**Supporting Information for Chakraborty *et al*.,** Genomic Instability Promoted by Overexpression of Mismatch Repair Factors in Yeast: A Model for Understanding Cancer Progression. Genetics doi: 10.1534/genetics.118.300923

**Legends for Supporting Figures and Tables**

**Figure S1 Comparison of gene expression patterns of *MSH2*, *MSH3* and *MSH6* across diverse human cancer and normal tissues using GENT database.** GENT (Gene Expression across Normal and Tumor tissue; http://medical-genome.kribb.re.kr/GENT/), a web-accessible database, was used to compare gene expression patterns of *MSH2*, *MSH3* and *MSH6* between tumor and normal tissues. This database analyzes more than 24,000 samples from multiple normal or cancer tissues, profiled by the gene expression array U133plus2. Presented are gene expression levels in multiple cancer tissues (C) and the corresponding normal tissues (N) analyzed using the U133Plus2 platform. The data are presented as boxplots and each circle represents an individual sample. The Y-axis of the plot indicates normalized expression measures. The line inside each box represents the median. Mann-Whitney tests were performed to compare cancer-normal expression levels and corrected for multiple testing using the Bonferroni correction. Differences were considered significant when $p < 0.05$.

**Figure S2 Correlation of MSH2 and MSH6 protein expression in TCGA tumor types with significantly upregulated *MSH2* and *MSH6* mRNA expression**. Normalized MSH2 (X-axis) and MSH6 (Y-axis) protein levels from individual tumors quantified by reverse phase protein arrays were obtained from The Cancer Proteome Atlas (http://tcpaportal.org/tcpa/). Correlation and p-value were calculated using Spearman correlation and Spearman rank test, respectively. TCGA tumor codes are: BRCA, breast invasive carcinoma; COAD, colon adenocarcinoma; HNSC, head and neck squamous cell carcinoma; LIHC, liver hepatocellular carcinoma; LUAD, lung adenocarcinoma; LUSC, lung squamous cell carcinoma; READ, rectum adenocarcinoma.

**Figure S3 Western blot analysis of Msh2 and Msh6 levels; independent measurements.** Detection of Msh2 (A) and Msh6 (B) levels in BJ5464 containing the indicated $2\mu$ vectors (Table S2; Figure 3). The indicated amounts of protein extract were loaded onto each lane and extracts were electrophoresed in an 8% SDS-PAGE gel and then stained with Coomassie blue or analyzed in Western blots. The indicated dilutions (1/4, 1/8) of extract were also analyzed in Western blots immunostained with antibodies specific to Msh2 and Msh6 (Materials and Methods).

**Figure S4 Western blot analysis of Msh2 and Msh6 levels in strains containing *ARS-CEN* plasmids.** Detection of Msh2 and Msh6 levels in BJ5464 containing the indicated *2μ* or *ARS-CEN* vectors (Table S2). 15 μg of protein extracts were loaded onto each lane and extracts were electrophoresed in an 8% SDS-PAGE gel. Gels were stained with Coomassie blue or analyzed in Western blots immunostained with antibodies specific to Msh2 and Msh6 (Materials and Methods).

**Table S1 Literature review of MMR proteins overexpressed in cancers.** Summary of studies that have found various MMR genes or proteins to be overexpressed in a variety of cancers and outcomes they have been linked to.

**Table S2 Strains and plasmids used in this study.** FY23 was obtained from Fred Winston (Winston *et al.* 1995); SJR328, SJR769, GCY615 and GCY559 from Sue Jinks-Robertson (Nicholson *et al.* 2000); JAY1201 from Lucas Argueso (Conover *et al.* 2015); BJ5464 from

Elizabeth Jones. The indicated genes were all cloned into pRS424 or pRS426 $2\mu$ plasmids (Christianson *et al*. 1992).

**Literature Cited**


Castrilli, G., A. Fabiano, G. La Torre, L. Marigo, C. Piantelli *et al.*, 2002 Expression of hMSH2 and hMLH1 proteins of the human DNA mismatch repair system in salivary gland tumors. J. Oral. Path. Med. 31**:** 234-238.

Christianson, T. W., R. S. Sikorski, M. Dante, J. H. Shero, and P. Hieter, 1992 Multifunctional yeast high-copy-number shuttle vectors. Gene 110**:** 119-122.

Conover, H. N., S. A. Lujan, M. J. Chapman, D. A. Cornelio, R. Sharif *et al.*, 2015 Stimulation of chromosomal rearrangements by ribonucleotides. Genetics 201**:** 951-961.

Friedrich, M., C. Villena-Heinsen, R. Meyberg, A. Woll-Hermann, K. Reitnauer *et al.*, 1999 Immunohistochemical analysis of DNA 'mismatch-repair' enzyme human Mut-S-homologon-2 in ovarian carcinomas. Histochem. J. 31**:** 717-722.

Hamid, A. A., M. Mandai, I. Konishi, K. Nanbu, Y. Tsuruta *et al.*, 2002 Cyclical change of hMSH2 protein expression in normal endometrium during the menstrual cycle and its overexpression in endometrial hyperplasia and sporadic endometrial carcinoma. Cancer 94**:** 997-1005.

Huang, S.C., S.F. Huang, Y.T. Chen, Y. Chang, Y.T. Chiu *et al.*, 2017 Overexpression of MutL homolog 1 and MutS homolog 2 proteins have reversed prognostic implications for stage I–II colon cancer patients. Biomed. J. 40**:** 39-48.

Jewell, R., C. Conway, A. Mitra, J. Randerson-Moor, S. Lobo *et al.*, 2010 Patterns of expression of DNA repair genes and relapse from melanoma. Clin. Cancer Res 16**:** 5211-5221.

Kauffmann, A., F. Rosselli, V. Lazar, V. Winnepenninckx, A. Mansuet-Lupo *et al.*, 2008 High expression of DNA repair pathways is associated with metastasis in melanoma patients. Oncogene 27**:** 565-573.

Leach, F. S., J. T. Hsieh, K. Molberg, M. H. Saboorian, J. D. McConnell *et al.*, 2000 Expression of the human mismatch repair gene hMSH2: a potential marker for urothelial malignancy. Cancer 88**:** 2333-2341.

Li, M., L. Liu, Z. Wang, L. Wang, Z. Liu *et al.*, 2008 Overexpression of hMSH2 and hMLH1 protein in certain gastric cancers and their surrounding mucosae. Oncol. Rep. 19: 401-406.

Li, M., Q. Zhang, L. Liu, W. Lu, H. Wei *et al.*, 2013 Expression of the mismatch repair gene hMLH1 Is enhanced in non-small cell lung cancer with EGFR mutations. PLoS One 8**:** e78500.

Nicholson, A., M. Hendrix, S. Jinks-Robertson, and G. F. Crouse, 2000 Regulation of mitotic homeologous recombination in yeast: functions of mismatch repair and nucleotide excision repair genes. Genetics 154**:** 133-146.

Norris, A. M., M. Gentry, D. M. Peehl, R. D'Agostino, and K. D. Scarpinato, 2009 The elevated expression of a mismatch repair protein Is a predictor for biochemical recurrence after radical prostatectomy. Cancer Epidemiol. Biomarkers Prev. 18**:** 57-64.

Norris, A. M., R. D. Woodruff, R. B. D'Agostino, J. E. Clodfelter, and K. D. Scarpinato, 2007 Elevated levels of the mismatch repair protein PMS2 are associated with prostate cancer. Prostate 67**:** 214-225.

Rass, K., P. Gutwein, C. Welter, V. Meineke, W. Tilgen *et al.*, 2001 DNA mismatch repair enzyme hMSH2 in malignant melanoma: increased immunoreactivity as compared to acquired melanocytic nevi and strong mRNA expression in melanoma cell lines. Histochem. J. 33**:** 459-467.

Srivastava, T., P. Chattopadhyay, A. K. Mahapatra, C. Sarkar, and S. Sinha, 2004 Increased

hMSH2 protein expression in glioblastoma multiforme. J. Neurooncol. 66**:** 51-57.
Stark, A. M., A. Doukas, H. H. Hugo, J. Hedderich, K. Hattermann *et al.*, 2015 Expression of DNA mismatch repair proteins MLH1, MSH2, and MSH6 in recurrent glioblastoma. Neurol. Res. 37**:** 95-105.
Vageli, D. P., S. Giannopoulos, S. G. Doukas, C. Kalaitzis, S. Giannakopoulos *et al.*, 2013 Mismatch repair hMSH2, hMLH1, hMSH6 and hPMS2 mRNA expression profiles in precancerous and cancerous urothelium. Oncol. Lett. 5**:** 283-294.
Velasco, A., P.S. Albert, H. Rosenberg, C. Martinez, and F.S. Leach, 2002 Clinicopathologic implications of hMSH2 gene expression. Cancer Biol. Ther. 1**:** 361-366.
Wagner, V. P., L. P. Webber, G. Salvadori, L. Meurer, F. P. Fonseca *et al.*, 2016 Overexpression of MutSα complex proteins predicts poor prognosis in oral squamous cell carcinoma. Medicine 95**:** e3725.
Wilczak, W., S. Rashed, C. Hube-Magg, M. Kluth, R. Simon *et al.*, 2017 Up-regulation of mismatch repair genes MSH6, PMS2 and MLH1 parallels development of genetic instability and is linked to tumor aggressiveness and early PSA recurrence in prostate cancer. Carcinogenesis 38**:** 19-27.
Winston, F., C. Dollard, and S. L. Ricupero-Hovasse, 1995 Construction of a set of convenient Saccharomyces cerevisiae strains that are isogenic to S288C. Yeast 11**:** 53-55.