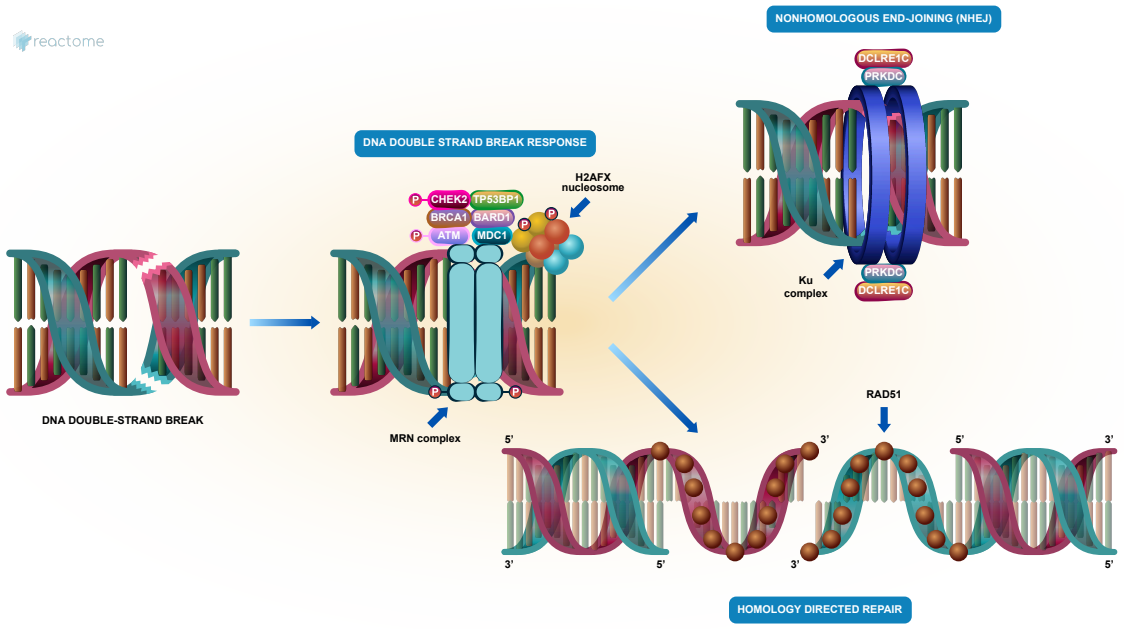
DNA Double-Strand Break Repair



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This is just an excerpt of a full-length report for this pathway. To access the complete report, please download it at the Reactome Textbook.

25/04/2024

Introduction

Reactome is open-source, open access, manually curated and peer-reviewed pathway database. Pathway annotations are authored by expert biologists, in collaboration with Reactome editorial staff and cross-referenced to many bioinformatics databases. A system of evidence tracking ensures that all assertions are backed up by the primary literature. Reactome is used by clinicians, geneticists, genomics researchers, and molecular biologists to interpret the results of high-throughput experimental studies, by bioinformaticians seeking to develop novel algorithms for mining knowledge from genomic studies, and by systems biologists building predictive models of normal and disease variant pathways.
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Fabregat, A., Sidiropoulos, K., Viteri, G., Forner, O., Marin-Garcia, P., Arnau, V. et al. (2017). Reactome pathway analysis: a high-performance in-memory approach. *BMC bioinformatics, 18*, 142. ↗

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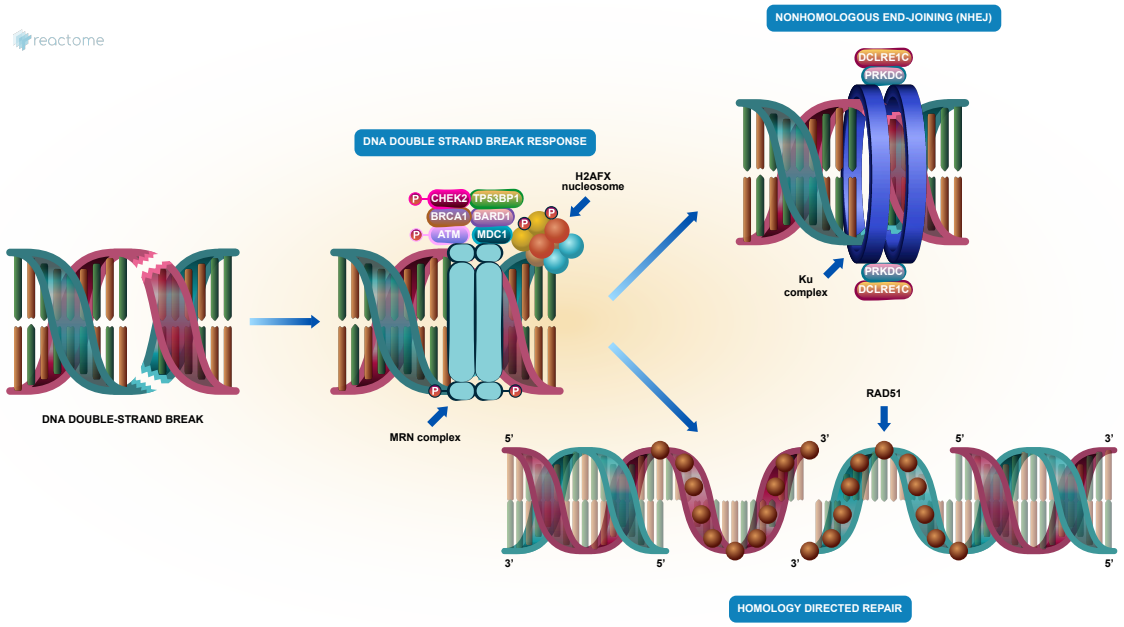
DNA Double-Strand Break Repair ↗

Stable identifier: R-HSA-5693532

Compartments: nucleoplasm



Double-strand breaks (DSBs), one of the most deleterious types of DNA damage along with interstrand crosslinks, are caused by ionizing radiation or certain chemicals such as bleomycin. DSBs also occur physiologically, during the processes of DNA replication, meiotic exchange, and V(D)J recombination.

DSBs are sensed (detected) by the MRN complex. Binding of the MRN complex to the DSBs usually triggers ATM kinase activation, thus initiating the DNA double strand break response. ATM phosphorylates a number of proteins involved in DNA damage checkpoint signaling, as well as proteins directly involved in the repair of DNA DSBs. DSBs are repaired via homology directed repair (HDR) or via nonhomologous end-joining (NHEJ).

HDR requires resection of DNA DSB ends. Resection creates 3'-ssDNA overhangs which then anneal with a homologous DNA sequence. This homologous sequence can then be used as a template for DNA repair synthesis that bridges the DSB. HDR preferably occurs through the error-free homologous recombination repair (HRR), but can also occur through the error-prone single strand annealing (SSA), or the least accurate microhomology-mediated end joining (MMEJ). MMEJ takes place when DSB response cannot be initiated.

While HRR is limited to actively dividing cells with replicated DNA, error-prone NHEJ pathway functions at all stages of the cell cycle, playing the predominant role in both the G1 phase and in S-phase regions of DNA that have not yet replicated. During NHEJ, the Ku70:Ku80 heterodimer (also known as the Ku complex or XRCC5:XRCC6) binds DNA DSB ends, competing away the MRN complex and preventing MRN-mediated resection of DNA DSB ends. The catalytic subunit of the DNA-dependent protein kinase (DNA-PKcs, PRKDC) is then recruited to DNA-bound Ku to form the DNA-PK holoenzyme. Two DNA-PK complexes, one at each side of the break, bring DNA DSB ends together, joining them in a synaptic complex. DNA-PK complex recruits DCLRE1C (ARTEMIS) to DNA DSB ends, leading to trimming of 3'- and 5'-overhangs at the break site, followed by ligation.

For review of this topic, please refer to Ciccia and Elledge 2010.

Literature references

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Elledge, SJ., Ciccia, A. (2010). The DNA damage response: making it safe to play with knives. *Mol. Cell, 40*, 179-204. ↗

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2003-07-14	Authored	Thompson, L., Lees-Miller, S.
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2015-05-12	Authored, Edited, Revised	Orlic-Milacic, M.
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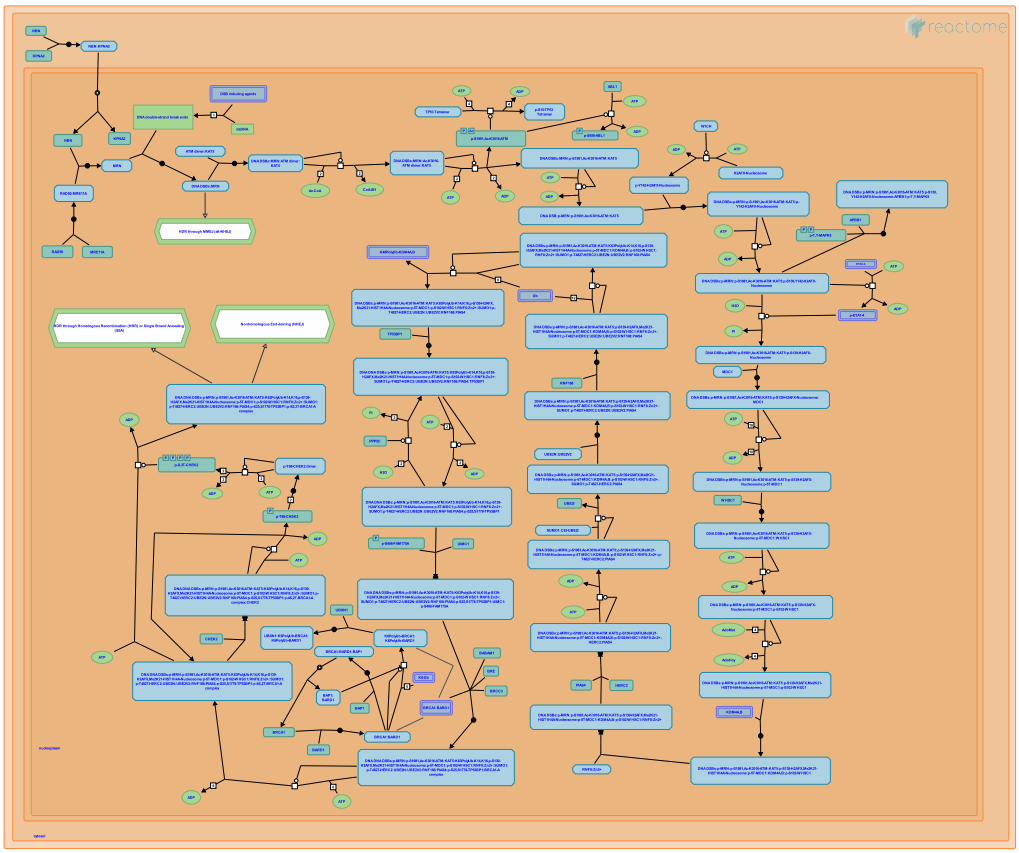
DNA Double Strand Break Response ↗

Location: DNA Double-Strand Break Repair

Stable identifier: R-HSA-5693606



DNA double-strand break (DSB) response involves sensing of DNA DSBs by the MRN complex which triggers ATM activation. ATM phosphorylates a number of proteins involved in DNA damage checkpoint signaling, as well as proteins directly involved in the repair of DNA DSBs. For a recent review, please refer to Ciccia and Elledge, 2010.

Literature references

Elledge, SJ., Ciccia, A. (2010). The DNA damage response: making it safe to play with knives. *Mol. Cell, 40*, 179-204. ↗

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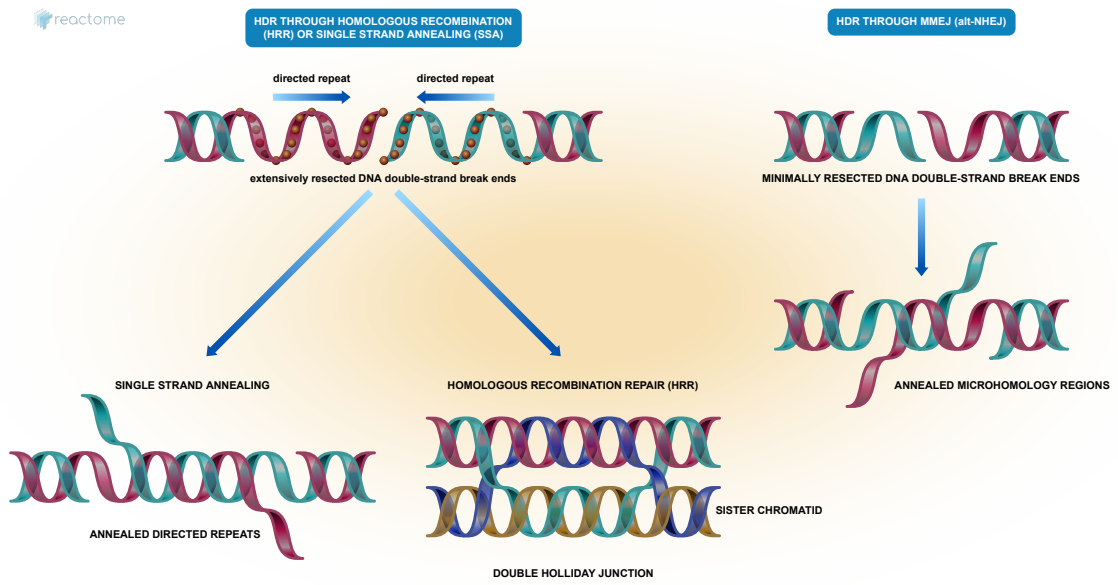
Homology Directed Repair ↗

Location: DNA Double-Strand Break Repair

Stable identifier: R-HSA-5693538

Compartments: nucleoplasm



Homology directed repair (HDR) of DNA double strand breaks (DSBs) requires resection of DNA DSB ends. Resection creates 3'-ssDNA overhangs which then anneal with a homologous DNA sequence. This homologous sequence can then be used as a template for DNA repair synthesis that bridges the DSB. HDR preferably occurs through the error-free homologous recombination repair (HRR), but can also occur through the error-prone single strand annealing (SSA), or the least accurate microhomology-mediated end joining (MMEJ).

HRR and SSA share the initial steps that involve ATM signaling, formation of the so-called ionizing radiation-induced foci (IRIF), extensive resection of DNA DSB ends and activation of ATR signaling. In homologous recombination, 3'-ssDNA overhangs anneal with complementary sister chromatid strands. In SSA, 3'-ssDNA overhangs anneal with each other through homologous direct repeats contained in each overhang, resulting in deletions of one of the repeats and the DNA sequence in between the repeats during DNA repair synthesis.

Contrary to HRR and SSA, which both involve annealing of long stretches of highly homologous DNA sequences, MMEJ entails annealing of short regions of two 3'-ssDNA overhangs (up to 20 nucleotides) and is therefore more promiscuous and more likely to join unrelated DNA molecules. The error rate of MMEJ is additionally increased by the low fidelity of the DNA polymerase theta (POLQ), which performs DNA repair synthesis in MMEJ.

For reviews of this topic, please refer to Khanna 2001, Thompson and Schild 2001, Thompson and Schild 2002, Thompson and Limoli 2003, Ciccia and Elledge 2010.

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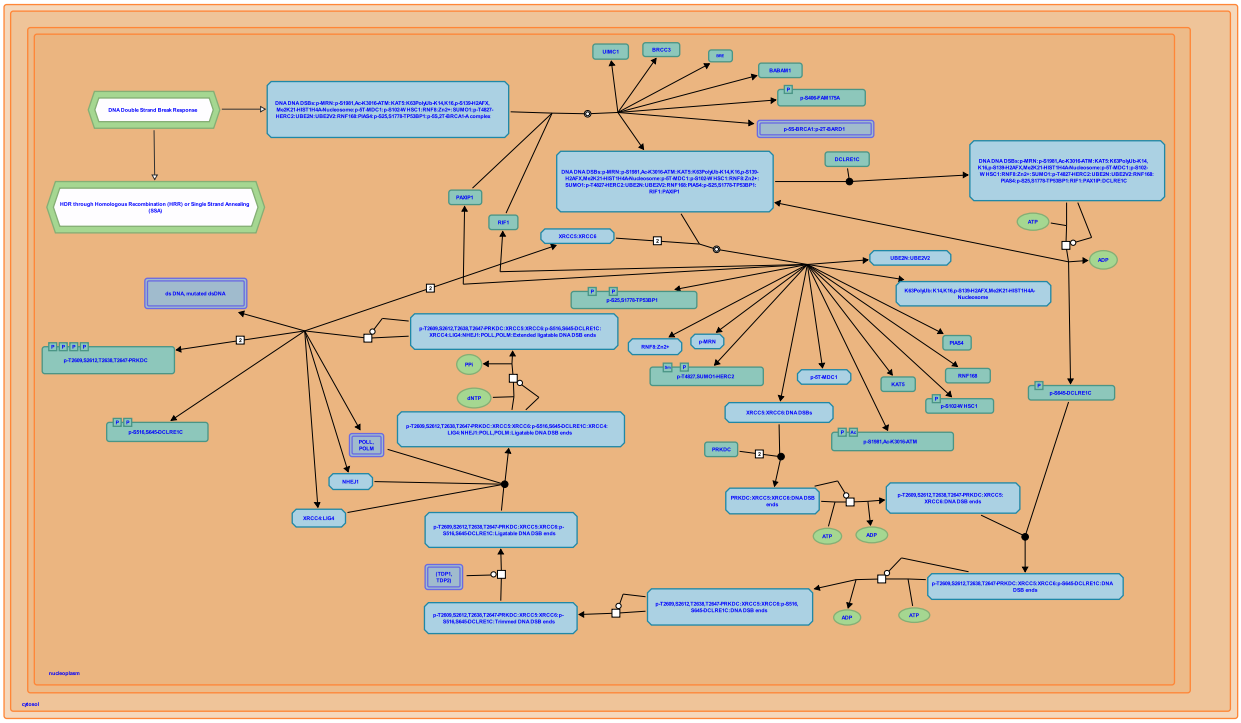
Nonhomologous End-Joining (NHEJ) ↗

Location: DNA Double-Strand Break Repair

Stable identifier: R-HSA-5693571

Compartments: nucleoplasm



reactome

The nonhomologous end joining (NHEJ) pathway is initiated in response to the formation of DNA double-strand breaks (DSBs) induced by DNA-damaging agents, such as ionizing radiation. DNA DSBs are recognized by the MRN complex (MRE11A:RAD50:NBN), leading to ATM activation and ATM-dependent recruitment of a number of DNA damage checkpoint and repair proteins to DNA DSB sites (Lee and Paull 2005). The ATM phosphorylated MRN complex, MDC1 and H2AFX-containing nucleosomes (gamma-H2AX) serve as scaffolds for the formation of nuclear foci known as ionizing radiation induced foci (IRIF) (Gatei et al. 2000, Paull et al. 2000, Stewart et al. 2003, Stucki et al. 2005). Ultimately, both BRCA1:BARD1 heterodimers and TP53BP1 (53BP1) are recruited to IRIF (Wang et al. 2007, Pei et al. 2011, Mallette et al. 2012), which is necessary for ATM-mediated CHEK2 activation (Wang et al. 2002, Wilson et al. 2008). In G1 cells, TP53BP1 promotes NHEJ by recruiting RIF1 and PAX1IP, which displaces BRCA1:BARD1 and associated proteins from the DNA DSB site and prevents resection of DNA DSBs needed for homologous recombination repair (HRR) (Escribano-Diaz et al. 2013, Zimmermann et al. 2013, Callen et al. 2013). TP53BP1 also plays an important role in ATM-mediated phosphorylation of DCLRE1C (ARTEMIS) (Riballo et al. 2004, Wang et al. 2014). Ku70:Ku80 heterodimer (also known as the Ku complex or XRCC5:XRCC6) binds DNA DSB ends, competing away the MRN complex and preventing MRN-mediated resection of DNA DSB ends (Walker et al. 2001, Sun et al. 2012). The catalytic subunit of the DNA-dependent protein kinase (DNA-PKcs, PRKDC) is then recruited to DNA-bound Ku to form the DNA-PK holoenzyme. Two DNA-PK complexes, one at each side of the break, bring DNA DSB ends together, joining them in a synaptic complex (Gottlieb 1993, Yoo and Dynan 2000). DNA-PK complex recruits DCLRE1C (ARTEMIS) to DNA DSB ends (Ma et al. 2002). PRKDC-mediated phosphorylation of DCLRE1C, as well as PRKDC autophosphorylation, enables DCLRE1C to trim 3'- and 5'-overhangs at DNA DSBs, preparing them for ligation (Ma et al. 2002, Ma et al. 2005, Niewolik et al. 2006). The binding of inositol phosphate may additionally stimulate the catalytic activity of PRKDC (Hanakahi et al. 2000). Other factors, such as polynucleotide kinase (PNK), TDP1 or TDP2 may remove unligatable damaged nucleotides from 5'- and 3'-ends of the DSB, converting them to ligatable substrates (Inamdar et al. 2002, Gomez-Herreros et al. 2013). DNA ligase 4 (LIG4) in complex with XRCC4 (XRCC4:LIG4) is recruited to ligatable DNA DSB ends together with the XLF (NHEJ1) homodimer and DNA polymerases mu (POLM) and/or lambda (POLL) (McElhinny et al. 2000, Hsu et al. 2002, Malu et al. 2002, Ahnesorg et al. 2006, Mahajan et al. 2002, Lee et al. 2004, Fan and Wu 2004). After POLL and/or POLM fill 1- or 2-nucleotide long single strand gaps at aligned DNA DSB ends, XRCC4:LIG4 performs the ligation of broken DNA strands, thus completing NHEJ. The presence of NHEJ1 homodimer facilitates the ligation step, especially at mismatched DSB ends (Tsai et al. 2007). Depending on other types of DNA damage present at DNA DSBs, NHEJ can result in error-free products, produce dsDNA with microdeletions and/or mismatched bases, or result in translocations (reviewed by Povrik et al. 2012).

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