Biol. Chem., Vol. 383, pp. 969–975, June 2002 · Copyright © by Walter de Gruyter · Berlin · New York

Minireview

DNA Binding Properties of the Yeast Msh2-Msh6 and Mlh1-Pms1 Heterodimers

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We describe here our recent studies of the DNA binding properties of Msh2-Msh6 and Mlh1-Pms1, two protein complexes required to repair mismatches generated during DNA replication. Mismatched DNA binding by Msh2-Msh6 was probed by mutagenesis based on the crystal structure of the homologous bacterial MutS homodimer bound to DNA. The results suggest that several amino acid side chains inferred to interact with the DNA backbone near the mismatch are critical for repair activity. These contacts, which are different in Msh2 and Msh6, likely facilitate stacking and hydrogen bonding interactions between side chains in Msh6 and the mismatched base, thus stabilizing a kinked DNA conformation that permits subsequent repair steps coordinated by the Mlh1-Pms1 heterodimer. Mlh1-Pms1 also binds to DNA, but independently of a mismatch. Mlh1-Pms1 binds short DNA substrates with low affinity and with a slight preference for single-stranded DNA. It also binds longer duplex DNA molecules, but with a higher affinity indicative of cooperative binding. Indeed, imaging by atomic force microscopy reveals cooperative DNA binding and simultaneous interaction with two DNA duplexes. The novel DNA binding properties of Mlh1-Pms1 may be relevant to signal transduction during DNA mismatch repair and to recombination, meiosis and cellular responses to DNA damage.

Key words: DNA binding / Mismatch repair / Mlh1 / Msh2 / Msh6 / Pms1.

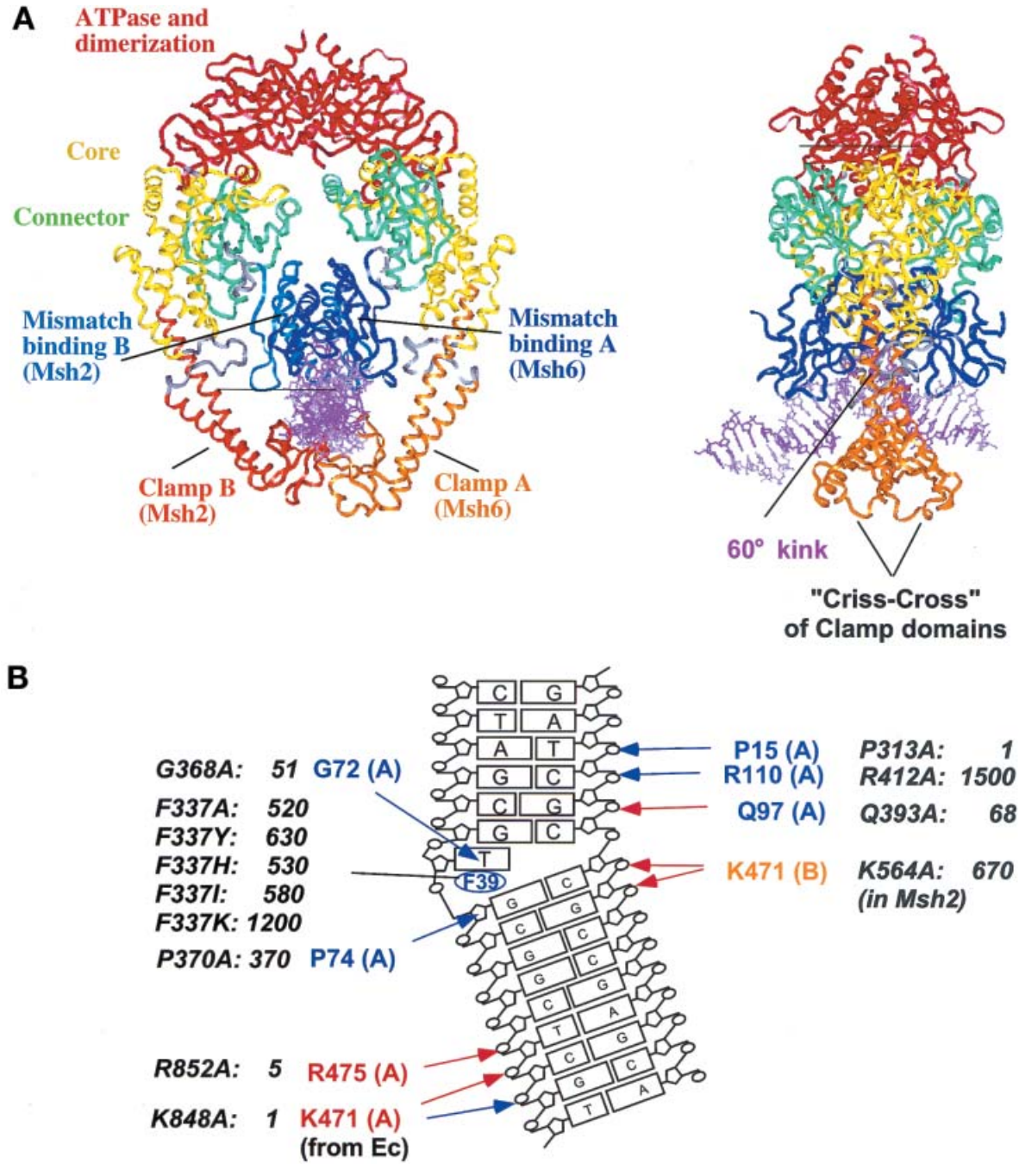
Introduction

DNA mismatch repair (MMR) contributes to genome stability by correcting DNA replication errors and by preventing recombination between similar but diverged DNA sequences (recently reviewed in Buermeyer *et al.*, 1999; Kolodner and Marsischky, 1999; Harfe and Jinks-Robertson, 2000; Hsieh, 2001). In the most thoroughly characterized *Escherichia coli* MMR system, which serves as a paradigm for eukaryotic MMR studies, initiation of MMR requires the products of *mutS*, *mutL* and *mutH* genes. A homodimer of MutS binds to mismatches in DNA and a homodimer of MutL coordinates mismatch recognition with the downstream steps in MMR that include nicking of the nascent DNA strand by MutH endonuclease, excision of the mismatch and resynthesis of the strand. In eukaryotes, MMR requires multiple MutS and MutL homologs that form several functionally distinct heterodimeric complexes. Two MutS-related heterodimers, Msh2-Msh6 and Msh2-Msh3, are responsible for mismatch recognition while at least two MutL-related heterodimers, Mlh1-Pms1 (MLH1-PMS2 in humans) and Mlh1-Mlh3, are thought to play a role in coordination of downstream MMR events, similar to *E.coli* MutL.

One of the key requirements for MMR is the capacity of MutS and its eukaryotic homologs to bind to mismatched DNA with higher specificity than to matched DNA. In addition, the *E.coli* MutL protein has also been shown to bind to DNA in a mismatch-independent manner (Bende and Grafstrom, 1991; Ban and Yang, 1998; Ban *et al.*, 1999; Mechanic *et al.*, 2000). This DNA binding capacity of MutL is not yet understood in the context of most current MMR models (Jiricny, 1998; Kolodner and Marsischky, 1999; Harfe and Jinks-Robertson, 2000). Moreover, it has become clear in recent years that eukaryotic MutS and MutL homologs participate in a variety of other DNA transactions. These include DNA damage recognition, cell cycle checkpoint control and apoptosis, transcription-coupled nucleotide excision repair, meiotic chromosome pairing and crossing-over, and possibly somatic hypermutation of immunoglobulin genes (reviewed in Buermeyer *et al.*, 1999; Harfe and Jinks-Robertson, 2000). Such widespread involvement of eukaryotic MutS and MutL homologs in DNA metabolic processes illustrates the importance of understanding how they interact with DNA. This article summarizes our recent studies of DNA binding by yeast Msh2-Msh6 (Drotschmann *et al.*, 2001) and yeast Mlh1-Pms1 (Hall *et al.*, 2001), as presented in October, 2001, at the 8th International Conference on Envionmental Mutagens in Shizuoka, Japan.

DNA Binding by Yeast Msh2-Msh6

Our studies of the DNA binding properties of yeast Msh2-Msh6 (Drotschmann *et al.*, 2001) were directed by the X-ray crystal structures of *Taq* and *E. coli* MutS proteins bound to mismatched DNA (Lamers *et al.*, 2000; Obmolova *et al.*, 2000). The MutS homodimer contains two identical polypeptide chains designated subunits A and B (Figure 1A). Each bacterial MutS subunit contains five discrete domains (colored differently in Figure 1A). Only two of these domains interact with DNA. In subunit A, amino acids in domain I (mismatch binding, in blue) and domain IV (clamp domain, in orange) interact in a sequence-independent manner with the DNA backbone *via*



van der Waals interactions or hydrogen bonding. The amino acid side chains in the mismatch binding domain of subunit A also contribute base-specific interactions, especially including stacking of a phenylalanine ring (F39 in *Taq* MutS, conserved as F337 in yeast Msh6, but not conserved in yeast Msh2) with the mismatched thymine. In contrast to the extensive interactions of subunit A with DNA, there are fewer interactions with subunit B, and none of these are base specific. Of special interest here is a lysine residue (K471 in *Taq* MutS, conserved as K564 in yeast Msh2) that interacts with the DNA backbone near the region where the DNA is kinked by 60°. Discrimination between homoduplex and heteroduplex DNA is thought to reflect an inherently higher deformability of duplex DNA at the site of a mismatch, such that binding induces a 60° kink in the DNA (Figure 1A, right) that can be stabilized by mismatched base-specific interactions.

This structural information indicates that the MutS homodimer binds to DNA in an asymmetric manner, *i.e.*, it is actually a functional heterodimer. The hypothesis implied by this structure and by amino acid sequence alignments is that Msh2 is functionally equivalent to subunit B of the MutS homodimer, while Msh6 is functionally equivalent to subunit A. To test this hypothesis, we substituted alternative amino acids for residues in yeast Msh6 and Msh2 that were inferred to be important for DNA binding based on amino acid sequence conservation with *Taq* MutS (Figure 1C; for additional details, see Drotschmann *et al.*, 2001).

Inferred Interactions of Msh2-Msh6 with the DNA Backbone

Figure 1B shows several amino acids in *Taq* MutS that contact the DNA backbone by Van der Waals interactions (blue arrows) and hydrogen bonds (red arrows). Homologous residues in yeast Msh2 and Msh6 were individually changed to alanine, and the ability of the mutants to complement the mutator phenotype of *msh2* or *msh3/6* mutant yeast strains was monitored by expression of the mutant genes from their natural promoter on single-copy plasmids. For this purpose, we used a reporter system that detects one base pair deletions in an A_{14} run in the *LYS2* gene. In comparison to expression of the wild-type Msh2 and Msh6 genes, elevated mutation rates characteristic of loss of MMR were observed upon expression of several of the mutant genes (Figure 1B). The results suggest that residues in the mismatch-binding domain of Msh6 and the clamp domain of Msh2 provide important sequence-independent contacts to the backbone. Note that the *msh6* mutations that give strong mutator phenotypes affect residues that are not conserved in Msh2. Also noteworthy is the fact that a strong mutator effect is conferred by the K564A substitution in the clamp domain of Msh2, whereas the homologous K848A mutant in Msh6 yields a wild type mutation rate. Importantly, changing side chain contacts with the sugar-phosphate backbone at and near the mismatch confer strong mutator effects, whereas putative more distant contacts have little effect.

A Mismatch-Specific Interaction of Msh2-Msh6 with DNA

Phe39 in the mismatch-binding domain of subunit A of *Taq* MutS is one of only two amino acids providing base-specific contacts with the mismatch. The side chain is observed to stack with the aromatic ring of the mismatched thymine base (Lamers *et al.*, 2000; Obmolova *et al.*, 2000). When the homologous residue in Msh6 (F337) is changed to any of five other amino acids, a strong mutator effect is observed (Figure 1B). In contrast, the mutation rate for the homologous Y42A mutant in Msh2 is only slightly elevated (9-fold, compared to 1000-fold of *msh2* strain, not shown).

Because mutator effects *in vivo* can result from inactivation of mismatch repair due to any of several defects in MutS homologs, we also tested the ability of purified heterodimers to bind to DNA *in vitro*. As an example, purified Msh2-Msh6 with either Ala or Tyr substituted for Phe337 binds to DNA with lower affinity and with lower discrimination between matched and mismatched DNA (Figure 2; also see Drotschmann *et al.*, 2001). These results are consistent with a number of studies by others (Hughes and Jiricny, 1992; Malkov *et al.*, 1997; Iaccarino *et al.*, 1998; Bowers *et al.*, 1999; Dufner *et al.*, 2000; Yamamoto

◀ **Fig. 1** Structure-Based Analysis of Mismatched DNA Binding by Yeast Msh2-Msh6.
Panel (A), left part: crystal structure of MutS from *Thermus aquaticus* in complex with heteroduplex DNA containing an unpaired thymine (adapted from Obmolova *et al.*, 2000). Individual domains are highlighted by different colors and labeled according to their inferred function. The right panel shows the same structure but rotated 90° along the vertical axis. The 60° kink introduced into DNA at the side of the mismatch is shown, as well as the 'criss-cross' of the clamp domains that embrace the DNA.
Panel (B): schematic representation of contacts between the mismatched DNA and amino acid residues in Taq MutS (colored) or inferred in yeast Msh2 and Msh6 (black; adapted from Drotschmann *et al*., 2001). Colors correspond to domain structure shown in panel (A), with residues in the mismatch binding domain I of subunit A in blue, residues in the clamp domain IV of subunit A in red, and residues from the clamp domain IV of subunit B in orange. Blue arrows represent Van der Waals interactions, and red arrows show hydrogen bonds. Mutator effects of amino acid changes to alanine (unless otherwise stated) in the homologous yeast Msh6 or Msh2 ('Msh') protein are given as relative rates in reference to expression of wild type *MSH6* or *MSH2* in an *msh3 msh6* or *msh2* strain, respectively, using a single-copy plasmid. The actual mutation rates for all variables are given in Drotschmann *et al.* (2001). 'Ec' in parenthesis refers to a comment only found in the *E. coli* MutS-DNA co-crystal.
Panel (C): alignment of TaqMutS and yeast Msh2 and Msh6 in the regions containing DNA-interaction residues. Coloring as shown in panel (B); mutations introduced are depicted underneath the respective amino acid (adapted from Drotschmann *et al*., 2001).

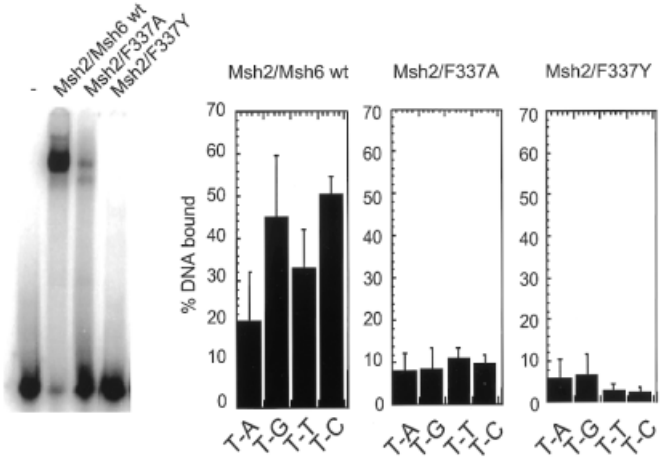


Fig. 2 Reduced DNA Binding by Altering a Side Chain Interaction with the Mismatched Base.
DNA binding activity of wild type and mutant yMsh2-yMsh6 heterodimer. 100 nM protein was incubated with 10 nM DNA, either homoduplex or heteroduplex DNA (34-mers) as indicated. Binding was visualized by retardation of bands on gels, and analyzed using PhosphoImager and ImageQuant software. The gel shows examples of differential binding to a +T heteroduplex substrate by wild type and mutant proteins. Quantitative analysis of binding to different substrates by wild type and mutant proteins contains mean values and standard deviations for at least three individual experiments.

et al., 2000; DasGupta and Kolodner, 2000). Collectively, they demonstrate the importance of this phenylalanine for mismatch repair in both bacterial and eukaryotic MutS proteins.

As predicted by the MutS structure, the overall effects described here and in other studies (cited in Drotschmann *et al.*, 2001) clearly imply asymmetric interactions of Msh6 and Msh2 with DNA, in which Msh6 and Msh2 correspond to subunits A and B of bacterial MutS, respectively. This binding asymmetry involves mismatched base-specific stacking with the phenylalanine as well as DNA backbone contacts involving amino acids in Msh6 that are not conserved in Msh2 and a lysine in Msh2 whose homolog in Msh6 is functionally less significant. In addition to this phenylalanine, there is a second mismatch-specific interaction involving a conserved glutamate side chain. Analysis of the importance of this residue in bacterial MutS and in yeast Msh6 can be found in (Schofield *et al.*, 2001) and (Drotschmann *et al.*, 2001), respectively.

Substitution of side chains inferred to interact within 2–3 base pairs on either side of the mismatch have a stronger effect on mutation rate than those interacting with DNA further away. This implies that local interactions with the backbone around the mismatch may facilitate a kinked DNA conformation, as observed in bacterial MutS-DNA complexes. This kinked DNA conformation may be critical for facilitating downstream events in mismatch repair, as has been proposed for sequential steps in base excision repair (for review, see Wilson and Kunkel, 2000).

DNA Binding by Yeast Mlh1-Pms1

Mlh1-Pms1 Has Intrinsic DNA Binding Activity

For our study of DNA binding by the yeast Mlh1-Pms1 heterodimer, the complex was purified to apparent homogeneity (Figure 3A) as described (Hall and Kunkel, 2001). To determine if Mlh1-Pms1 binds to DNA, we initially used size exclusion chromatography, an approach first performed with the *E. coli* MutL protein (Bende and Grafstrom, 1991). Free single-stranded M13mp2 DNA and free Mlh1-Pms1 elute from a Bio-Gel A-15m size exclusion column at different positions due to their large difference in size (Figure 3B). However, when Mlh1-Pms1 was applied to a column equilibrated with M13mp2 DNA, the protein eluted in the void volume at the position of the DNA alone, suggesting that the heterodimer bound to the DNA. Immunoblots of these column fractions using yMlh1- and yPms1-specific polyclonal antibodies (not shown) confirmed that the proteins in the void volume were Mlh1-Pms1. This result implies that Mlh1-Pms1 binds to DNA. This binding appears relatively stable based on the fact that the elution peaks of DNA and protein coincide. The majority of the protein was found in the peak coinciding with DNA, indicating that most or all of the Mlh1-Pms1 preparation is competent for DNA binding. We also observed Mlh1-Pms1 binding to DNA using a band shift assay and a duplex oligonucleotide substrate (Figure 3C). This result is similar to observations on *E. coli* MutL protein (Bende and Grafstrom, 1991; Ban *et al.*, 1999).

Higher Binding Affinity with Long DNA Substrates

To determine the ability of Mlh1-Pms1 to bind to other DNA substrates, filter-binding assays were performed

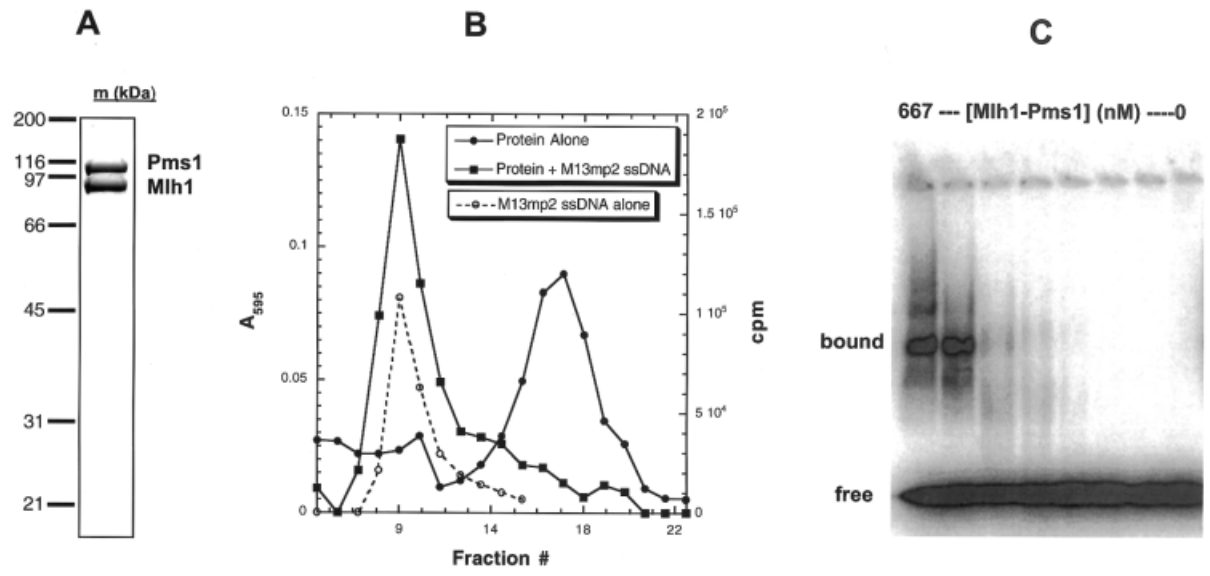


Fig. 3 Mlh1-Pms1 Binding to DNA.
(A) Purified yeast Mlh1-Pms1. (B) Binding of Mlh1-Pms1 to a DNA cellulose affinity column. Protein elution was followed by the Bradford dye assay and ^{3}H-DNA elution by scintillation counting. (C) Band shift analysis of Mlh1-Pms1 binding to a matched duplex 59-mer oligonucleotide.

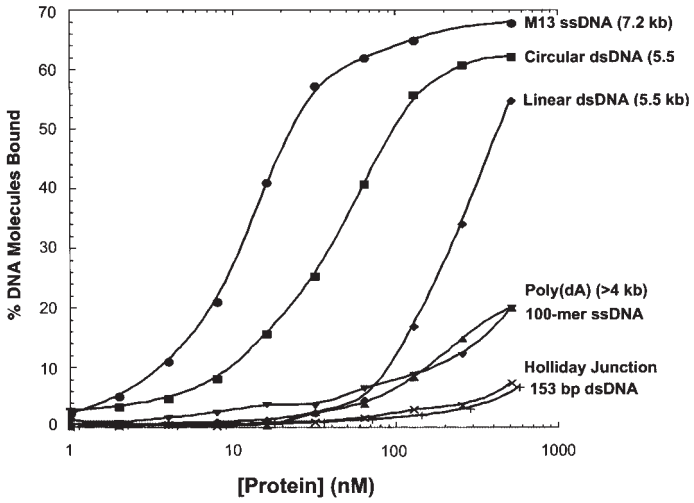


Fig. 4 Filter Binding Analysis of Mlh1-Pms1 Binding to DNA.
Filter binding assays were performed as a function of protein concentration as described in Hall *et al.* (2001). Data represent the average of three or four individual trials, and the results are independent of substrate concentration.

with a variety of ss- and dsDNA molecules (Figure 4). All DNA molecules consisting of relatively short DNA oligonucleotides were bound with low affinity, including substrates that mimic recombination intermediates (Holliday junction in Figure 4 or Y-junction, not shown) and dsDNA oligonucleotides with 5′ or 3′ ssDNA extensions (not shown). With short substrates, Mlh1-Pms1 binds to ssDNA with slightly higher affinity than to dsDNA, and binding to dsDNA was not altered by the presence of a mismatched base pair. Interestingly, the apparent binding affinity of Mlh1-Pms1 for long duplex DNA molecules was much higher (Figure 4). Moreover, when we probed the duplex DNA chain length required for high affinity binding, a sharp increase in affinity was seen as the length increased from 241 to 513 base pairs (see Figure 4 in Hall *et al.*, 2001). One explanation for such high binding affinity of Mlh1-Pms1 to long DNA substrates is positive cooperativity. To further investigate whether Mlh1-Pms1 binds to DNA cooperatively, atomic force microscopy (AFM) was used to directly visualize Mlh1-Pms1 bound to dsDNA. These images revealed that Mlh1-Pms1 usually bound in long, continuous tracts of protein coating the DNA (Figure 5 and additional images in Hall *et al.*, 2001). In the majority of images of continuous tracts, two separate dsDNA regions appeared to be in contact with the protein tracts (*e.g.*, Figure 5). These AFM images suggest

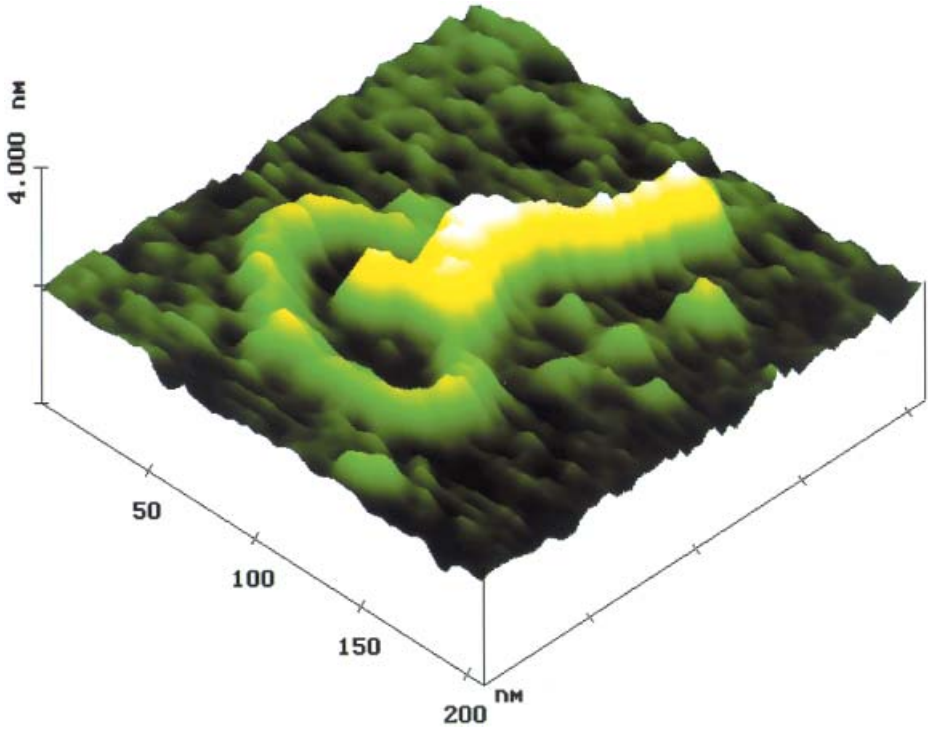


Fig. 5 Atomic Force Microscopy (AFM) Image of Mlh1-Pms1 Bound to Circular Duplex DNA.
A 1.8 kb plasmid DNA was deposited onto mica in the presence of Mlh1-Pms1 and visualized by AFM as described in Hall *et al.* (2001). The scan size is 200 nm and the plane of the mica was inclined by 40°.

that Mlh1-Pms1 contains more than one DNA binding site and that binding of two duplex DNA strands by Mlh1-Pms1 promotes the formation of long tracts of cooperatively bound protein.

These observations provide direct evidence that yeast Mlh1-Pms1 binds to both ssDNA and dsDNA in a mismatch-independent and sequence non-specific manner. The low affinity of Mlh1-Pms1 for short DNA molecules and the small preference for ssDNA over dsDNA is consistent with previous work on *E. coli* MutL (Ban *et al.*, 1999). The higher affinity of Mlh1-Pms1 for long DNA molecules, additional binding studies to circular *versus* linear duplex DNA substrates (not shown, but see Hall *et al.*, 2001) and the AFM images reveal strong positive cooperativity in DNA binding, a property not previously reported for MMR proteins. The AFM images also suggest that the Mlh1-Pms1 heterodimer has more than one DNA binding site. This differs from a model proposed for DNA binding by the MutL homodimer (Ban *et al.*, 1999), which was suggested to bind ssDNA (but not dsDNA) in a groove created by the dimerization of MutL N-terminal domains. It was further predicted that the C-terminal residues of intact MutL might contribute to formation of a dimeric protein structure that encircles the DNA molecule, thus implying a single DNA binding site within a central hole in the protein. Our results for binding of Mlh1-Pms1 to long duplex DNA imply that alternatives to this model may exist for the eukaryotic heterodimer. Nonetheless, extensive homology and conservation of function between bacterial MutL and eukaryotic MutL homologs implies that similarities may exist in certain DNA binding properties.

Most current models for DNA mismatch repair (Jiricny, 1998; Kolodner and Marsischky, 1999; Harfe and Jinks-Robertson, 2000) do not yet invoke a direct interaction of MutL homologs with DNA. Our results, demonstrating the ability of Mlh1-Pms1 to bind with high affinity to long dsDNA, suggest that these models may require revision. There is already some evidence that DNA binding by MutL plays a role in mismatch repair in *E. coli*. The interaction of MutL with DNA is important for its ability to stimulate DNA helicase II (Mechanic *et al.*, 2000), the helicase involved in the excision step of the mismatch repair pathway. Furthermore, DNA enhances the ATPase activity of MutL (Ban *et al.*, 1999), which is required for mismatch repair (Spampinato and Modrich, 2000) and is proposed to trigger the transformation of MutL from an initiation mode to a processing mode (Ban *et al.*, 1999). The presence of multiple DNA binding sites on Mlh1-Pms1 may be important for communication between the strand discrimination signal (*e.g.*, possibly a nick or the primer termini at a replication fork) and proteins bound at the mismatch, such as Msh2-Msh6. For example, in the presence of ATP, binding of *E. coli* MutS protein to a mismatch results in formation of α-loop structures (Allen *et al.*, 1997) that may be intermediates in the search for the strand discrimination signal. *E. coli* MutL enhances the yield of α-loops and it is found in a complex with MutS at the base of these structures where two dsDNA regions are juxtaposed. Perhaps MutL stabilizes this putative repair inter-

mediate not only by interacting with MutS, but also by binding simultaneously to the two dsDNA regions that merge at the base of the α-loop. Other possible functions for multiple DNA binding sites exist as well. Two Mlh1-Pms1 binding sites for dsDNA may be relevant to repair on the leading and lagging strands during replication. Current models of DNA mismatch repair also do not accommodate the present evidence for the cooperativity of DNA binding by Mlh1-Pms1 that could facilitate communication between the strand discrimination signal and the mismatch. Cooperative interactions between Mlh1-Pms1 heterodimers, and DNA binding in general, may be relevant to the other DNA transactions in which these MutL homologs participate. These processes include meiotic recombination, transcription-coupled excision repair of DNA adducts and other cellular responses to agents that damage DNA, cell cycle checkpoint control and apoptosis (for reviews on biological roles of mismatch repair proteins see Buermeyer *et al.*, 1999; Harfe and Jinks-Robertson, 2000). It will be interesting to determine if the DNA binding capacity of these MutL homologs is required for these processes. The possible role of DNA binding by Mlh1-Pms1 in genetic recombination is of particular interest. Recombination involves homologous pairing of two DNA molecules over relatively long stretches. The cooperative, multiple-site DNA binding activity of Mlh1-Pms1 could be relevant here, although the two duplexes with which Mlh1-Pms1 simultaneously interacts need not be homologous. It would be also interesting to determine whether other dimeric MutL complexes (Mlh1-Mlh2 or Mlh1-Mlh3) possess a similar high affinity cooperative DNA binding activity. This may improve our understanding of the biological functions of these other heterodimers.

References

Allen, D.J., Makhov, A., Grilley, M., Taylor, J., Thresher, R., Modrich, P., and Griffith, J.D. (1997). MutS mediates heteroduplex loop formation by a translocation mechanism. EMBO J. *16*, 4467–4476.

Ban, C., and Yang, W. (1998). Crystal structure and ATPase activity of MutL: implications for DNA repair and mutagenesis. Cell *95*, 541–552.

Ban, C., Junop, M., and Yang, W. (1999). Transformation of MutL by ATP binding and hydrolysis: a switch in DNA mismatch repair. Cell *97*, 85–97.

Bende, S.M., and Grafstrom, R.H. (1991). The DNA binding properties of the MutL protein isolated from *Escherichia coli*. Nucleic Acids Res. *19*, 1549–1555.

Bowers, J., Sokolsky, T., Quach, T., and Alani, E. (1999). A mutation in the Msh6 subunit of the *Saccharomyces cerevisiae* Msh2-Msh6 complex disrupts mismatch recognition. J. Biol. Chem. *274*, 16115–16125.

Buermeyer, A.B., Deschenes, S.M., Baker, S.M., and Liskay, R.M. (1999). Mammalian DNA mismatch repair. Annu. Rev. Genet. *33*, 533–564.

Das Gupta, R., and Kolodner, R.D. (2000). Novel dominant mutatons in *Saccharomyces cerivisiae MSH6*. Nature Genet. *24*, 53–56.

Drotschmann, K., Yang, W., Brownewell, F.E., Kool, E.T., and Kunkel, T.A. (2001). Asymmetric recognition of DNA local distortion. Structure-based functional studies of eukaryotic Msh2-Msh6. J. Biol. Chem. *276*, 46225–46229.

Dufner, P., Marra, G., Raschle, M., and Jiricny, J. (2000). Mismatch recognition and DNA-dependent stimulation of the ATPase activity of hMutSα is abolished by a single mutation in the hMSH6 subunit. J. Biol. Chem. *275*, 36550–36555.

Hall, M.C., and Kunkel, T.A. (2001). Purification of eukaryotic MutL homologs from *Saccharomyces cerevisiae* using self-affinity technology. Protein Expr. Purif. *21*, 333–342.

Hall, M.C., Wang, H., Erie, D.A., and Kunkel, T.A. (2001). High affinity cooperative DNA binding by the yeast Mlh1-Pms1 heterodimer. J. Mol. Biol. *312*, 637–647.

Harfe, B.D., and Jinks-Robertson, S. (2000). DNA mismatch repair and genetic instability. Annu. Rev. Genet. *34*, 359–399.

Hsieh, P. (2001). Molecular mechanisms of DNA mismatch repair. Mutat. Res. *486*, 71–87.

Hughes, M.J., and Jiricny, J. (1992). The purification of a human mismatch-binding protein and identification of its associated atpase and helicase activities. J. Biol. Chem. *267*, 23876–23882.

Iaccarino, I., Marra, G., Palombo, F., and Jiricny, J. (1998). hMsh2 and hMsh6 play distinct roles in mismatch binding and contribute differently to the ATPase activity of hMutSα. EMBO J. *17*, 2677–2686.

Jiricny, J. (1998). Eukaryotic mismatch repair: an update. Mutat. Res. *409*, 107–121.

Kolodner, R.D., and Marsischky, G.T. (1999). Eukaryotic DNA mismatch repair. Curr. Opin. Genet. Dev. *9*, 89–96.

Lamers, M.H., Perrakis, A., Enzlin, J.H., Winterwerp, H.H., de Wind, N., and Sixma, T.K. (2000). The crystal structure of DNA mismatch repair protein MutS binding to a G x T mismatch. Nature *407*, 711–717.

Malkov, V.A., Biswas, I., Camerini-Otero, R.D., and Hsieh, P. (1997). Photocross-linking of the NH2-terminal region of *Taq* MutS protein to the major groove of a heteroduplex DNA. J. Biol. Chem. *272*, 23811–23817.

Mechanic, L.E., Frankel, B.A., and Matson, S.W. (2000). *Escherichia coli* MutL loads DNA helicase II onto DNA. J. Biol. Chem. *275*, 38337–38346.

Obmolova, G., Ban, C., Hsieh, P., and Yang, W. (2000). Crystal structures of mismatch repair protein MutS and its complex with a substrate DNA. Nature *407*, 703–710.

Schofield, M.J., Brownewell, F.E., Nayak, S., Du, C., Kool, E.T., and Hsieh, P. (2001). The Phe-X-Glu DNA binding motif in MutS: the role of hydrogen bonding in mismatch recognition. J. Biol. Chem. *276*, 45505–45508.

Spampinato, C., and Modrich, P. (2000). The MutL ATPase is required for mismatch repair. J. Biol. Chem. *275*, 9863–9869.

Wilson, S.H., and Kunkel, T.A. (2000). Passing the baton in base excision repair. Nature Struct. Biol. *7*, 176–178.

Yamamoto, A., Schofield, M.J., Biswas, I., and Hsieh, P. (2000). Requirement for Phe36 for DNA binding and mismatch repair by *Escherichia coli* MutS protein. Nucleic Acids Res. *28*, 3564–3569.