YALE JOURNAL OF BIOLOGY AND MEDICINE 86 (2013), pp.453-461.

FOCUS: 50 YEARS OF DNA REPAIR: THE YALE SYMPOSIUM REPORTS

Investigations of Homologous Recombination Pathways and Their Regulation

James M. Daley, YoungHo Kwon, Hengyao Niu, and Patrick Sung*

Molecular Biophysics & Biochemistry, Yale School of Medicine, New Haven, Connecticut

The DNA double-strand break (DSB†), arising from exposure to ionizing radiation or various chemotherapeutic agents or from replication fork collapse, is among the most dangerous of chromosomal lesions. DSBs are highly cytotoxic and can lead to translocations, deletions, duplications, or mutations if mishandled. DSBs are eliminated by either homologous recombination (HR), which uses a homologous template to guide accurate repair, or by nonhomologous end joining (NHEJ), which simply rejoins the two broken ends after damaged nucleotides have been removed. HR generates error-free repair products and is also required for generating chromosome arm crossovers between homologous chromosomes in meiotic cells. The HR reaction includes several distinct steps: resection of DNA ends, homologous DNA pairing, DNA synthesis, and processing of HR intermediates. Each occurs in a highly regulated fashion utilizing multiple protein factors. These steps are being elucidated using a combination of genetic tools, cell-based assays, and *in vitro* reconstitution with highly purified HR proteins. In this review, we summarize contributions from our laboratory at Yale University in understanding HR mechanisms in eukaryotic cells.

*To whom all correspondence should be addressed: Patrick Sung, Yale School of Medicine, 333 Cedar St., SHM C-130, New Haven, CT 06520; Tele: 203-785-4569; Fax: 203-785-6404; Email: patrick.sung@yale.edu.

†Abbreviations: DSB, double-strand break; HR, homologous recombination; NHEJ, nonhomologous end joining; SDSA, synthesis-dependent strand annealing; OB, oligonucleotide/oligosaccharide-binding; RPA, replication protein A; MRX, Mre11-Rad50-Xrs2; MRN, MRE11-RAD50-NBS1.

Keywords: DNA repair, homologous recombination, double-strand breaks, resection, recombinase, presynaptic filament, synaptic complex, double Holliday junction

Author contributions: James M. Daley, YoungHo Kwon, and Hengyao Niu contributed equally.

INTRODUCTION

DNA is continually damaged by a plethora of endogenous and exogenous agents, including reactive oxygen species generated during cellular metabolism, ultraviolet light from the sun, and ionizing radiation. Accurate repair of damaged DNA is necessary to avoid accumulation of mutations. Dysfunctions of DNA repair pathways often lead to cancer and other diseases.

The study of DNA repair at Yale has a long and storied history. In the homologous recombination arena, pioneering work from the Yale laboratories of Charles Radding and Paul Howard-Flanders has led to insights regarding HR mechanisms in bacteria and provided essential experimental frameworks to guide similar studies in eukaryotes. For instance, Charles Radding contributed to the idea that recombination begins with the invasion of a homologous duplex by single-stranded DNA [1]. Over the course of many years, the Howard-Flanders and Radding laboratories continually made major findings that helped elucidate the biochemical mechanism by which the RecA recombinase promotes the homologous DNA pairing and strand exchange reaction that underlies HR-mediated processes. The intellectual capital contributed by our Yale colleagues has provided the impetus for us to address HR mechanisms in eukaryotes.

HOMOLOGOUS RECOMBINATION OVERVIEW

Double-strand break repair by homologous recombination is summarized in Figure 1. The process begins with DNA end resection, in which nucleolytic degradation of the 5' strand leaves a long 3' single-stranded DNA overhang. This ssDNA is then coated by replication protein A (RPA), which is subsequently displaced by a recombinase (Rad51 or Dmc1, the latter being meiosis-specific) to yield the presynaptic filament. Recombination mediator proteins such as Rad52 and BRCA2 are involved in this process. The synaptic complex is formed when the filament pairs with homologous dsDNA and involves the proteins PALB2 and RAD51AP1, as well as the Hop2-Mnd1 complex. Rad54 and Rdh54 then promote invasion of the homologous duplex, forming a structure called the displacement loop or D-loop. After DNA synthesis extends the D-loop, the structure can be dismantled by Mph1 (yeast) or FANCM (human), leading to a noncrossover repair outcome. Alternatively, a double Holliday junction can form, which can be resolved by specialized nucleases termed resolvases. The orientation of the DNA incisions introduced by the resolvase determines whether a crossover or noncrossover recombinant is made. These junctions can also be dissolved by a helicase-topoisomerase complex (BLM-Topo IIIα-RMI1-RMI2) to yield noncrossover products. Altogether, recombination is a highly complex reaction involving a multitude of enzymatic activities, with potential for regulation at many points. Our laboratory has worked to unravel the mechanisms of many of these steps, and some of our endeavors are reviewed below.

DNA END RESECTION

In order to recruit the proteins that catalyze DSB repair, 3' ssDNA tails must first be created at the break site in a process termed DNA end resection (Figure 1). Central to this resection process is the MRX (yeast) or MRN (human) protein complex comprised of Mre11, Rad50, and Xrs2/NBS1. MRX/MRN possesses 3' to 5' exonuclease and structure-specific endonuclease activities and is one of the first protein complexes recruited to DSB ends. The MRX/MRN complex acts as a sensor in DNA damage checkpoint signaling as well. Interestingly, MRX is also indispensable for NHEJ in yeast. Our laboratory has contributed findings regarding the assembly of this complex and the regulation of the Mre11 nuclease activities by Rad50 and Xrs2 [2-4].

Genetic studies in yeast have identified MRX as one of three nucleases that function in DNA end resection [5,6]. Specifically, working in conjunction with Sae2 (CtIP in humans), MRX trims DNA ends at the vicinity of the break. Long range resection

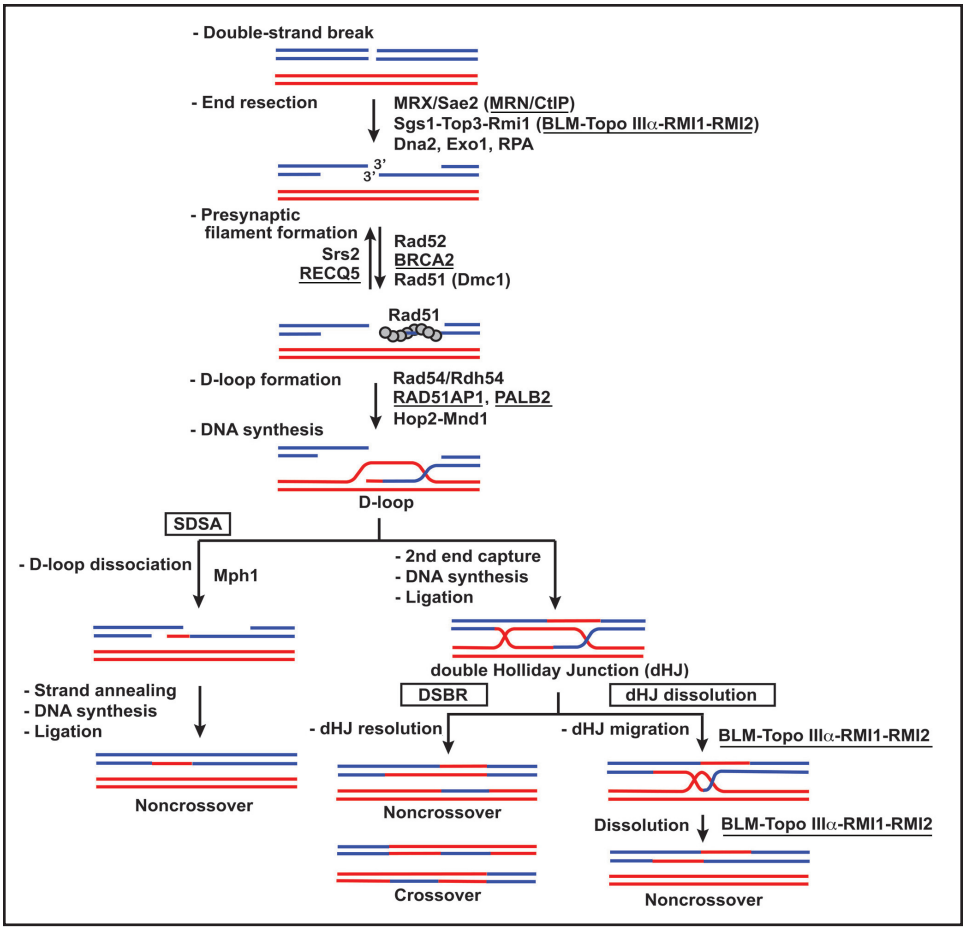

Figure 1. Protein factors that process DNA double-strand breaks and mediate homologous pairing. Three distinct HR pathways (synthesis dependent strand annealing (SDSA), double-strand break repair (DSBR), and double Holliday (dHJ) dissolution) are shown. *S. cerevisiae* and human proteins (underlined) addressed in this review are indicated.

is catalyzed by either the 5' to 3' exonuclease Exo1 or the ssDNA endonuclease Dna2. Unlike Exo1, which is active on dsDNA, the action of Dna2 relies on duplex unwinding by a 3' to 5' helicase, Sgs1. Sgs1 is orthologous to human BLM, which is mutated in the cancer-prone disease Bloom syndrome. Our laboratory has successfully reconstituted the Sgs1-Dna2-depenent resection pathway and provided mechanistic information regarding the action of the Sgs1-Dna2 helicase-nuclease ensemble [7]. Specifically, our results have revealed the roles of the single-strand DNA binding protein RPA and the MRX and Top3-Rmi1 complexes in resection by Sgs1-Dna2 (Figure 1). Importantly, the Sgs1-Dna2-catalyzed resection is regulated in a cell cycle-dependent manner via Cdk1-mediated phosphorylation of Dna2, a mechanism that serves to activate HR in the S/G2 phase of the cell cycle [8]. In collaboration with Craig Peterson's group at the University of Massachusetts, we have examined the influence of nucleosome dynamics on DNA end resection and found that while a mononucleosome completely blocks Exo1-catalyzed resection, the Sgs1-Dna2 path can partially overcome the nucleosomal barrier [9]. Taken together, the results described above have led to mechanistic understanding of the DNA end resection pathways and their regulation during the cell cycle and by chromatin structure. Our reconstituted systems have also set the stage

for tackling additional questions regarding DSB processing in mitotic and meiotic cells.

EUKARYOTIC RECOMBINASES: Rad51 AND Dmc1

The ssDNA derived from DNA end resection is first engaged by RPA, which is then replaced by a general recombinase, either Rad51 or Dmc1, to mediate homologous DNA pairing. This leads to the formation of a DNA joint between the ssDNA and donor DNA molecule. Rad51 is required for both mitotic DSB repair and meiotic HR, while the role of Dmc1 is limited to meiosis [10]. Our biochemical studies on *S. cerevisiae* and human Rad51 and Dmc1 proteins have revealed key properties of the recombinase protein filament assembled on ssDNA, commonly referred to as the presynaptic filament [11-17]. The presynaptic filament has a right-handed helical architecture, with ~18 nucleotides being engaged by ~6 protomers of the recombinase in each repeat. Within the presynaptic filament, the DNA is stretched to give an axial rise of ~5.4Å between adjacent nucleotides [14,18,19]. This extended DNA conformation is characteristic of a catalytically active presynaptic filament. In the homologous pairing reaction, the presynaptic filament engages the duplex DNA molecule and samples it for homology. Once homology is found, the recombining DNA molecules become aligned in homologous registry in a higher order ensemble called the synaptic complex. Finally, invasion of the duplex by the presynaptic filament yields a DNA joint called the displacement loop, or D-loop. As discussed later, studies by us and others have led to the identification of specific HR factors that facilitate the assembly of the presynaptic filament and synaptic complex and that promote the DNA strand invasion reaction (Figure 1).

PROMOTION OF PRESYNAPTIC FILAMENT ASSEMBLY BY RECOMBINATION MEDIATORS

Owing to its high affinity for ssDNA, RPA poses a challenge to the timely assembly of the presynaptic filament [20]. Biochemical and cell-based studies have revealed recombination mediator proteins that facilitate the assembly of the Rad51 and Dmc1 presynaptic filaments on RPA-coated ssDNA. The most well-studied HR mediators are *S. cerevisiae* Rad52 and human BRCA2 (Breast Cancer Susceptibility 2). Purified Rad52 efficiently overcomes the inhibitory action of RPA on Rad51-mediated homologous DNA pairing strand exchange [21]. Rad52 is a ring-shaped oligomer that harbors domains conferring DNA binding activity and the ability to interact with Rad51 and RPA. These Rad52 domains contribute to its mediator function *in vitro* [22,23] and in cells [24]. BRCA2, long known to be essential for HR and the maintenance of genetic stability in mammalian cells, binds DNA and interacts with RAD51 via several BRC repeats located in the middle portion of the protein and also a distinct module within its C-terminus [10]. We have shown that a polypeptide derived from BRCA2 containing the BRC repeats 3 and 4 and the DNA binding domain possesses recombination mediator activity [25]. It remains to be determined how the C-terminal RAD51 interaction domain of BRCA2 and several BRCA2-associated proteins, such as DSS1 and PALB2 [10], affect presynaptic filament assembly.

HR FACTORS THAT FACILITATE SYNAPTIC COMPLEX ASSEMBLY

Several HR factors, namely, RAD51AP1, the tumor suppressor PALB2, and the Hop2-Mnd1 complex, enhance the efficiency of synaptic complex assembly. RAD51AP1, which is needed for HR and is vertebrate-specific [26-28], was identified in a yeast two-hybrid screen for proteins that interact with human RAD51 by Radding and colleagues [26]. It was later found in our laboratory to also associate with DMC1 [29,30]. PALB2 interacts with RAD51 as well and additionally with RAD51AP1 [31]. Both RAD51AP1 and PALB2 bind DNA and can, individually, co-operate with the RAD51 presynaptic filament to capture du-

plex DNA and assemble the synaptic complex [28,31,32].

Importantly, RAD51AP1 and PALB2 act synergistically in synaptic complex assembly in a manner that relies on complex formation between the two proteins [31]. RAD51AP1 also functions with DMC1 in synaptic complex assembly [29,30], but the role, if any, of PALB2 or the RAD51AP1-PALB2 complex in DMC1-mediated HR remains to be determined.

In the budding and fission yeasts, the Hop2 and Mnd1 proteins form a heterodimeric complex to promote crossover formation during meiosis [10,33] via enhancement of Dmc1-mediated homologous DNA pairing [34]. The mammalian Hop2-Mnd1 complex functions with both Rad51 and Dmc1 [35]. Biochemical analyses by us and others have revealed that via its DNA binding activity and physical interaction with the presynaptic filament, Hop2-Mnd1 helps capture the duplex DNA partner to assemble the synaptic complex [36,37].

MULTIFACETED ROLE OF Rad54 AND Rdh54 IN HOMOLOGOUS DNA PAIRING

Rad54 and Rdh54 are members of the Swi2/Snf2 family of DNA motor proteins that, at the expense of ATP hydrolysis, are capable of translocation on duplex DNA [38]. Purified *S. cerevisiae* Rad54 and Rdh54 interact with Rad51 and Dmc1 and greatly enhance the homologous DNA pairing reaction mediated by these recombinases [18,39-42]. When translocating on DNA, Rad54 and Rdh54 generate extensive negative supercoiling that induces transient separation of DNA strands, which is believed to promote DNA strand invasion by the Rad51 presynaptic filament [41-43]. Additionally, both proteins possess a chromatin remodeling activity that enables D-loop formation in a chromatinized template [44-46]. Interestingly, these proteins also mediate the migration of the nascent Holliday structure made during HR and remove Rad51 and Dmc1 from dsDNA [39,47-53]. The latter attribute has been implicated in the repair DNA synthesis reaction and in avoiding the accumulation of cytotoxic nucleoprotein intermediates.

While Rad54 and Rdh54 possess very similar enzymatic activities and clearly provide overlapping functions in HR, they are not strictly redundant biologically. For instance, Rad54 functions in intrachromosomal recombination and repair of DSBs, whereas Rdh54 appears to be more important in the promotion of interhomolog recombination that is dependent on Dmc1 and prevention of Dmc1 accumulation at non-recombination sites [54,55]. In addition, Rdh54 is needed for the adaptation to checkpoint-mediated G2/M arrest induced by DSBs, which is not replaceable with Rad54 [56]. Moreover, Rdh54 is phosphorylated by Mec1 and Rad53 [57], whereas phosphorylation of Rad54 occurs during meiotic recombination by Mek1 [58].

PROMOTION OF THE NON-CROSSOVER SYNTHESIS-DEPENDENT STRAND ANNEALING PATHWAY BY THE Mph1 HELICASE

Early on in meiosis, DNA crossovers generated by the HR machinery serve to stably tie homologous chromosome pairs until it is time for their segregation in the first division. However, owing to the inherent danger of chromosome translocations and loss of heterozygosity associated with crossover HR, this mode of recombination is actively suppressed by several distinct mechanisms. In *S. cerevisiae*, the Mph1 helicase is a major negative regulator of crossover HR, and it acts by resolving D-loop intermediates via the non-crossover pathway of synthesis-dependent strand annealing (SDSA) (Figure 1). Specifically, Mph1 utilizes its helicase function to dissociate the invading strand from the D-loop structure (Figure 1; [59]). It should be noted that Mph1 also promotes the regression of a model DNA replication fork *in vitro*, an activity that is likely germane for DNA replication repair in cells [60]. Fml1 and FANCM, orthologs of Mph1 in the fission yeast and humans, respectively, have enzymatic attributes similar to what we have described for Mph1 [61,62].

CROSSOVER SUPPRESSION VIA DOUBLE HOLLIDAY JUNCTION DISSOLUTION BY BLM-TOPO IIIα-RMI1-RMI2

Double Holliday junction (dHJ) dissolution is another conserved mechanism by which a crossover outcome is prevented in HR [63]. Whereas dHJ processing by resolvases can yield either crossover or noncrossover products, dissolution always leads to noncrossovers. This entails convergent branch migration of the two Holliday junctions generated via second DNA end capture in HR and untangling of the hemicatenae by a specialized topoisomerase (Figure 1). The dHJ dissolution reaction is mediated by the BLM helicase in conjunction with the type IA topoisomerase Topo IIIα. RPA and the OB (Oligonucleotide/Oligosaccharide-Binding)-fold containing RMI1-RMI2 complex associate with BLM-Topo IIIα and enhance the dHJ dissolution reaction [63-70]. The higher order ensemble of the aforementioned proteins has been termed the dHJ dissolvasome. BLM's involvement in the suppression of crossover formation is consistent with genetic results from the budding yeast showing a similar role of its ortholog Sgs1 and provides a satisfactory explanation as to the elevated frequency of sister chromatid exchanges in cells of Bloom syndrome patients [71-73]. As discussed above, BLM/Sgs1 also plays an important early role in DNA end resection.

Srs2 AND RECQ5: NEGATIVE HR REGULATORS THAT DISASSEMBLE THE PRESYNAPTIC FILAMENT

Even though HR provides an important means for chromosome damage repair, it can generate cytotoxic intermediates and deleterious chromosome rearrangements [20,74]. Our laboratory has contributed the finding that the budding yeast Srs2 helicase helps suppress spurious HR events via disruption of the Rad51 presynaptic filament (Figure 1) [75]. This process is stimulated by RPA and requires complex formation between Srs2 and Rad51 [76]. In humans, the RecQ family helicase RECQ5 is the functional equivalent of Srs2. Specifically, RECQ5-deficient mouse cells exhibit a hyper-recombination phenotype and mutant animals are predisposed to cancer [77]. Purified RECQ5 interacts with RAD51 and inhibits presynaptic filament assembly in a manner that is stimulated by RPA [77] and dependent on complex formation with RAD51 [78].

CONCLUSION

Our studies have provided insights into how the HR machinery forms and subsequently processes DNA joints during recombination. We have also contributed toward the delineation of an intricate network of regulatory mechanisms that influence the frequency and outcome of HR. These studies have benefited greatly from the intellectual framework established by Yale colleagues whose work preceded ours. We anticipate that our future endeavors will continue to help elucidate the mechanism and regulation of HR and its role in genome maintenance.

REFERENCES

1. Meselson MS, Radding CM. A general model for genetic recombination. Proc Natl Acad Sci USA. 1975;72(1):358-61.
2. Trujillo KM, Roh DH, Chen L, Van Komen S, Tomkinson A, Sung P. Yeast xrs2 binds DNA and helps target rad50 and mre11 to DNA ends. J Biol Chem. 2003;278(49):48957-64.
3. Trujillo KM, Sung P. DNA structure-specific nuclease activities in the Saccharomyces cerevisiae Rad50*Mre11 complex. J Biol Chem. 2001;276(38):35458-64.
4. Trujillo KM, Yuan SS, Lee EY, Sung P. Nuclease activities in a complex of human recombination and DNA repair factors Rad50, Mre11, and p95. J Biol Chem. 1998;273(34):21447-50.
5. Mimitou EP, Symington LS. Sae2, Exo1 and Sgs1 collaborate in DNA double-strand break processing. Nature. 2008;455(7214):770-4.
6. Zhu Z, Chung WH, Shim EY, Lee SE, Ira G. Sgs1 helicase and two nucleases Dna2 and Exo1 resect DNA double-strand break ends. Cell. 2008;134(6):981-94.
7. Niu H, Chung WH, Zhu Z, Kwon Y, Zhao W, Chi P, et al. Mechanism of the ATP-dependent DNA end-resection machinery from Saccharomyces cerevisiae. Nature. 2010;467(7311):108-11.
8. Chen X, Niu H, Chung WH, Zhu Z, Papusha A, Shim EY, et al. Cell cycle regulation of

DNA double-strand break end resection by Cdk1-dependent Dna2 phosphorylation. Nat Struct Mol Biol. 2012;18(9):1015-9.
9. Adkins NL, Niu H, Sung P, Peterson CL. Nucleosome dynamics regulates DNA processing. Nat Struct Mol Biol. 2013;20(7):836-42.
10. San Filippo J, Sung P, Klein H. Mechanism of eukaryotic homologous recombination. Annu Rev Biochem. 2008;77:229-57.
11. Chi P, Van Komen S, Sehorn MG, Sigurdsson S, Sung P. Roles of ATP binding and ATP hydrolysis in human Rad51 recombinase function. DNA Repair (Amst). 2006;5(3):381-91.
12. Robertson RB, Moses DN, Kwon Y, Chan P, Chi P, Klein H, et al. Structural transitions within human Rad51 nucleoprotein filaments. Proc Natl Acad Sci USA. 2009;106(31):12688-93.
13. Robertson RB, Moses DN, Kwon Y, Chan P, Zhao W, Chi P, et al. Visualizing the disassembly of S. cerevisiae Rad51 nucleoprotein filaments. J Mol Biol. 2009;388(4):703-20.
14. Sehorn MG, Sigurdsson S, Bussen W, Unger VM, Sung P. Human meiotic recombinase Dmc1 promotes ATP-dependent homologous DNA strand exchange. Nature. 2004;429(6990):433-7.
15. Sigurdsson S, Trujillo K, Song B, Stratton S, Sung P. Basis for avid homologous DNA strand exchange by human Rad51 and RPA. J Biol Chem. 2001;276(12):8798-806.
16. Sung P. Catalysis of ATP-dependent homologous DNA pairing and strand exchange by yeast RAD51 protein. Science. 1994;265(5176):1241-3.
17. Sung P, Robberson DL. DNA strand exchange mediated by a RAD51-ssDNA nucleoprotein filament with polarity opposite to that of RecA. Cell. 1995;82(3):453-61.
18. Busygina V, Gaines WA, Xu Y, Kwon Y, Williams GJ, Lin SW, et al. Functional attributes of the Saccharomyces cerevisiae meiotic recombinase Dmc1. DNA Repair (Amst). 2013;12(9):707-12.
19. Sheridan SD, Yu X, Roth R, Heuser JE, Sehorn MG, Sung P, et al. A comparative analysis of Dmc1 and Rad51 nucleoprotein filaments. Nucleic Acids Res. 2008;36(12):4057-66.
20. Sung P, Klein H. Mechanism of homologous recombination: mediators and helicases take on regulatory functions. Nat Rev Mol Cell Biol. 2006;7(10):739-50.
21. Sung P. Function of yeast Rad52 protein as a mediator between replication protein A and the Rad51 recombinase. J Biol Chem. 1997;272(45):28194-7.
22. Krejci L, Song B, Bussen W, Rothstein R, Mortensen UH, Sung P. Interaction with Rad51 is indispensable for recombination mediator function of Rad52. J Biol Chem. 2002;277(42):40132-41.
23. Seong C, Sehorn MG, Plate I, Shi I, Song B, Chi P, et al. Molecular anatomy of the recombination mediator function of Saccharomyces cerevisiae Rad52. J Biol Chem. 2008;283(18):12166-74.
24. Plate I, Hallwyl SC, Shi I, Krejci L, Muller C, Albertsen L, et al. Interaction with RPA is necessary for Rad52 repair center formation and for its mediator activity. J Biol Chem. 2008;283(43):29077-85.
25. San Filippo J, Chi P, Sehorn MG, Etchin J, Krejci L, Sung P. Recombination mediator and Rad51 targeting activities of a human BRCA2 polypeptide. J Biol Chem. 2006;281(17):11649-57.
26. Kovalenko OV, Golub EI, Bray-Ward P, Ward DC, Radding CM. A novel nucleic acid-binding protein that interacts with human rad51 recombinase. Nucleic Acids Res. 1997;25(24):4946-53.
27. Mizuta R, LaSalle JM, Cheng HL, Shinohara A, Ogawa H, Copeland N, et al. RAB22 and RAB163/mouse BRCA2: proteins that specifically interact with the RAD51 protein. Proc Natl Acad Sci USA. 1997;94(13):6927-32.
28. Wiese C, Dray E, Groesser T, San Filippo J, Shi I, Collins DW, et al. Promotion of homologous recombination and genomic stability by RAD51AP1 via RAD51 recombinase enhancement. Mol Cell. 2007;28(3):482-90.
29. Dray E, Dunlop MH, Kauppi L, Filippo JS, Wiese C, Tsai MS, et al. Molecular basis for enhancement of the meiotic DMC1 recombinase by RAD51 associated protein 1 (RAD51AP1). Proc Natl Acad Sci USA. 2011;108(9):3560-5.
30. Dunlop MH, Dray E, Zhao W, Tsai MS, Wiese C, Schild D, et al. RAD51-associated protein 1 (RAD51AP1) interacts with the meiotic recombinase DMC1 through a conserved motif. J Biol Chem. 2011;286(43):37328-34.
31. Dray E, Etchin J, Wiese C, Saro D, Williams GJ, Hammel M, et al. Enhancement of RAD51 recombinase activity by the tumor suppressor PALB2. Nat Struct Mol Biol. 2010;17(10):1255-9.
32. Dunlop MH, Dray E, Zhao W, San Filippo J, Tsai MS, Leung SG, et al. Mechanistic insights into RAD51-associated protein 1 (RAD51AP1) action in homologous DNA repair. J Biol Chem. 2012;287(15):12343-7.
33. Tsubouchi H, Roeder GS. The Mnd1 protein forms a complex with hop2 to promote homologous chromosome pairing and meiotic double-strand break repair. Mol Cell Biol. 2002;22(9):3078-88.
34. Ploquin M, Petukhova GV, Morneau D, Dery U, Bransi A, Stasiak A, et al. Stimulation of fission yeast and mouse Hop2-Mnd1 of the Dmc1 and Rad51 recombinases. Nucleic Acids Res. 2007;35(8):2719-33.
35. Petukhova GV, Pezza RJ, Vanevski F, Ploquin M, Masson JY, Camerini-Otero RD. The Hop2 and Mnd1 proteins act in concert with Rad51 and Dmc1 in meiotic recombination. Nat Struct Mol Biol. 2005;12(5):449-53.

36. Chi P, San Filippo J, Sehorn MG, Petukhova GV, Sung P. Bipartite stimulatory action of the Hop2-Mnd1 complex on the Rad51 recombinase. Genes Dev. 2007;21(14):1747-57.
37. Pezza RJ, Voloshin ON, Vanevski F, Camerini-Otero RD. Hop2/Mnd1 acts on two critical steps in Dmc1-promoted homologous pairing. Genes Dev. 2007;21(14):1758-66.
38. Heyer WD, Li X, Rolfsmeier M, Zhang XP. Rad54: the Swiss Army knife of homologous recombination? Nucleic Acids Res. 2006;34(15):4115-25.
39. Chi P, Kwon Y, Seong C, Epshtein A, Lam I, Sung P, et al. Yeast recombination factor Rdh54 functionally interacts with the Rad51 recombinase and catalyzes Rad51 removal from DNA. J Biol Chem. 2006;281(36):26268-79.
40. Petukhova G, Stratton S, Sung P. Catalysis of homologous DNA pairing by yeast Rad51 and Rad54 proteins. Nature. 1998;393(6680):91-4.
41. Petukhova G, Sung P, Klein H. Promotion of Rad51-dependent D-loop formation by yeast recombination factor Rdh54/Tid1. Genes Dev. 2000;14(17):2206-15.
42. Van Komen S, Petukhova G, Sigurdsson S, Stratton S, Sung P. Superhelicity-driven homologous DNA pairing by yeast recombination factors Rad51 and Rad54. Mol Cell. 2000;6(3):563-72.
43. Tan TL, Essers J, Citterio E, Swagemakers SM, de Wit J, Benson FE, et al. Mouse Rad54 affects DNA conformation and DNA-damage-induced Rad51 foci formation. Curr Biol. 1999;9(6):325-8.
44. Jaskelioff M, Van Komen S, Krebs JE, Sung P, Peterson CL. Rad54p is a chromatin remodeling enzyme required for heteroduplex DNA joint formation with chromatin. J Biol Chem. 2003;278(11):9212-8.
45. Kwon Y, Chi P, Roh DH, Klein H, Sung P. Synergistic action of the Saccharomyces cerevisiae homologous recombination factors Rad54 and Rad51 in chromatin remodeling. DNA Repair (Amst). 2007;6(10):1496-506.
46. Kwon Y, Seong C, Chi P, Greene EC, Klein H, Sung P. ATP-dependent chromatin remodeling by the Saccharomyces cerevisiae homologous recombination factor Rdh54. J Biol Chem. 2008;283(16):10445-52.
47. Bugreev DV, Mazina OM, Mazin AV. Rad54 protein promotes branch migration of Holliday junctions. Nature. 2006;442(7102):590-3.
48. Nimonkar AV, Amitani I, Baskin RJ, Kowalczykowski SC. Single molecule imaging of Tid1/Rdh54, a Rad54 homolog that translocates on duplex DNA and can disrupt joint molecules. J Biol Chem. 2007;282(42):30776-84.
49. Chi P, Kwon Y, Visnapuu ML, Lam I, Santa Maria SR, Zheng X, et al. Analyses of the yeast Rad51 recombinase A265V mutant reveal different in vivo roles of Swi2-like factors. Nucleic Acids Res. 2011;39(15):6511-22.
50. Holzen TM, Shah PP, Olivares HA, Bishop DK. Tid1/Rdh54 promotes dissociation of Dmc1 from nonrecombinogenic sites on meiotic chromatin. Genes Dev. 2006;20(18):2593-604.
51. Santa Maria SR, Kwon Y, Sung P, Klein HL. Characterization of the Interaction between the Saccharomyces cerevisiae Rad51 Recombinase and the DNA Translocase Rdh54. J Biol Chem. 2013;288(30):21999-2005.
52. Shah PP, Zheng X, Epshtein A, Carey JN, Bishop DK, Klein HL. Swi2/Snf2-related translocases prevent accumulation of toxic Rad51 complexes during mitotic growth. Mol Cell. 2010;39(6):862-72.
53. Solinger JA, Kiianitsa K, Heyer WD. Rad54, a Swi2/Snf2-like recombinational repair protein, disassembles Rad51:dsDNA filaments. Mol Cell. 2002;10(5):1175-88.
54. Klein HL. RDH54, a RAD54 homologue in Saccharomyces cerevisiae, is required for mitotic diploid-specific recombination and repair and for meiosis. Genetics. 1997;147(4):1533-43.
55. Shinohara M, Shita-Yamaguchi E, Buerstedde JM, Shinagawa H, Ogawa H, Shinohara A. Characterization of the roles of the Saccharomyces cerevisiae RAD54 gene and a homologue of RAD54, RDH54/TID1, in mitosis and meiosis. Genetics. 1997;147(4):1545-56.
56. Lee SE, Pellicioli A, Malkova A, Foiani M, Haber JE. The Saccharomyces recombination protein Tid1p is required for adaptation from G2/M arrest induced by a double-strand break. Curr Biol. 2001;11(13):1053-7.
57. Ferrari M, Nachimuthu BT, Donnianni RA, Klein H, Pellicioli A. Tid1/Rdh54 translocase is phosphorylated through a Mec1- and Rad53-dependent manner in the presence of DSB lesions in budding yeast. DNA Repair (Amst). 2013;12(5):347-55.
58. Niu H, Wan L, Busygina V, Kwon Y, Allen JA, Li X, et al. Regulation of meiotic recombination via Mek1-mediated Rad54 phosphorylation. Mol Cell. 2009;36(3):393-404.
59. Prakash R, Satory D, Dray E, Papusha A, Scheller J, Kramer W, et al. Yeast Mph1 helicase dissociates Rad51-made D-loops: implications for crossover control in mitotic recombination. Genes Dev. 2009;23(1):67-79.
60. Zheng XF, Prakash R, Saro D, Longerich S, Niu H, Sung P. Processing of DNA structures via DNA unwinding and branch migration by the S. cerevisiae Mph1 protein. DNA Repair (Amst). 2011;10(10):1034-43.
61. Gari K, Decaillet C, Stasiak AZ, Stasiak A, Constantinou A. The Fanconi anemia protein FANCM can promote branch migration of Holliday junctions and replication forks. Mol Cell. 2008;29(1):141-8.
62. Sun W, Nandi S, Osman F, Ahn JS, Jakovleska J, Lorenz A, et al. The FANCM

ortholog Fml1 promotes recombination at stalled replication forks and limits crossing over during DNA double-strand break repair. Mol Cell. 2008;32(1):118-28.
63. Wu L, Hickson ID. The Bloom's syndrome helicase suppresses crossing over during homologous recombination. Nature. 2003;426(6968):870-4.
64. Bussen W, Raynard S, Busygina V, Singh AK, Sung P. Holliday junction processing activity of the BLM-Topo IIIalpha-BLAP75 complex. J Biol Chem. 2007;282(43):31484-92.
65. Chang M, Bellaoui M, Zhang C, Desai R, Morozov P, Delgado-Cruzata L, et al. RMI1/NCE4, a suppressor of genome instability, encodes a member of the RecQ helicase/Topo III complex. EMBO J. 2005;24(11):2024-33.
66. Raynard S, Bussen W, Sung P. A double Holliday junction dissolvasome comprising BLM, topoisomerase IIIalpha, and BLAP75. J Biol Chem. 2006;281(20):13861-4.
67. Raynard S, Niu H, Sung P. DNA double-strand break processing: the beginning of the end. Genes Dev. 2008;22(21):2903-7.
68. Singh TR, Ali AM, Busygina V, Raynard S, Fan Q, Du CH, et al. BLAP18/RMI2, a novel OB-fold-containing protein, is an essential component of the Bloom helicase-double Holliday junction dissolvasome. Genes Dev. 2008;22(20):2856-68.
69. Xu D, Guo R, Sobeck A, Bachrati CZ, Yang J, Enomoto T, et al. RMI, a new OB-fold complex essential for Bloom syndrome protein to maintain genome stability. Genes Dev. 2008;22(20):2843-55.
70. Yin J, Sobeck A, Xu C, Meetei AR, Hoatlin M, Li L, et al. BLAP75, an essential component of Bloom's syndrome protein complexes that maintain genome integrity. EMBO J. 2005;24(7):1465-76.
71. Amin AD, Chaix AB, Mason RP, Badge RM, Borts RH. The roles of the Saccharomyces cerevisiae RecQ helicase SGS1 in meiotic genome surveillance. PLoS One. 2010;5(11):e15380.
72. Cejka P, Kowalczykowski SC. The full-length Saccharomyces cerevisiae Sgs1 protein is a vigorous DNA helicase that preferentially unwinds holliday junctions. J Biol Chem. 2010;285(11):8290-301.
73. Cejka P, Plank JL, Bachrati CZ, Hickson ID, Kowalczykowski SC. Rmi1 stimulates decatenation of double Holliday junctions during dissolution by Sgs1-Top3. Nat Struct Mol Biol. 2010;17(11):1377-82.
74. Wu L, Hickson ID. DNA helicases required for homologous recombination and repair of damaged replication forks. Annu Rev Genet. 2006;40:279-306.
75. Krejci L, Van Komen S, Li Y, Villemain J, Reddy MS, Klein H, et al. DNA helicase Srs2 disrupts the Rad51 presynaptic filament. Nature. 2003;423(6937):305-9.
76. Colavito S, Macris-Kiss M, Seong C, Gleeson O, Greene EC, Klein HL, et al. Functional significance of the Rad51-Srs2 complex in Rad51 presynaptic filament disruption. Nucleic Acids Res. 2009;37(20):6754-64.
77. Hu Y, Raynard S, Sehorn MG, Lu X, Bussen W, Zheng L, et al. RECQL5/Recql5 helicase regulates homologous recombination and suppresses tumor formation via disruption of Rad51 presynaptic filaments. Genes Dev. 2007;21(23):3073-84.
78. Schwendener S, Raynard S, Paliwal S, Cheng A, Kanagaraj R, Shevelev I, et al. Physical interaction of RECQ5 helicase with RAD51 facilitates its anti-recombinase activity. J Biol Chem. 2010;285(21):15739-45.