

Perspective

Genome Instability Caused by a Germline Mutation in the Human DNA Repair Gene *POLB*

Robert W. Sobol^{1,2,3*}

1 Department of Pharmacology and Chemical Biology, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania, United States of America, **2** University of Pittsburgh Cancer Institute, Hillman Cancer Center, Pittsburgh, Pennsylvania, United States of America, **3** Department of Human Genetics, University of Pittsburgh Graduate School of Public Health, Pittsburgh, Pennsylvania, United States of America

DNA polymerase β (Pol β) is recognized as an essential DNA repair protein [1,2]. Although the smallest of the human DNA polymerases [3,4], this 335-amino-acid protein is the primary DNA polymerase in the base excision repair (BER) pathway [5]. A majority of the 20,000 DNA lesions per day that each human cell is faced with are repaired by the BER pathway [6]. These include products of base depurination and depyrimidination (abasic sites), deamination of cytosine and 5-methylcytosine, oxidation products such as 8-oxo-7,8-dihydro-2'-deoxyguanosine (8-oxodG), thymine glycol and lipid peroxidation products, as well as methylation modifications such as N7-guanine [7,8]. Failure to repair these spontaneous or endogenously induced DNA base lesions as well as the numerous base modifications that arise from environmental or exogenous sources can result in multiple cellular effects, including cell death, gene mutations, gene rearrangements, and/or decreased cell growth rate. Pol β facilitates the repair of these base lesions in concert with different proteins of the BER pathway depending on the lesion [2,7]. Once the base lesion is removed by one of 11 DNA glycosylase enzymes and the resulting abasic site is hydrolysed by the endonuclease APE1, Pol β is recruited to the lesion via an interaction with the BER scaffold protein XRCC1 [9,10] and the DNA damage sensor PARP1 [11–13]. Pol β then conducts two essential enzymatic functions: 5'dRP lyase-mediated gap tailoring and DNA polymerase-mediated DNA synthesis to fill the gap [2,3]. The 5'dRP lyase activity functions to “tailor” the gap by removing the sugar-phosphate residue that remains after APE1 cleaves the DNA backbone, and then the polymerase activity adds the newly synthesized nucleotide that was removed during repair. Considering the critical and essential role of these two enzymatic activities, the important protein-protein interactions between Pol β and several BER proteins [14], as well as the increasing number of post-translational modifications suggested to affect Pol β function and stability [15], it may not be

surprising that a significant number of somatic mutations in *POLB* have been observed in cancer (Table 1). Within the 33 Kb *POLB* gene (PubMed geneID #5423), as many as 567 SNPs have been identified (see dbSNP). However, only 34 SNPs are in or near the coding region (22 are found in exons), and only two have been confirmed in larger cohorts. These two germline *POLB* mutants (R137Q; rs12678588 and P242R; rs3136797) have been reported to be present in as much as 0.6% and 2.4% of the human population, respectively [16,17]. However, little is known about the functional impact that results from these single amino acid alterations. An earlier study on the Pol β (R137Q) mutant (rs12678588) suggested that the R137Q mutation impairs function of the purified protein. Further, when produced in mouse cells, the R137Q mutant protein interfered with Pol β binding to PCNA [18] and the response of mouse cells to DNA-damaging agents, although no information was provided on the impact of this mutation on genome stability. Whereas the Pol β (P242R) mutant allele (rs3136797) has been linked with altered incidence of cancer in several studies [19–21], there have been few or no studies defining the impact of this SNP on Pol β function, DNA repair capacity, and genome maintenance in human cells.

In this issue of *PLOS Genetics*, Sweasy and colleagues conducted a detailed analysis of the *POLB* germline-coding SNP rs3136797 [22]. This polymorphism alters amino acid 242, changing the amino acid from a proline (P; Pro) to an arginine (R;

Arg). To determine whether the P242R mutation affected genome stability in human or mouse cells, the wild-type (WT) or P242R protein was produced in human normal mammary epithelial cells (MCF10A) and in mouse embryonic fibroblasts (MEFs). In both cell lines, the synthesis of the P242R protein led to an increase in genomic alterations. Analysis of metaphase spreads showed that the P242R protein induced an increase in chromosome breaks and a significant increase in fragmented chromosomes and chromosome fusions. In other reports, cancer-specific mutations in Pol β (e.g., Y265C) induced an increase in mutant frequency [23,24] that could explain the increase in chromosome alterations seen with P242R. However, cells producing the P242R protein were found to have the same mutant frequency as those expressing WT Pol β . The lack of an increase in mutations together with the increase in chromosomal instability suggested that the Pol β (P242R) protein may promote the accumulation of DNA strand breaks during repair. To test this hypothesis, the cells were treated with methyl methanesulfonate (MMS) to induce DNA damage repaired by Pol β [5]. As suspected, exposure of the cells expressing the P242R mutant to MMS induced a greater level of single-strand and double-strand DNA breaks. The increase in single-strand breaks and related BER intermediates was measured by the alkaline Comet assay [25], and an increase in the number of DNA double-strand breaks was indirectly determined by measuring an increase in γ -H2AX foci

Citation: Sobol RW (2012) Genome Instability Caused by a Germline Mutation in the Human DNA Repair Gene *POLB*. *PLoS Genet* 8(11): e1003086. doi:10.1371/journal.pgen.1003086

Editor: Susan M. Rosenberg, Baylor College of Medicine, United States of America

Published: November 8, 2012

Copyright: © 2012 Robert W. Sobol. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Funding: RWS is supported by grants from the National Institutes of Health (NIH) [GM087798; CA148629; ES019498, R43ES021116 & R43GM099213]. The funder had no role in the preparation of the article.

Competing Interests: The author has declared that no competing interests exist.

* E-mail: rws9@pitt.edu

Table 1. Germline and somatic *POLB* mutants.*

WT Residue	Residue Number	Mutant Residue	Pol β Domain	Functional or Correlative Effect of Mutation	Citation
Gln	8	Arg	8K	<i>n.d.</i>	[17]
Leu	22	Pro	8K	Loss of 5' dRP lyase activity	[37]
Leu	22	Pro	8K	Suppressed BER activity	[38]
Lys	27	Asn	8K	Decreased catalytic (5' dRP lyase) activity	[39]
Tyr	39	Cys	8K	<i>n.d.</i>	[38]
Gly	80	Arg	8K	<i>n.d.</i>	[40]
Ile	88	Val	8K	<i>n.d.</i>	[41,42]
Phe	114	Ser	Fingers	<i>n.d.</i>	[41]
Gly	118	Glu	Fingers	<i>n.d.</i>	[41]
Glu	123	Lys	Fingers	Decreased catalytic (polymerase) activity	[39]
Arg	137	Gln	Fingers	Decreased interaction with PCNA, reduced polymerase activity	[18]
Arg	137	Gln	Fingers	<i>n.d.</i>	[17]
Arg	137	Gln	Fingers	Haplotype analysis	[16]
Asp	160	Asn	Palm	<i>n.d.</i>	[38]
Asp	160	Asn	Palm	Increase in cellular transformation	[27]
Lys	167	Ile	Palm	<i>n.d.</i>	[41]
Gly	179	Arg	Palm	<i>n.d.</i>	[41]
Arg	182	Gly	Palm	<i>n.d.</i>	[43]
Arg	183	Gly	Palm	<i>n.d.</i>	[41]
Glu	186	Gly	Palm	<i>n.d.</i>	[41,42]
Glu	216	Lys	Palm	No observed change in activity	[39]
Gly	231	Asp	Palm	Decreased catalytic rate and decreased binding affinity of nucleotides	[44]
Glu	232	Lys	Palm	Decreased catalytic (polymerase) activity	[39]
Met	236	Leu	Palm	Decreased catalytic (polymerase) activity	[39]
Cys	239	Arg	Palm	<i>n.d.</i>	[38]
Pro	242	Arg	Palm	<i>n.d.</i>	[17,45]
Pro	242	Arg	Palm	Decreased catalytic (polymerase) activity	[39]
Pro	242	Arg	Palm	Haplotype analysis	[16]
Pro	242	Arg	Palm	Decreased risk of colorectal cancer	[19]
Pro	242	Arg	Palm	Increase in cellular transformation and genome instability	[22]
Ile	260	Met	Palm	Misalignment-mediated errors in dipyrimidine sequences	[46]
Ile	260	Met	Palm	Increase in cellular transformation	[28]
Tyr	265	Cys	Thumb	Increase in mutation frequency	[23,24]
Tyr	265	Cys	Thumb	Increase in BER intermediates, chromosome aberrations, and DNA breaks	[47]
Asn	281	Ser	Thumb	<i>n.d.</i>	[40]
Glu	288	Lys	Thumb	Increase in mutations at A/T base pairs	[48]
Lys	289	Met	Thumb	Increase in mutation frequency	[49]
Lys	289	Met	Thumb	<i>n.d.</i>	[45,50]
Lys	289	Met	Thumb	Increase in cellular transformation	[28]
Asn	294	Asp	Thumb	<i>n.d.</i>	[38]
Glu	295	Lys	Thumb	Decreased polymerase activity, acts as a dominant negative	[51]
Glu	295	Lys	Thumb	Loss of BER and DNA polymerase activity	[38]
Glu	295	Lys	Thumb	Decreased polymerase activity that may stem from steric interaction with Arg258	[52]

n.d. = not determined.

*See [53] for an extensive list of *POLB* gene mutations recently identified in colorectal tumors.
doi:10.1371/journal.pgen.1003086.t001

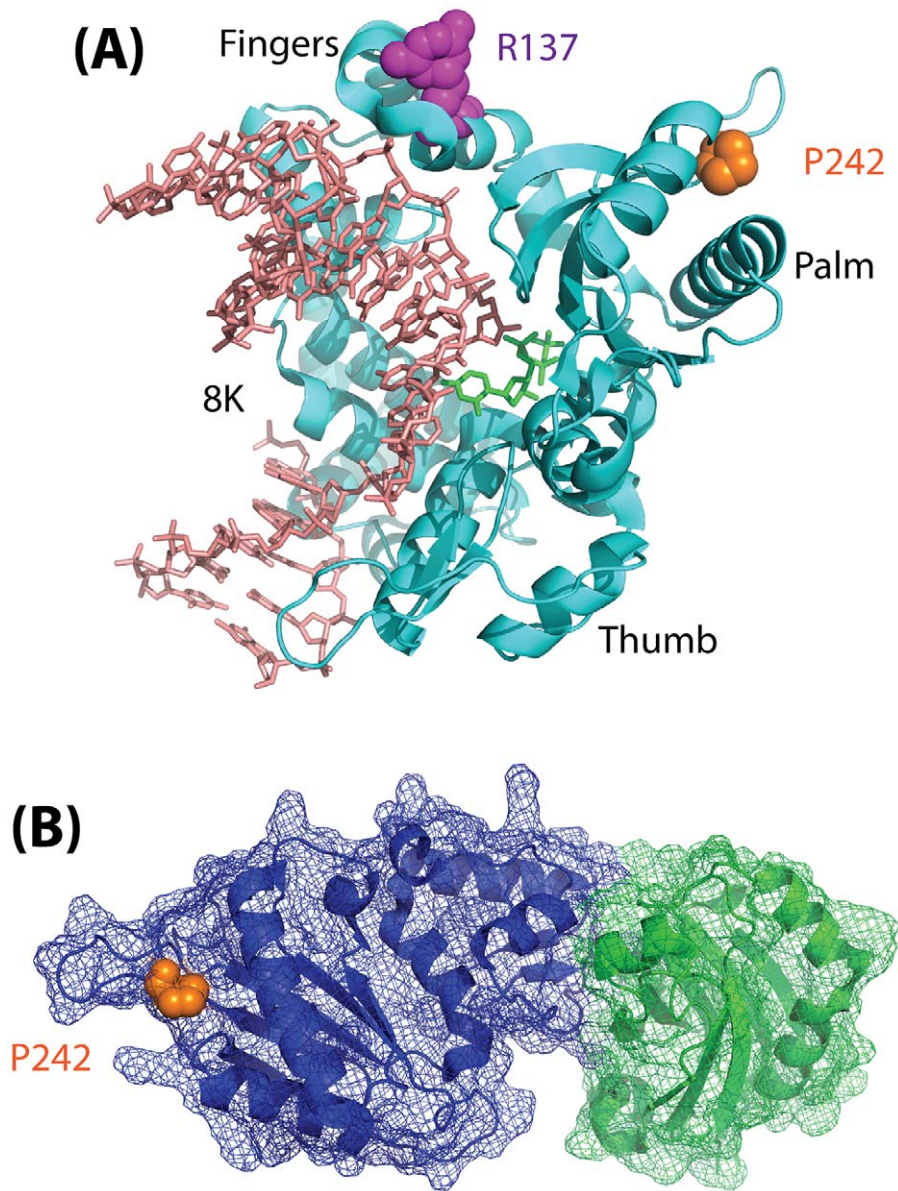


Figure 1. Model depicting the structure of Pol β . (A) Structure (pdb2fms) depicting DNA Polymerase β (Pol β) with a gapped DNA substrate and dUMPNPP with magnesium in the catalytic site [35]. The image is a cartoon rendition of the polypeptide chain of Pol β in teal, the gapped DNA substrate in salmon, and the incoming dUMPNPP base in green. Amino acids known to be altered by germline mutations are shown using a space-filling rendering: R137 (magenta) and P242 (orange). The fingers, palm, and thumb domains of Pol β are indicated. The 8K domain is at the back of the structure, facing away from the plane of the image, and is shown behind the DNA in this orientation. (B) Structure (pdb3lqc) depicting oxidized XRCC1 bound to the Pol β palm/thumb domains [36]. The image is a cartoon rendition of the palm and thumb domains of Pol β in blue, with a mesh illustrating the surface of the structure (amino acids 150–335), and a cartoon rendition of the oxidized form of XRCC1 in green, with a mesh illustrating the surface of the structure (amino acids 1–151). Amino acid P242 (orange) is shown using a space-filling rendering. The images were generated using PyMOL (Molecular Graphics System, Version 1.2r3pre; Schrödinger, LLC; <http://pymol.org/>). doi:10.1371/journal.pgen.1003086.g001

[26]. A second phenotype that Sweasy and colleagues have linked with cancer mutants of Pol β is the ability to induce cellular transformation, as was seen with the D160N, I260M, and K289M Pol β mutants [27,28]. Similarly, production of the P242R mutant protein in mouse cells (C127 λ) or human cells (MCF10A) increased growth in soft agar significantly in comparison with expression of WT Pol β .

Yamitch et al. [22] then used both cellular analysis and biochemical measurements to evaluate the functional impact of the P242R mutation. WT MEF cells (expressing endogenous WT Pol β) were engineered to produce either WT human Pol β or the P242R mutant protein and were then exposed to MMS to measure cellular survival. Both WT- and P242R-expressing cells responded equally except

at the highest doses of MMS. Next, Pol β -knock out (KO) MEFs engineered to express either WT human Pol β or the P242R mutant were exposed to MMS to determine whether the P242R mutant could restore (complement) resistance to MMS. In this case, there was a small but significant difference in response, suggesting that the P242R mutant was mildly defective in BER. A strength of the Sweasy

lab's study is the use of cell biology analyses as well as detailed biochemical evaluation of these mutant proteins. Yamtich et al. [22] expressed and purified the WT and P242R mutant proteins from *E. coli* and measured the rate of DNA polymerase activity by two separate kinetic analyses. This provided the opportunity to determine whether the decreased BER capacity observed in the Pol β -KO MEFs expressing the P242R mutant in response to MMS and the increase in DNA breaks were the result of a defect in the polymerase activity of the P242R mutant. In both cases, they found that the mutation (P242R) caused a decrease in the rate of DNA polymerase activity. However, the protein bound to the DNA substrate with affinity equal to that of the WT enzyme. The slow polymerase activity of the P242R protein therefore is likely to promote the accumulation of BER intermediates, inducing genome alterations when the cell is exposed to DNA-damaging agents [29].

Defects in Pol β can have significant cellular ramifications, especially in response to DNA-damaging agents that require Pol β and BER for repair. Complete loss of Pol β function can trigger an increase in cell death in response to high doses

of genotoxins [5,30] and an increase in genome alterations even at low doses [29]. Additional cellular responses to DNA damage when Pol β is defective may include PARP1 activation and alterations in bioenergetic metabolites such as NAD⁺ [31]. The steady-state expression level of Pol β is also reported to be regulated by the proteasome via ubiquitylation [32], suggesting that some Pol β mutants may have altered stability. In this regard, the observation that the P242R mutant protein has a functional defect now opens the door for further studies to clarify the mechanisms and cellular impacts of other defects in Pol β . It has been suggested that tumor-specific defects in BER, such as a defect in Pol β , may be exploited for selective therapeutic options [33]. Cells producing the mutant protein (P242R) have a higher level of DNA strand breaks and increased cellular transformation, and so it is possible that the Pol β (P242R) protein may be considered a driver of cancer formation. It remains to be determined whether the presence of this mutant protein (P242R) provides therapeutic selectivity.

Finally, it remains to be determined how a mutation (P242R) so distant from the

Pol β active site, and which does not interfere with binding to XRCC1 (Figure 1), can have such a significant effect on the function of Pol β . Given the subtle yet significant impact of the Pol β (P242R) mutant on cellular function and genome stability in response to DNA damage as described by Yamtich et al [22], further analysis of this mutant protein is warranted. The P242 amino acid is located in a loop domain that is essential for enzymatic activity [34], so it is likely that the alteration of the amino acid from P to R changes the movement of the loop and may also change the overall architecture of the protein. To more completely appreciate the subtle yet significant defect associated with this germline mutation, it is therefore suggested that future studies be conducted to determine the structure of the ternary complex of Pol β (P242R) with DNA and an incoming nucleotide. In addition, whole animal studies should be considered so as to determine whether the genome instability and cellular transformation results described [22] extend to additional cell types. As a germline mutation, analysis of the P242R mutant protein in an animal model will provide valuable insight into the possible effects on human health.

References

- Burgers PM, Koonin EV, Bruford E, Blanco L, Burtis KC, et al. (2001) Eukaryotic DNA polymerases: proposal for a revised nomenclature. *J Biol Chem* 276: 43487–43490.
- Almeida KH, Sobol RW (2007) A unified view of base excision repair: lesion-dependent protein complexes regulated by post-translational modification. *DNA Repair* 6: 695–711.
- Beard WA, Wilson SH (2006) Structure and mechanism of DNA polymerase Beta. *Chem Rev* 106: 361–382.
- Lange SS, Takata K, Wood RD (2011) DNA polymerases and cancer. *Nature Reviews Cancer* 11: 96–110.
- Sobol RW, Horton JK, Kuhn R, Gu H, Singhal RK, et al. (1996) Requirement of mammalian DNA polymerase- β in base-excision repair. *Nature* 379: 183–186.
- Lindahl T (1993) Instability and decay of the primary structure of DNA. *Nature* 362: 709–715.
- Svilar D, Goellner EM, Almeida KH, Sobol RW (2011) Base excision repair and lesion-dependent sub-pathways for repair of oxidative DNA damage. *Antioxid Redox Signal* 14: 2491–2507.
- Friedberg EC, Walker GC, Siede W, Wood RD, Schultz RA, et al. (2006) DNA repair and mutagenesis, 2nd edition. Washington, D.C.: ASM Press. 1,164 p.
- Caldecott KW, Aoufouchi S, Johnson P, Shall S (1996) XRCC1 polypeptide interacts with DNA polymerase β and possibly poly (ADP-ribose) polymerase, and DNA ligase III is a novel molecular 'nick-sensor' *in vitro*. *Nucleic Acids Research* 24: 4387–4394.
- Lan L, Nakajima S, Oohata Y, Takao M, Okano S, et al. (2004) In situ analysis of repair processes for oxidative DNA damage in mammalian cells. *Proceedings of the National Academy of Science* 101: 13738–13743.
- Jelezcova E, Trivedi RN, Wang XH, Tang JB, Brown AR, et al. (2010) Parp1 activation in mouse embryonic fibroblasts promotes Pol beta-dependent cellular hypersensitivity to alkylation damage. *Mutat Res* 686: 57–67.
- Tang JB, Svilar D, Trivedi RN, Wang XH, Goellner EM, et al. (2011) N-methylpurine DNA glycosylase and DNA polymerase beta modulate BER inhibitor potentiation of glioma cells to temozolomide. *Neuro Oncol* 13: 471–486.
- Masaoka A, Horton JK, Beard WA, Wilson SH (2009) DNA polymerase beta and PARP activities in base excision repair in living cells. *DNA Repair (Amst)* 8: 1290–1299.
- Almeida KH, Sobol RW (2005) Increased specificity and efficiency of base excision repair through complex formation. In: Siede W, Doetsch PW, Kow YW, editors. DNA damage recognition. New York: Marcel Dekker Inc. pp. 33–64.
- Sobol RW (2008) CHIPping away at base excision repair. *Molecular Cell* 29: 413–415.
- Yamtich J, Speed WC, Straka E, Kidd JR, Sweasy JB, et al. (2009) Population-specific variation in haplotype composition and heterozygosity at the POLB locus. *DNA Repair (Amst)* 8: 579–584.
- Mohrenweiser HW, Xi T, Vazquez-Matias J, Jones IM (2002) Identification of 127 amino acid substitution variants in screening 37 DNA repair genes in humans. *Cancer Epidemiology Biomarkers & Prevention* 11: 1054–1064.
- Guo Z, Zheng L, Dai H, Zhou M, Xu H, et al. (2009) Human DNA polymerase beta polymorphism, Arg137Gln, impairs its polymerase activity and interaction with PCNA and the cellular base excision repair capacity. *Nucleic Acids Res* 37: 3431–3441.
- Moreno V, Gemignani F, Landi S, Gioia-Patricola L, Chabrier A, et al. (2006) Polymorphisms in genes of nucleotide and base excision repair: risk and prognosis of colorectal cancer. *Clin Cancer Res* 12: 2101–2108.
- Matakidou A, el Galta R, Webb EL, Rudd MF, Bridle H, et al. (2007) Genetic variation in the DNA repair genes is predictive of outcome in lung cancer. *Hum Mol Genet* 16: 2333–2340.
- Sellick GS, Wade R, Richards S, Oscier DG, Catovsky D, et al. (2008) Scan of 977 nonsynonymous SNPs in CLL4 trial patients for the identification of genetic variants influencing prognosis. *Blood* 111: 1625–1633.
- Yamtich J, Nemeec AA, Keh A, Sweasy JB (2012) A germline polymorphism of DNA polymerase beta induces genomic instability and cellular transformation. *PLoS Genet* 8: e1003052. doi:10.1371/journal.pgen.1003052
- Clairmont CA, Narayanan L, Sun KW, Glazer PM, Sweasy JB (1999) The Tyr-265-to-Cys mutator mutant of DNA polymerase beta induces a mutator phenotype in mouse LN12 cells. *Proceedings of the National Academy of Sciences USA* 96: 9580–9585.
- Opresko PL, Sweasy JB, Eckert KA (1998) The mutator form of polymerase beta with amino acid substitution at tyrosine 265 in the hinge region displays an increase in both base substitution and frame shift errors. *Biochemistry* 37: 2111–2119.
- Fortini P, Raspaglio G, Falchi M, Dogliotti E (1996) Analysis of DNA alkylation damage and repair in mammalian cells by the comet assay. *Mutagenesis* 11: 169–175.
- Paull TT, Rogakou EP, Yamazaki V, Kirchgessner CU, Gellert M, et al. (2000) A critical role for histone H2AX in recruitment of repair factors to nuclear foci after DNA damage. *Current Biology* 10: 886–895.
- Donigan KA, Hile SE, Eckert KA, Sweasy JB (2012) The human gastric cancer-associated DNA polymerase beta variant D160N is a mutator that induces cellular transformation. *DNA Repair (Amst)* 11: 381–390.
- Sweasy JB, Lang T, Starcevic D, Sun KW, Lai CC, et al. (2005) Expression of DNA polymerase

- {beta} cancer-associated variants in mouse cells results in cellular transformation. *Proc Natl Acad Sci U S A* 102: 14350–14355.
29. Sobol RW, Kartalou M, Almeida KH, Joyce DF, Engelward BP, et al. (2003) Base excision repair intermediates induce p53-independent cytotoxic and genotoxic responses. *Journal of Biological Chemistry* 278: 39951–39959.
 30. Sobol RW, Prasad R, Evenski A, Baker A, Yang XP, et al. (2000) The lyase activity of the DNA repair protein β -polymerase protects from DNA-damage-induced cytotoxicity. *Nature* 405: 807–810.
 31. Tang J, Goellner EM, Wang XW, Trivedi RN, St. Croix CM, et al. (2010) Bioenergetic metabolites regulate base excision repair-dependent cell death in response to DNA damage. *Molecular Cancer Research* 8: 67–79.
 32. Parsons JL, Tait PS, Finch D, Dianova, II, Allinson SL, et al. (2008) CHIP-mediated degradation and DNA damage-dependent stabilization regulate base excision repair proteins. *Mol Cell* 29: 477–487.
 33. Neijenhuis S, Verwijs-Janssen M, van den Broek LJ, Begg AC, Vens C (2010) Targeted radiosensitization of cells expressing truncated DNA polymerase β . *Cancer Research* 70: 8706–8714.
 34. Lin GC, Jaeger J, Eckert KA, Sweasy JB (2009) Loop II of DNA polymerase beta is important for discrimination during substrate binding. *DNA Repair (Amst)* 8: 182–189.
 35. Batra VK, Beard WA, Shock DD, Krahn JM, Pedersen LC, et al. (2006) Magnesium-induced assembly of a complete DNA polymerase catalytic complex. *Structure* 14: 757–766.
 36. Cuneo MJ, London RE (2010) Oxidation state of the XRCC1 N-terminal domain regulates DNA polymerase beta binding affinity. *Proceedings of the National Academy of Sciences USA* 107: 6805–6810.
 37. Dalal S, Chikova A, Jaeger J, Sweasy JB (2008) The Leu22Pro tumor-associated variant of DNA polymerase beta is dRP lyase deficient. *Nucleic Acids Res* 36: 411–422.
 38. Iwanaga A, Ouchida M, Miyazaki K, Hori K, Mukai T (1999) Functional mutation of DNA polymerase β found in human gastric cancer: inability of the base excision repair in vitro. *Mutation Research* 435: 121–128.
 39. An CL, Chen D, Makridakis NM (2011) Systematic biochemical analysis of somatic missense mutations in DNA polymerase beta found in prostate cancer reveal alteration of enzymatic function. *Hum Mutat* 32: 415–423.
 40. Forbes SA, Bindal N, Bamford S, Cole C, Kok CY, et al. (2011) COSMIC: mining complete cancer genomes in the Catalogue of Somatic Mutations in Cancer. *Nucleic Acids Res* 39: D945–950.
 41. Dong Z, Zhao G, Zhao Q, Yang H, Xue L, et al. (2002) [A study of DNA polymerase beta mutation in human esophageal cancer]. *Zhonghua Yi Xue Za Zhi* 82: 899–902.
 42. Zhao GQ, Wang T, Zhao Q, Yang HY, Tan XH, et al. (2005) Mutation of DNA polymerase beta in esophageal carcinoma of different regions. *World J Gastroenterol* 11: 4618–4622.
 43. Han LP, Qiao YH, Dong ZM, Shi HR, Zhao GQ, et al. (2003) [Study on DNA polymerase beta gene mutation in human cervical cancer]. *Zhonghua Fu Chan Ke Za Zhi* 38: 618–620.
 44. Nemeec AA, Donigan KA, Murphy DL, Jaeger J, Sweasy JB (2012) Colon cancer-associated DNA polymerase beta variant induces genomic instability and cellular transformation. *Journal of Biological Chemistry* 287: 23840–23849.
 45. Sliwinski T, Ziemba P, Morawiec Z, Kowalski M, Zdrozny M, et al. (2007) Polymorphisms of the DNA polymerase beta gene in breast cancer. *Breast Cancer Res Treat* 103: 161–166.
 46. Dalal S, Hile S, Eckert KA, Sun KW, Starcevic D, et al. (2005) Prostate-cancer-associated I260M variant of DNA polymerase beta is a sequence-specific mutator. *Biochemistry* 44: 15664–15673.
 47. Senejani AG, Dalal S, Liu Y, Nottoli TP, McGrath JM, et al. (2012) Y265C DNA polymerase beta knockin mice survive past birth and accumulate base excision repair intermediate substrates. *Proceedings of the National Academy of Sciences USA* 109: 6632–6637.
 48. Murphy DL, Donigan KA, Jaeger J, Sweasy JB (2012) The E288K colon tumor variant of DNA polymerase beta is a sequence specific mutator. *Biochemistry* 51: 5269–5275.
 49. Lang T, Maitra M, Starcevic D, Li SX, Sweasy JB (2004) A DNA polymerase beta mutant from colon cancer cells induces mutations. *Proc Natl Acad Sci U S A* 101: 6074–6079.
 50. Wang L, Patel U, Ghosh L, Banerjee S (1992) DNA polymerase β mutations in human colorectal cancer. *Cancer Research* 52: 4824–4827.
 51. Lang T, Dalal S, Chikova A, Dimaio D, Sweasy JB (2007) The E295K DNA polymerase beta gastric cancer-associated variant interferes with base excision repair and induces cellular transformation. *Mol Cell Biol* 27: 5587–5596.
 52. Li Y, Gridley CL, Jaeger J, Sweasy JB, Schlick T (2012) Unfavorable electrostatic and steric interactions in DNA polymerase beta E295K mutant interfere with the enzyme's pathway. *J Am Chem Soc* 134: 9999–10010.
 53. Donigan KA, Sun KW, Nemeec AA, Murphy DL, Cong X, et al. (2012) Human POLB gene is mutated in high percentage of colorectal tumors. *Journal of Biological Chemistry* 287: 23830–23839.