

Supplementary Data legend

Supplementary table1.

Six week old female athymic nude mice were injected subcutaneously with 1×10^6 cells suspended in Matrigel. Animals were euthanized and necropsies performed when tumors reached 1-1.5 cm in diameter or after 6 months if no tumors were observed. Each cell line was tested in at least 5 animals.

Supplementary Figure 1.

RhoA-immortalized cells are anchorage-dependent. 2×10^4 cells were suspended in 0.3% of agarose and plated on top of 0.6% agarose in DMEM medium as described in Materials and Methods. The number of colonies was counted after two weeks. Breast cancer cell line Hs578T cells used as positive control.

Supplementary Figure 2.

Quantitative PCR analysis of gene expression in normal and RhoA immortal cells.

Total RNA was isolated, single-stranded cDNA was produced by reverse transcription and quantitative PCR was performed using 25 ng cDNA per reaction. Results are given in fold change as compared to normal parental cells, 70N. The data shown here is mean \pm S.D. from one representative experiment performed in triplicates. B-actin (ACTB) was used as normalization control.

The primer sequences used in this study:

CLCA2 forward	GCAAGATGGCAGAGGCTGACAGA
CLCA2 reverse	GGTGGGCAGATATGAAACCAGCAA
ELF3 forward	GAGTTCATCCGGGACATCCTCATC
ELF3 reverse	CAGGATCTCCCGTTTGTAGTAGTACCTCAT
S100P forward	CCAGGCTTCCTGCAGAGTGGAA
S100P reverse	GGCTCTGCCAGGAATCTGTGACA
ZNF217 forward	CCAGCTCGACGTTAGAAGGAAAAAGG
ZNF217 reverse	GGGAGTAAGCACTGACATCCACCAA
ACTB forward	CCTTCCTTCCTGGGCATGGA
ACTB reverse	CTGGGTGCCAGGGCAGTGAT

Supplementary Fig.3.

Data from Oncomine database (1) show that S100P is overexpressed in breast cancer and its overexpression is correlated with tumor grade and invasive. **A.** S100P is overexpressed in tumors in one breast cancer data set (2). Class 1: normal breast (7 samples), class 2: breast carcinoma (40 samples). **B.** S100P expression is higher in invasive breast cancers(IDC) as compared to breast ductal carcinoma in situ (DCIS)(3). Class 1: DCIS(3 samples), class 2: IDC (33 samples). **C.** S100P overexpression is correlated with tumor grade in one breast cancer data set (4). Class 1: Elston grade 1(68 samples), class 2: Elston grade (126 samples), class 3: Elston grade 3 (55 samples). **D.** S100P overexpression is correlated with tumor grade in another breast cancer data set (5). Class 1: Elston grade 1(67 samples), class 2: Elston grade (128 samples), class 3: Elston grade 3 (54 samples).

Supplementary Fig.4.

Data from Oncomine database (1) show that DAB2 is down-regulated in breast tumor , its downregulation correlate with tumor grade and lymphocytic infiltrate. **A.** DAB2 is down-regulated in breast tumor in one data set (2). Class 1: normal breast (7 samples), class 2: breast carcinoma (40 samples). **B.** DAB2 downregulation is correlate with lymphocytic infiltrate in one data set (6). Class 1: lymphocytic infiltrate negative (89 samples), class 2: lymphocytic infiltrate positive(28 samples). **C.** DAB2 down-regulation is correlate with tumor grade in another breast cancer data set(1). Class 1: Elston grade 1(30 samples), class 2: Elston grade 2 (107 samples), class 3: Elston grade 3 (54 samples).

Supplementary References for Oncomine data

1. Rhodes DR, Yu J, Shanker K, et al. A cancer microarray database and integrated data-mining platform, *Neoplasia* 2004; 6:1–6.
2. Richardson AL, Wang ZC, De Nicolo A, et al. X chromosomal abnormalities in basal-like human breast cancer. *Cancer Cell* 2006; 9:121-32.
3. Radvanyi L, Singh-Sandhu D, Gallichan S, et al. The gene associated with trichorhinophalangeal syndrome in humans is overexpressed in breast cancer. *Proc Natl Acad Sci U S A* 2005;102:11005-10.
4. Ivshina AV, George J, Senko O, et al. Genetic reclassification of histologic grade delineates new clinical subtypes of breast cancer. *Cancer Res* 2006;66:10292-301.

5. Miller LD, Smeds J, George J, et al. An expression signature for p53 status in human breast cancer predicts mutation status, transcriptional effects, and patient survival. *Proc Natl Acad Sci U S A* 2005;102:13550-5.
6. van 't Veer LJ, Dai H, van de Vijver MJ, et al. Gene expression profiling predicts clinical outcome of breast cancer. *Nature* 2002;415:530-6.