylation of ZDHHC22 in 64 breast cancer tissue samples. **B.**
3 Validation of ZDHHC22 and ZDHHC22-(C111A) mRNA and protein expression by qPCR and
4 western blot analyses. **C-D.** Cell vitality was detected with CCK-8 assay and colony formation
5 assay after transfection with Vector, ZDHHC22 and ZDHHC22-(C111A) plasmids in YCC-B1 cells.
6 **E-F.** Cell cycle and cell apoptosis in YCC-B1 cells at 48 hours post-transfection of Vector,
7 ZDHHC22 and ZDHHC22-(C111A) plasmids was examined by flow cytometry analysis of PI
8 staining and Annexin V-APC/PI staining, respectively. **G** Immunofluorescence assays were used to
9 detect the protein expression of PHLPP2 after transfected with ZDHHC22 plasmids. Values
10 represent mean \pm SD. from three independent experiments, $**p < 0.01$. $***p < 0.001$. ZDC22:
11 ZDHHC22, ZDC22-mut: ZDHHC22-(C111A) mutation.



1

2

1 **FIGURE S2. A.** Immunofluorescence assays were used to detect the protein expression and
2 co-localization of mTOR at the indicated time in BT-549 and SK-BR-3 cells transfected with
3 ZDHHC22-(C11A) plasmids. **B.** The ZDHHC22 mainly co-localized with the endoplasmic
4 reticulum. **C.** BT-549 and SK-BR-3 cells were transfected with Vector and ZDHHC22-mut
5 plasmids for 48 hours, followed by 200µg/ml cycloheximide (CHX) treatment for the indicated
6 times. mTOR protein expression levels were detected with western blot and quantified with Image J
7 software. ZDC22:ZDHHC22, ZDC22-mut: ZDHHC22-(C11A) mutation.



1 **TABLE S1.** List of expression primers used in this study.

PCR	Primer	Sequence (5'-3')	Product size	PCR Cycles
RT-PCR	ZDHHC22F	GTGACCTTCGTGCTGCAGCT	167bp	35
	ZDHHC22R	AGGTCGTCTGGGGAGTTCTG		35
	GAPDHF	GGAGTCAACGGATTTGGT	206bp	23
	GAPDHR	GTGATGGGATTTCCATTGAT		23
qRT-PCR	mTORF	TGTGGGCAGCATCACTCTT	80bp	
	mTORR	GGCGAACAAATTGGGTCAG		
	GAPDHF	CCAGCAAGAGCACAAGAGGAA	114bp	
	GAPDHR	CAAGGGGTCTACATGGCAACT		

Note: RT-PCR: Semiquantitative reverse transcription PCR;

qRT-PCR: quantitative real-time PCR.

2

3

1 **TABLE S2.** List of MSP primers used in this study.

MSP	Primer	Sequence (5'-3')
ZDHHC22m1	ATAAGAGGAGTTTCGGACGTC	101bp
ZDHHC22m2	CCAATTCCCGAAACGAAACG	
ZDHHC22u1	GATAAGAGGAGTTTTGGATGTT	104bp
ZDHHC22u2	ACCCAATTCCCAAACAAAACA	

Note: MSP: methylation-specific PCR

2

3

1 **TABLE S3.** Prediction of mTORC2 complex and AKT palmitoylation

Protein	N-terminal sequence or entry code	Predicted N-terminus of the mature protein	Likelihood (%)
mTOR	NP_004949.1	*M (1)	100
Deptor-X1	NP_073620.2	Ac-M (1)	100
Deptor-X2	NP_001269941.1	Ac-M (1)	100
Protor1-X1	NP_851850.1	M (1)	89
Protor1-X2	NP_001017528.1	AC-S (2)	85
Protor1-X3	NP_00107529.1	V (2)	100
Protor1-X4	NP_001185650.1	V (2)	100
Sin1-X1	NP_001006618.1	Ac-A (2)	83
Sin1-X2	NP_007022.1	Ac-A (2)	83
Sin1-X3	NP_001006620.1	Ac-A (2)	83
Sin1-X4	NP_001006621.1	Ac-T (2)	77
Sin1-X5	NP_001006619.1	Ac-A (2)	83
mLST8-Xa	NP_001186102.1	Ac-M (1)	67
mLST8-Xb	NP_001186104.1	Ac-M (1)	67
mLST8-Xc	NP_001338986.1	Ac-M (1)	67
mLST8-Xd	NP_001338988.1	M (1)	89
Rictor-X1	NP_689969.2	Ac-A (2)	83
Rictor-X2	NP_001272368.1	Ac-A (2)	83
Rictor-X3	NP_001272369.1	My-G (2)	45
AKT-X1	NP_001014431.1	AC-S (2)	85
AKT-X2	NP_001014432.1	AC-S (2)	85
AKT-X3	NP_005154.2	AC-S (2)	85

2