Supporting Information

Latimer et al. 10.1073/pnas.0914772107

Fig. S1. Supervised analysis of genomic instability using a set of 676 probes representing genes from all five DNA repair pathways (NER, base excision repair, mismatch repair, nonhomologous end-joining, and recombinational repair), translesion synthesis, replication, and chromosome segregation, assembled from commercial sources and Castro et al. (1). The dendrogram shows two major clusters representing nondiseased BRE and breast TSI explants. The color bars are set so that lower than the fifth centile of expression is the brightest green color and greater than the 95th centile is the brightest red.

1. Castro MAA, Mombach JC, de Almeida RM, Moreira JC (2007) Impaired expression of NER gene network in sporadic solid tumors. *Nucleic Acids Res* 35:1859–1867.

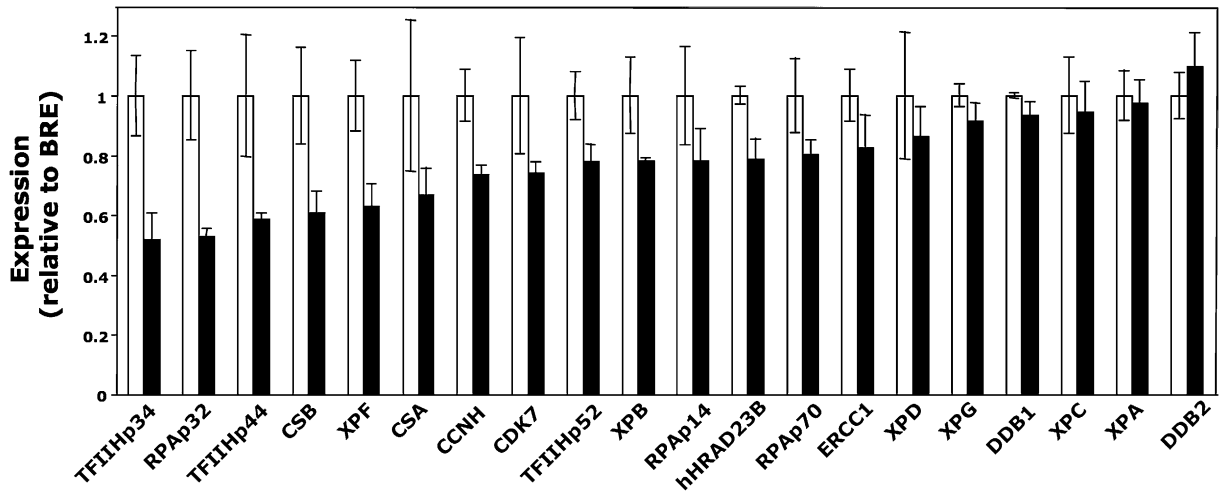

Fig. S2. Triplicate microarray analysis of NER genes in the BRE and TSI samples further processed for RPA and Western analysis. Summary of results for 20 NER genes, showing mRNA expression in tumor (solid bars) versus age-matched normal breast epithelium (open bars). Gene expression was reduced in the TSI samples for 19 of the NER genes relative to that in BRE. This reduction was individually significant for six genes: *TFIIHp34* ($P = 0.040$), *RPAp32* ($P = 0.023$), *CSB* ($P = 0.047$), *XPF* ($P = 0.034$), *CCNH* ($P = 0.036$), and *hHR23B* ($P = 0.020$); and came close to significance for four others: *TFIIHp44* ($P = 0.086$), *TFIIHp44* ($P = 0.083$), *RPAp70* ($P = 0.086$), and *XPG* ($P = 0.061$).

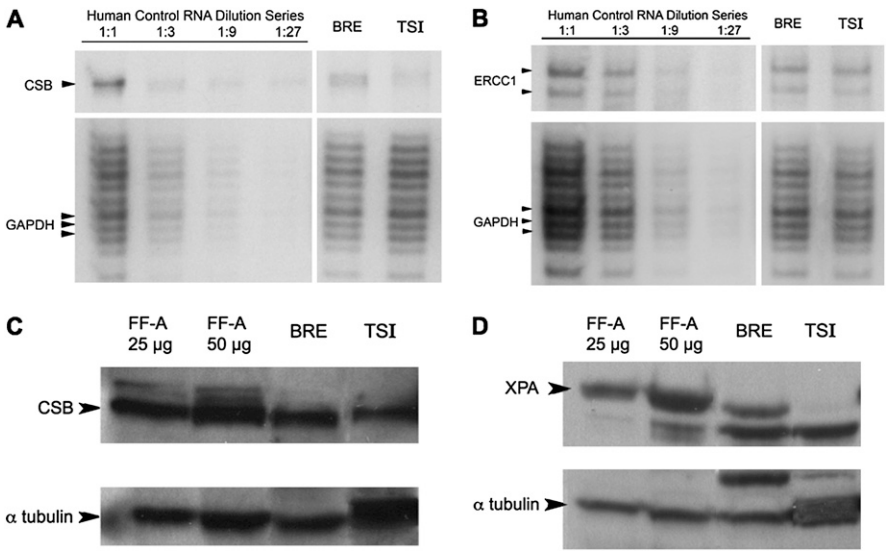

Fig. S3. Representative molecular analyses of NER genes in nondiseased BRE and breast TSI explant cultures. Quantitative RPA of the (*A*) *CSB* and (*B*) *ERCC1* genes. Acrylamide gels of mRNA specific to these genes demonstrate significantly lower expression of *CSB*, but not *ERCC1*, mRNA in tumor versus normal breast epithelium. Densitometric quantification was achieved by comparison with the control human RNA dilution series after normalization to the signal from the housekeeping gene *GAPDH*. Protected bands densitometrically quantified for each transcript are indicated by arrows. Western analysis of the products of the (*C*) *CSB* and (*D*) *XPA* genes. Expression of protein products specific to these genes demonstrates significantly lower expression of both gene products in tumor versus normal breast epithelium. Quantification was achieved by comparison with protein from human FFs (FF-A), which have fivefold higher baseline NER capacity than breast epithelium (1), after normalization to the signal from the structural protein α-tubulin. Specific bands quantified for each protein are indicated by arrows.

1. Latimer JJ, et al. (2003) Unique tissue-specific level of DNA nucleotide excision repair in primary human mammary epithelial cultures. *Exp Cell Res* 291:111–121.

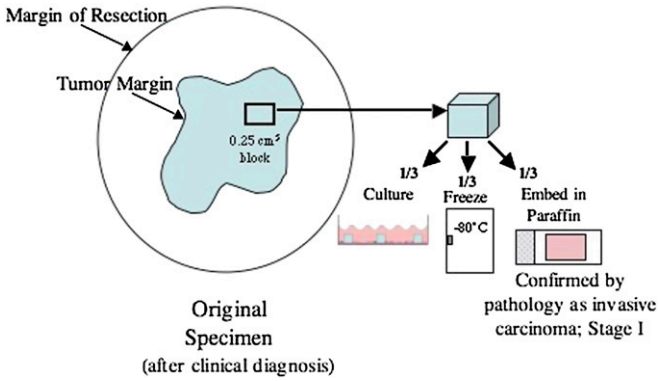

Fig. S4. Schematic of the processing of TSI tissue samples after clinical diagnosis. A block excised from within the tumor margins was split into three pieces: one piece was used for in vitro culture and subsequent NER analysis, another piece was used for histopathological confirmation by creation of a second paraffin-embedded block of fixed tissue, and a third piece was frozen for future analysis.