Mitochondrial DNA polymerase γ (pol γ) is active in base excision repair of AP (apurinic/apyrimidinic) sites in DNA. Usually AP site repair involves cleavage on the 5′ side of the deoxyribose phosphate by AP endonuclease. Previous experiments suggested that DNA pol γ acts to catalyze the removal of a 5′-deoxyribose phosphate (dRP) group in addition to playing the conventional role of a DNA polymerase. We confirm that DNA pol γ is an active dRP lyase and show that other members of the family A of DNA polymerases including Escherichia coli DNA pol I also possess this activity. The dRP lyase reaction proceeds by formation of a covalent enzyme-DNA intermediate that is converted to an enzyme-dRP intermediate following elimination of the DNA. Both intermediates can be cross-linked with NaBH₄. For both DNA pol γ and the Klenow fragment of pol I, the enzyme-dRP intermediate is extremely stable. This limits the overall catalytic rate of the dRP lyase, so that family A DNA polymerases, unlike pol β, may only be able to act as dRP lyases in repair of AP sites when they occur at low frequency in DNA.

Abasic (AP) sites in DNA are produced frequently by spontaneous base loss or by the action of DNA glycosylases that initiate base excision repair of damaged bases in DNA (1, 2). If AP sites are not repaired quickly, DNA polymerases are capable of replicating through these non-instructional lesions, resulting in frequent misincorporation. Cells have adapted vigorous mechanisms for the repair of AP sites, usually beginning with incision of the phosphodiester backbone on the 5′ side of the lesion by AP endonuclease to produce a 3′-OH terminus adjacent to a 5′-deoxyribose phosphate, or 5′-dRP group (3, 4). The 3′-OH terminus provides a primer for repair synthesis by DNA polymerase. The 5′-dRP moiety must be removed in the course of repair. This is frequently accomplished by a β-elimination reaction catalyzed by an AP lyase activity. When an AP lyase acts on an exposed 5′-dRP residue produced by a class II AP endonuclease, the activity may be considered as a dRP lyase. The dRP lyase mechanism involves nucleophilic attack on the C-1 position of the 5′-dRP group by a free amino group of the enzyme (5). This produces a transient covalent intermediate in which the DNA substrate is linked to the enzyme as a Schiff base that can be stabilized by treatment with strong reducing agents, such as sodium borohydride. This borohydride trapping reaction has been used to identify a number of DNA repair enzymes with AP lyase activity, including several DNA glycosylases (6, 7).

Eukaryotic cells have redundant pathways for repairing AP sites in nuclear DNA. Under most circumstances, AP sites are repaired by a pathway that employs the dedicated repair polymerase, DNA pol β. Recently, Matsumoto and Kim (8) showed that the pol β is especially well adapted to function in base excision repair, since it contains a dRP lyase activity in a small domain not required for polymerase activity. The active site of the pol β dRP lyase has been localized to a helix-hairpin-helix domain similar to that found in repair glycosylases with associated AP lyase activity (7–10). Thus, pol β is capable of binding to an incised AP site and employing its polymerase and dRP lyase activities in concerted reactions to prepare the DNA for ligation to complete the repair reaction. This mechanism permits repair to be accomplished with a single base patch size. However, in some instances, pol β may participate in longer patch repair (11). The generation of cell lines devoid of pol β activity implies that pol β is not absolutely indispensable for base excision repair (12). Our laboratory and others have shown that another polymerase, either pol δ or ε, can function in a repair pathway that employs PCNA and the 5′ flap endonuclease, FEN I (13–16).

We recently attempted to characterize enzymes capable of acting in base excision repair of lesions in mtDNA. We constituted a complete pathway for repair of AP sites using highly purified mitochondrial enzymes (17). In the course of these experiments, we found that both mtDNA pol γ and mtDNA ligase were active in a borohydride trapping assay. This represented the first observation of potential dRP lyase activity in a DNA polymerase other than DNA pol β, and the first observation of an AP lyase activity in a DNA ligase. We found that other ATP-dependent DNA ligases, including T4 and T7 DNA ligases, also contain AP lyase activity (18).

In this paper, we present additional experiments to characterize the dRP lyase activity in DNA pol γ in greater detail. Since pol γ is a member of the family A group of DNA polymerases (22, 23), we also tested other members of this family for dRP lyase activity. All family members tested, including DNA pol I, T7 DNA polymerase, and Moloney murine leukemia virus reverse transcriptase are active in borohydride trapping reactions at dRP sites. The family A DNA polymerases appear to initiate attack at dRP sites quickly, but are slow to complete the β-elimination of the DNA from the dRP group, leading to a very low turnover rate for the overall reaction. We show that the AP lyase activity in DNA pol γ and the Klenow fragment of DNA polymerase I is sufficient to permit these enzymes to function in base excision repair in the absence of other detectable sources of AP lyase activity.
EXPERIMENTAL PROCEDURES

Materials—All chemicals used were reagent grade. Nucleoside triphosphates were obtained as HPLC purified reagents from Amersham Pharmacia Biotech. Radioactively labeled nucleotides were obtained from ICN Radiochemicals. Micrococcal nuclease was obtained from New England Biolabs. Uracil DNA glycosylase (UNG), was from Epicentre Technologies. Moloney murine leukemia virus reverse transcriptase (MMLV RT) was obtained from Life Technologies. DNA pol I and the Klenow fragment were obtained from Roche Molecular Biochemicals. T7 DNA polymerase was obtained from Amersham Pharmacia Biotech. Escherichia coli endonuclease IV was obtained from Trevigen. E. coli DNA ligase preparations, from Life Technologies, New England Biolabs, and Roche Biochemicals, were screened for dRP lyase activity. None of these preparations were active in borohydride trapping. The preparation from Roche Molecular Biochemicals was selected for repair assays. DNA pol γ was purified from an Xenopus laevis oocyte homogenate as described (19, 20). The preparation used in this study had a specific activity of 30 units/ml where 1 unit corresponds to incorporation of 1 nmol of TMP on a poly(dA)-oligo(dT) template-primer in 60 min at 30 °C. Mitochondrial APE was prepared as described (17). Recombinant E. coli FPG protein (also known as formamidopyrimidine glycosylase or as 8-oxo-guanine glycosylase) was a gift from Drs. J. Tchou and A. P. Grollman. Recombinant rat DNA pol β was a gift from Dr. Y. Matsumoto (Fox Chase Cancer Center). All commercial DNA polymersases used in this study were analyzed by SDS-PAGE in parallel with quantitative protein standards of similar molecular weight. Densitometric analysis of the Coomassie Blue-stained gel was used to determine the approximate protein concentration.

Oligonucleotides—Oligonucleotides were synthesized on a Beckman oligonucleotide synthesizer or were obtained from Operon Technologies. All oligonucleotides were purified by preparative gel electrophoresis. Duplex oligonucleotides were prepared by heating to 70 °C in a solution containing 10 mM Tris, pH 8.0, 0.1 mM EDTA and either 0.1 or 0.2 mM NaCl and slowly cooling to room temperature over at least 3 h. The oligonucleotide substrate used for borohydride trapping reactions was prepared by annealing a 15-mer (5′-CATGGGCCGACATG) and a 5′-cleotide substrate used for borohydride trapping reactions was pre-

![Fig. 1. Borohydride trapping of family A DNA polymerases to oligonucleotides bearing AP sites. A duplex oligonucleotide containing a 5′-phosphorylated U residue at an internal nick was prepared and pretreated with HK-UNG as described under “Experimental Procedures.” 50 fmoles of oligonucleotide was added to 10-μl binding reactions with approximately 100 fmoles of DNA pol γ (lanes 1 and 2), DNA pol I (lanes 3 and 4), Klenow fragment of pol I (lanes 5 and 6), MMLV reverse transcriptase (lanes 7 and 8), T7 DNA pol (lanes 9 and 10), or DNA pol β (lanes 11 and 12). Reactions were assembled on ice and 20 mM NaBH₄ was added before reactions were transferred to a water bath at 25 °C. Reactions were incubated at 25 °C for 30 min. One-half of each reaction was stopped by addition of SDS sample loading buffer (odd numbered lanes), the remaining half was treated with micrococcal nuclease (MN) as described under “Experimental Procedures.” Proteins were precipitated with 10% trichloroacetic acid and fractionated by SDS-PAGE. A PhosphorImager analysis of the dried gels is shown. The numbers on the left of each panel indicate the molecular masses (kd) of prestained protein mobility markers.

RESULTS

dRP Lyase Activity of DNA Polymerases—We tested the abilities of a variety of family A DNA polymerases to react with an oligonucleotide substrate containing a 5′-32P-labeled dRP residue at an internal nick. This oligonucleotide substrate was prepared immediately before incubation with DNA polymerase by treatment of an oligonucleotide containing a single uracil residue with uracil DNA glycosylase (UDG). The resulting oligonucleotide contains an internal 5′-32P[dRP] residue in a structure identical to that expected following incision of an AP site by a class II AP endonuclease. Fig. 1 shows that DNA pol γ, DNA pol I, the pol I Klenow fragment, MMLV RT, T7 DNA polymerase, and DNA pol β are active in the borohydride trapping assay. In each case, the primary trapped product (odd numbered lanes) has a slower gel mobility than the unmodified polymerase due to cross-linking to the oligonucleotide. Treatment with micrococcal nuclease generates a product with essentially the same mobility as the unmodified polymerase (even numbered lanes). DNA pol β represents a positive control for these reactions, since this enzyme is known to contain dRP lyase activity. The additional mass of the oligonucleotide makes a larger contribution to the mobility of a small polymerase like pol β than to the larger polymerases. We have repeatedly seen that cross-linking the MMLV RT preparation gives a doublet of retarded protein bands. We have not explored the basis for this different pattern of reactivity. We conclude that all of the polymerases used in the experiment in Fig. 1 are capable of reacting with 5′-dRP residues in DNA to form a Schiff base that can be reduced with NaBH₄.

To study the kinetics and mechanism of the dRP lyase reaction in greater detail, we selected pol γ and Klenow pol for additional experiments to compare the kinetics of borohydride cross-linking with these enzymes to that observed for pol β as...
DNA polb was incubated with DNA to enzyme-dRP intermediates. The mobility and mass in kDa of the unmodified DNA polymerases as visualized by staining. The time course of action of DNA polymerases at AP sites suggests the dRP lyase reaction proceeds from enzyme-DNA to enzyme-dRP intermediates. For polγ, Klenow pol incubated at 37 °C; polβ, incubated at 30 °C; polβ, incubated at 10 °C.

A positive control. We sought to determine whether the yield of cross-linked product would be improved if the DNA polymerase were preincubated with the oligonucleotide for varied intervals before addition of NaBH₄. The timing of borohydride-trapping reactions with AP sites is of critical importance since NaBH₄ can also reduce the 5'-dRP substrate, effectively preventing further reaction with an dRP lyase. The kinetics of the borohydride trapping reactions are shown in Fig. 2. All three DNA polymerases initiated an attack on the substrate within 5 min. At the earliest time points, two radioactive cross-linked products were observed, a lower band with the same mobility as the unmodified protein and an upper band with the mobility of the enzyme-oligonucleotide complex. Preincubation of pol β with the substrate at 37 °C resulted in a loss of borohydride-trapped product after the first 30 min. This is predicted for an enzyme with a potent dRP lyase activity that efficiently resolves the dRP enzyme intermediate. The lack of labeling at later times reflects the fact that pol β has consumed all of the substrate. Pol γ and Klenow pol clearly have a much lower overall turnover rate. Both enzymes facilitate β-elimination of the DNA from the enzyme-oligonucleotide complex, leading to loss of the upper band in the cross-linking reaction. The persistence of the lower band implies that these enzymes do not release the dRP product as efficiently as does pol β.

We performed a more detailed analysis of the kinetics of the dRP lyase mechanism in an additional experiment in which we monitored the disappearance of the substrate oligonucleotide, the appearance of the borohydride-trapped polymerase intermediates and the generation of an ethanol-soluble product. The results in Fig. 4 document the disappearance of the free labeled oligonucleotide substrate concomitant with the appearance of the labeled enzyme-DNA and later of the enzyme-dRP intermediates. Free [132P]dRP is released slowly as the final product of the reaction. A similar reaction rate was observed with pol γ (data not shown). In experiments performed with DNA pol β we have observed the same sequence of events, but at an accelerated pace (Fig. 5; note the change in time scale compared with Fig. 4). In reactions in which 400 fmol of oligonucleotide was incubated with either 100 or 1000 fmol of DNA pol β, release of the internal 5'-[32P]dRP oligonucleotide for 5, 15, 30, or 60 min (lanes 1–4, respectively) before addition of 20 mM NaBH₄. Following an additional 15-min incubation at room temperature, the proteins were analyzed by SDS-PAGE. Gels were stained with Coomassie Blue, dried, and exposed to a PhosphorImager. Panel A, DNA pol γ incubated at 30 °C; B, Klenow pol incubated at 37 °C; C, DNA pol β incubated at 37 °C; D, DNA pol β incubated at 10 °C. Arrows on the left of panels A–D indicate the mobility and mass in kDa of the unmodified DNA polymerases as visualized by staining.

The initial substrate contains a 15-mer and a 5' -end labeled 17-mer annealed to a complementary strand. Treatment with UDG generates a labeled 5'-dRP group adjacent to a nick, as would result from action of AP endonuclease. Binding of DNA pol (enzyme) permits attack on the C1 residue of the dRP group as shown. This overall scheme is adapted from Sun et al. (10) and Zharkov et al. (30). The β-elimination reaction involves the primary attack to form the Schiff base intermediate and also abstraction of a proton by a second nucleophilic center in the enzyme (enz) to promote elimination of the DNA. Both enzyme-DNA and enzyme-dRP intermediates are capable of reaction with NaBH₄, which releases the dRP product. 

![Fig. 2](http://www.jbc.org/Downloaded from http://www.jbc.org/) The time course of action of DNA polymerases at AP sites suggests the dRP lyase reaction proceeds from enzyme-DNA to enzyme-dRP intermediates. DNA pol was incubated with the internal 5'-[32P]dRP oligonucleotide for 5, 15, 30, or 60 min (lanes 1–4, respectively) before addition of 20 mM NaBH₄. Following an additional 15-min incubation at room temperature, the proteins were analyzed by SDS-PAGE. Gels were stained with Coomassie Blue, dried, and exposed to a PhosphorImager. Panel A, DNA pol γ incubated at 30 °C; B, Klenow pol incubated at 37 °C; C, DNA pol β incubated at 37 °C; D, DNA pol β incubated at 10 °C. Arrows on the left of panels A–D indicate the mobility and mass in kDa of the unmodified DNA polymerases as visualized by staining.

![Fig. 3](http://www.jbc.org/Downloaded from http://www.jbc.org/) Proposed scheme for the dRP lyase reaction. The initial substrate contains a 15-mer and a 5'-end labeled 17-mer annealed to a complementary strand. Treatment with UDG generates a labeled 5'-dRP group adjacent to a nick, as would result from action of AP endonuclease. Binding of DNA pol (enzyme) permits attack on the C1 residue of the dRP group as shown. This overall scheme is adapted from Sun et al. (10) and Zharkov et al. (30). The β-elimination reaction involves the primary attack to form the Schiff base intermediate and also abstraction of a proton by a second nucleophilic center in the enzyme (enz) to promote elimination of the DNA. Both enzyme-DNA and enzyme-dRP intermediates are capable of reaction with NaBH₄, which releases the dRP product. 

![Fig. 4](http://www.jbc.org/Downloaded from http://www.jbc.org/) The time course of action of DNA polymerases at AP sites suggests the dRP lyase reaction proceeds from enzyme-DNA to enzyme-dRP intermediates. DNA pol was incubated with the internal 5'-[32P]dRP oligonucleotide for 5, 15, 30, or 60 min (lanes 1–4, respectively) before addition of 20 mM NaBH₄. Following an additional 15-min incubation at room temperature, the proteins were analyzed by SDS-PAGE. Gels were stained with Coomassie Blue, dried, and exposed to a PhosphorImager. Panel A, DNA pol γ incubated at 30 °C; B, Klenow pol incubated at 37 °C; C, DNA pol β incubated at 37 °C; D, DNA pol β incubated at 10 °C. Arrows on the left of panels A–D indicate the mobility and mass in kDa of the unmodified DNA polymerases as visualized by staining.

A positive control. We sought to determine whether the yield of cross-linked product would be improved if the DNA polymerase were preincubated with the oligonucleotide for varied intervals before addition of NaBH₄. The timing of borohydride-trapping reactions with AP sites is of critical importance since NaBH₄ can also reduce the 5'-dRP substrate, effectively preventing further reaction with an dRP lyase. The kinetics of the borohydride trapping reactions are shown in Fig. 2. All three DNA polymerases initiated an attack on the substrate within 5 min. At the earliest time points, two radioactive cross-linked products were observed, a lower band with the same mobility as the unmodified protein and an upper band with the mobility of the enzyme-oligonucleotide complex. At later times, the intensity of the upper band diminished while the intensity of the lower band increased. This behavior is consistent with the proposed reaction scheme shown in Fig. 3, which suggests that the dRP lyase reaction proceeds from an enzyme-DNA to an enzyme-dRP complex. NaBH₄ is capable of reducing both intermediates to trap the label on the protein. It is important to recognize that, unlike the experiment in Fig. 1, micrococcal nuclease was not employed in the experiment in Fig. 2. Thus, the conversion from the species with lower mobility to that of higher mobility represents the natural course of the reaction. For pol β, which has a very active dRP lyase, it was necessary to perform the reaction at a reduced temperature to document the upper band.
soluble dRP product is essentially complete in 10 min. Interestingly, the enzyme-DNA intermediate is more easily visualized by borohydride trapping in reactions containing a higher relative concentration of oligonucleotide (panel B) than in those containing excess pol β (panel C). The persistence of the enzyme-DNA intermediate is apparent due to the use of substrate-excess conditions. A major difference between the dRP lyase reaction catalyzed by pol β and that of Klenow pol is that the release of the dRP residue from the enzyme is a very slow rate-limiting step for Klenow pol.

We performed additional experiments with pol γ and Klenow pol to characterize the ethanol soluble product of the reaction. When the dRP lyase reaction is conducted in the presence of thioglycolate, an anionic species is generated that has characteristic chromatographic properties on anion exchange HPLC (8, 24). Fig. 6 shows that the ethanol soluble products generated by DNA pol γ and Klenow pol in the presence of thioglycolate have the same chromatographic properties as the species produced by a well characterized AP lyase, E. coli FPG protein.

We conclude that both of these enzymes are capable of acting as authentic dRP lyase enzymes to catalyze release of a 5’ dRP residue at an incised AP site. However, additional experiments revealed that the overall rate of catalysis by these DNA polymerases is quite limited. Despite extensive efforts to determine a turnover rate for the AP lyase in Klenow pol we have not been able to document a release of more than 0.7 dRP group per enzyme molecule in reactions incubated for as long as 1 h.

Is the dRP Lyase in Pol γ and Klenow Pol Sufficient for Complete Repair of AP Sites?—The experiments presented above show that our preliminary report of borohydride trapping activity in pol γ (17) and the related activity in Klenow pol do represent authentic dRP lyase activity. Our earlier experiments did not permit the conclusion that the dRP lyase activity completing repair of an AP site in E. coli DNA. Our current experiments with thioglycolate indicate that this may be the case.
in pol γ was sufficient for repair in the absence of other sources of dRP lyase since we observed that the mtDNA ligase was also a potential source of AP lyase activity. We performed repair experiments to test whether the dRP lyase activity in DNA pol γ or Klenow pol is sufficient for repair in reactions in which all other enzymes employed in the repair reaction lack AP lyase activity. Control experiments with the uracil glycosylase, mtAPE, endonuclease IV, and E. coli DNA ligase used in these repair reactions failed to show any indication of dRP lyase activity as assessed by borohydride trapping or dRP release assays (data not shown).

In the repair experiments in Fig. 7, we used a closed circular DNA substrate containing a single U residue. The substrate was prelabeled with $^{32}$P at a single site 5 base pairs preceding the lesion to permit us to follow the course of the repair reaction. One labeled substrate was used for two separate sets of reactions using different sources of DNA polymerase and AP endonuclease. In reaction A, the template was incised using mitochondrial APE and repaired using DNA pol γ. In reaction B, the template was incised using E. coli endonuclease IV and repaired using the Klenow fragment of DNA pol I. In each case, the complete repair reactions included E. coli DNA ligase. The intermediate and final products were cleaved at Hinfl sites flanking the lesion to permit a detailed analysis of the intermediates and products of the repair reaction following electrophoresis of the fragments on a 20% PAGE-urea gel. The gel analysis of the treated samples is shown in Fig. 7. The substrate was efficiently incised by either mitochondrial APE (lane A2) or E. coli endonuclease IV (lane B6). The incised DNAs were then incubated either with E. coli DNA ligase alone (lanes A3 and B7), with polymerase alone (DNA pol γ in lane A4, Klenow pol in lane B8), or with both E. coli DNA ligase and the appropriate polymerase (lanes A5 and B9). Complete repair was observed in reactions including AP endonuclease, either pol γ or Klenow pol, and E. coli DNA ligase. We conclude that both pol γ and Klenow pol can participate in repair reactions in which they provide the only detectable source of dRP lyase.

**DISCUSSION**

We initially reported that DNA pol γ was active in a borohydride trapping reaction as a preliminary observation in a study of the overall base excision repair reaction conducted by mitochondrial proteins (17). Subsequent to this, Longley et al. (25) published a more thorough characterization of the dRP lyase reaction of recombinant human DNA pol γ. The dRP lyase assay employed by Longley et al. (25) monitored the shift in mobility of a 3′-labeled oligonucleotide upon release of the 5′-dRP group. Our current work provides the first evidence that a DNA pol γ liberates a product in the presence of thioglycolic acid with the chromatographic properties characteristic of a dRP release product (Fig. 6). Longley et al. (25) concluded that the rate of the overall dRP lyase reaction was markedly slower for pol γ than for pol β. Our current work suggests that the rate-limiting step in this reaction is the slow release of the dRP group from the enzyme.
Since DNA pol γ shares significant sequence homology with other members of the family A DNA polymerases, we asked whether other enzymes in this class contained a similar dRP lyase activity. The experiment in Fig. 1 shows that three other family A DNA polymerases were readily labeled by borohydride trapping reactions with the appropriate AP site substrate. We selected the Klenow fragment of DNA pol I for more extensive studies and found that this enzyme, like pol γ, has a very low catalytic rate limited by slow release of the dRP group from the enzyme. We note that there is at least one precedent in the case of E. coli mutY protein for a bona fide AP lyase that exhibits a very slow release of the dRP product (26).

A recurrent issue in the literature on AP lyase activities is the concern that the β-elimination of a 5′-dRP group is a facile reaction that occurs at a measurable rate in the absence of enzymes. Moreover, release can be accelerated by binding of nonspecific basic proteins and even by simple peptides or by elevated pH. One standard that is applied to ask whether an AP lyase activity is authentic is to determine whether it depends on native enzyme structure. We found that both DNA pol γ and Klenow pol are inactive in borohydride trapping assays following thermal denaturation (data not shown). Nevertheless, it may be argued that the rather languid lyase reaction of family A DNA polymerases indicates that this is not an important activity. However, it is clear that DNA polymerases bind nicked AP sites avidly and initiate attack on the C1′ residue of the dRP group rapidly. Once the reaction has been initiated in this manner, it is only the subsequent mechanistic step of product release that is kinetically slow. This slow overall reaction indicates that family A DNA polymerases would be unlikely to handle a large load of AP site damage successfully without the aid of other sources of AP lyase activity. For wild type E. coli, the catalytic inefficiency of the polymerase-associated dRP lyase may be of little consequence, since this organism contains the mutM gene product as an alternative source of AP lyase. The polymerase-associated dRP lyase may be a factor in the survival of E. coli bearing mutM and recE mutations (27). It is possible that mitochondria contain additional sources of AP lyase activity that may contribute to efficient repair of AP sites in vivo or in crude extracts (28).

Our observation that E. coli DNA pol I has an associated dRP lyase activity may appear to be at odds with the classical work of Mosbaugh and Linn (29) on the mechanism of action of E. coli DNA pol I at incised AP sites in DNA. These workers found that DNA pol I did not appear to act efficiently at incised AP sites generated by random depurination of PM2 DNA. This early work did not involve studies of site-specific lesions. Since the lyase mechanism for removal of dRP groups was not appreciated at the time this work was done, these authors were more concerned with the question of whether the 5′-3′ exonuclease of DNA pol I would remove a 5′-dRP group. They concluded that the exonuclease activity was not able to act on the 5′-dRP group at an incised AP site, and that the polymerase was able to engage in strand displacement replication on these templates. Our results suggest that the dRP lyase activity in DNA pol I may permit the enzyme to remove a limited number of 5′-dRP residues and that the strand displacement reported by Mosbaugh and Linn (29) may be increasingly common on templates with a high frequency of AP sites. Thus, the alternate sources of AP lyase in E. coli mentioned above clearly have an important role to limit the number of instances in which pol I is required to act as a dRP lyase.

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Characterization of a Catalytically Slow AP Lyase Activity in DNA Polymerase γ and Other Family A DNA Polymerases
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