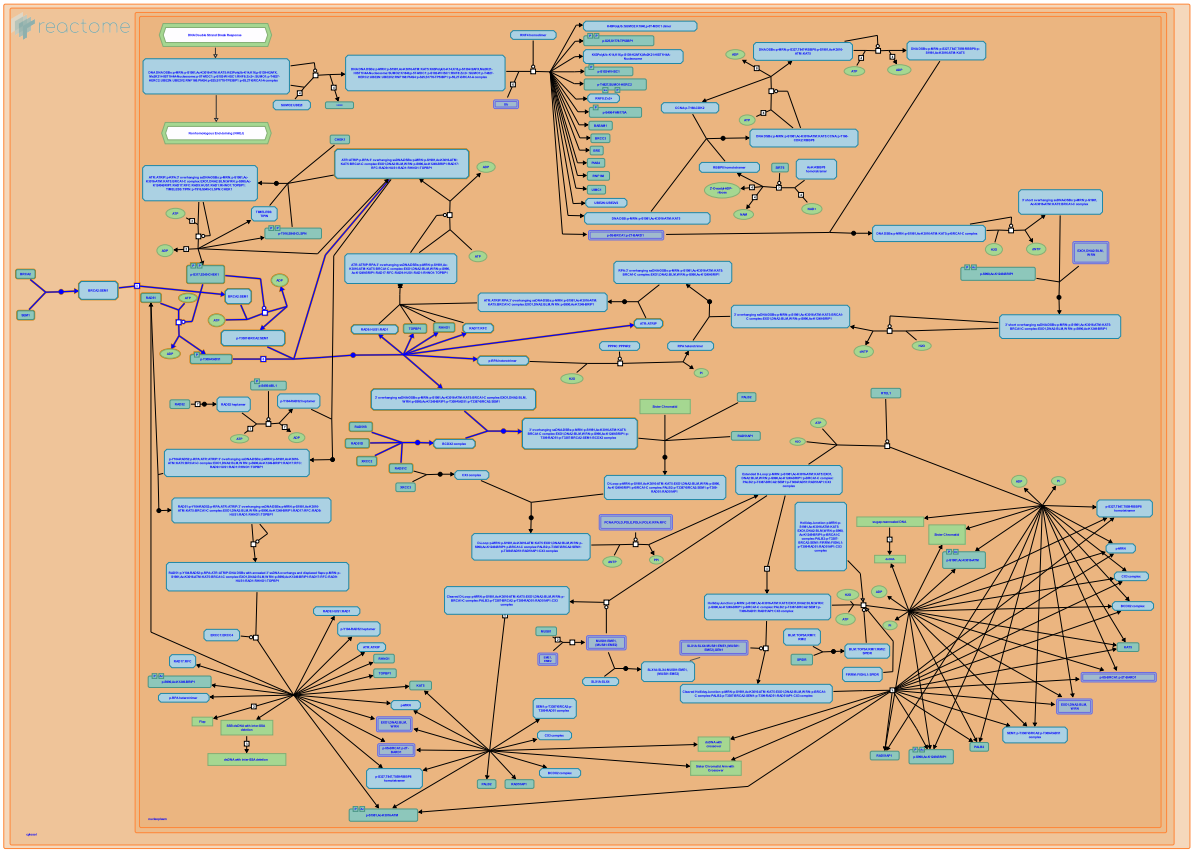
Presynaptic phase of homologous DNA pairing and strand exchange



Borowiec, JA., Heyer, WD., Le, HP., Liu, J., Matthews, L., Orlic-Milacic, M., Thompson, L.

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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.
The development of Reactome is supported by grants from the US National Institutes of Health (P41 HG003751), University of Toronto (CFREF Medicine by Design), European Union (EU STRP, EMI-CD), and the European Molecular Biology Laboratory (EBI Industry program).

Literature references

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. ↗

Sidiropoulos, K., Viteri, G., Sevilla, C., Jupe, S., Webber, M., Orlic-Milacic, M. et al. (2017). Reactome enhanced pathway visualization. *Bioinformatics, 33*, 3461-3467. ↗

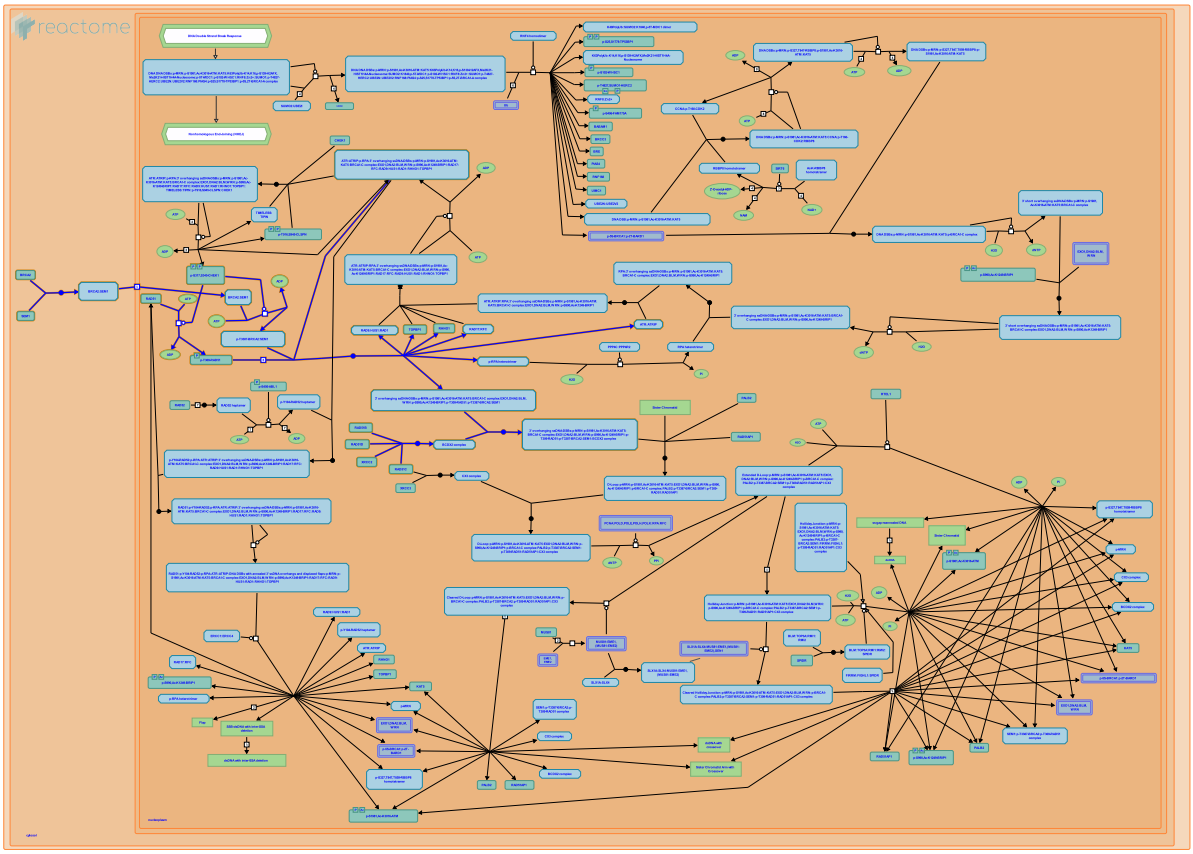
Fabregat, A., Jupe, S., Matthews, L., Sidiropoulos, K., Gillespie, M., Garapati, P. et al. (2018). The Reactome Pathway Knowledgebase. *Nucleic Acids Res, 46*, D649-D655. ↗

Fabregat, A., Korninger, F., Viteri, G., Sidiropoulos, K., Marin-Garcia, P., Ping, P. et al. (2018). Reactome graph database: Efficient access to complex pathway data. *PLoS computational biology, 14*, e1005968. ↗

Presynaptic phase of homologous DNA pairing and strand exchange ↗

Stable identifier: R-HSA-5693616

Compartments: nucleoplasm



The presynaptic phase of homologous DNA pairing and strand exchange during homologous recombination repair (HRR) begins with the displacement of RPA from ssDNA (Thompson and Limoli 2003) by the joint action of RAD51 and BRCA2. CHEK1-mediated phosphorylation of RAD51 and BRCA2 (Sorensen et al. 2005, Bahassi et al. 2008) is needed for BRCA2-mediated nucleation of RAD51 on 3'-ssDNA overhangs, RPA displacement and formation of RAD51 nucleofilaments (Yang et al. 2005, Jensen et al. 2010, Liu et al. 2010, Thorslund et al. 2010). Invasive RAD51 nucleofilaments are stabilized by the BCDX2 complex composed of RAD51B, RAD51C, RAD51D and XRCC2 (Masson et al. 2001, Chun et al. 2013, Amunugama et al. 2013).

Literature references

West, SC., Lekomtsev, S., Compton, SA., McIlwraith, MJ., Petronczki, M., Thorslund, T. et al. (2010). The breast cancer tumor suppressor BRCA2 promotes the specific targeting of RAD51 to single-stranded DNA. *Nat. Struct. Mol. Biol., 17*, 1263-5. ↗

Caldecott, KW. (2004). Origin, Recognition, Signaling and Repair of DNA Double-Strand Breaks in Mammalian Cells, Eukaryotic DNA Damage Surveillance and Repair. *Springer*, 1-40.

Buechelmaier, ES., Powell, SN., Chun, J. (2013). Rad51 paralog complexes BCDX2 and CX3 act at different stages in the BRCA1-BRCA2-dependent homologous recombination pathway. *Mol. Cell. Biol., 33*, 387-95. ↗

Ovesen, JL., Riesenberg, AL., Bahassi, EM., Stambrook, PJ., Hasty, PE., Bernstein, WZ. (2008). The checkpoint kinases Chk1 and Chk2 regulate the functional associations between hBRCA2 and Rad51 in response to DNA damage. *Oncogene, 27*, 3977-85. ↗

Benson, FE., Shah, R., Masson, JY., West, SC., Stasiak, AZ., Tarsounas, MC. et al. (2001). Identification and purification of two distinct complexes containing the five RAD51 paralogs. *Genes Dev., 15*, 3296-307. ↗

Editions

2003-07-14	Authored	Thompson, L.
2015-05-12	Authored, Edited, Revised	Orlic-Milacic, M.
2015-06-12	Reviewed	Borowiec, JA.

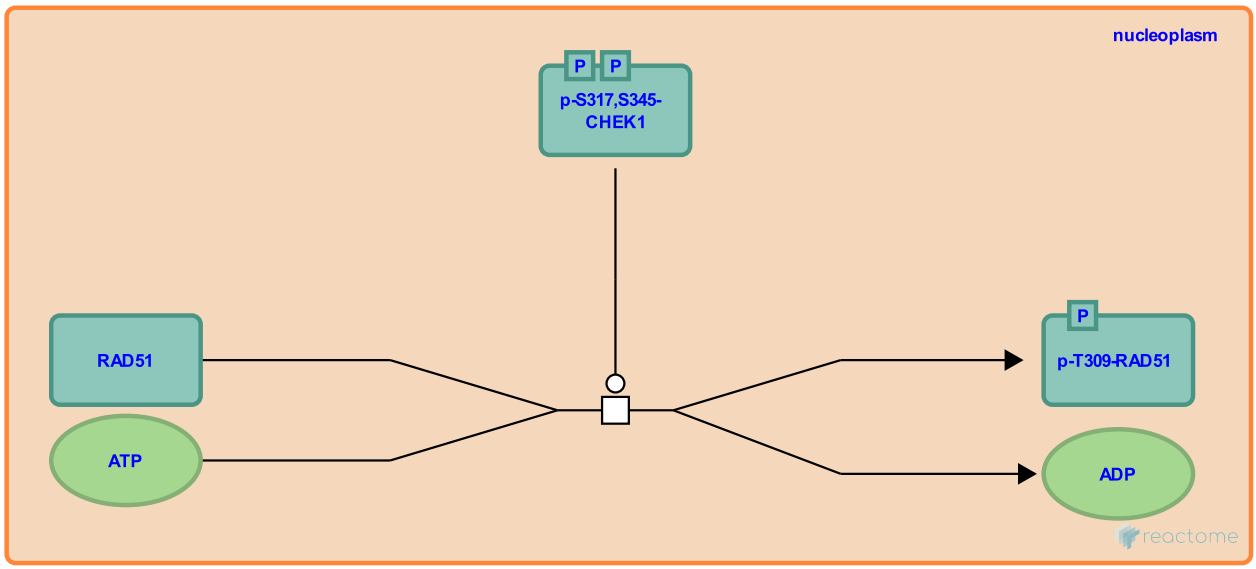
CHEK1 phosphorylates RAD51 ↗

Location: Presynaptic phase of homologous DNA pairing and strand exchange

Stable identifier: R-HSA-5685230

Type: transition

Compartments: nucleoplasm



Activated CHEK1 phosphorylates RAD51 on threonine residue T309, which is necessary for RAD51 association with chromatin (Sorensen et al. 2005).

Followed by: RAD51 binds BRCA2 at resected DNA DSBs

Literature references

Lundin, C., Bartek, J., Dziegielewski, J., Helleday, T., Hansen, LT., Sørensen, CS. et al. (2005). The cell-cycle checkpoint kinase Chk1 is required for mammalian homologous recombination repair. *Nat. Cell Biol., 7*, 195-201. ↗

Editions

2015-05-12	Authored, Edited	Orlic-Milacic, M.
2015-06-12	Reviewed	Borowiec, JA.

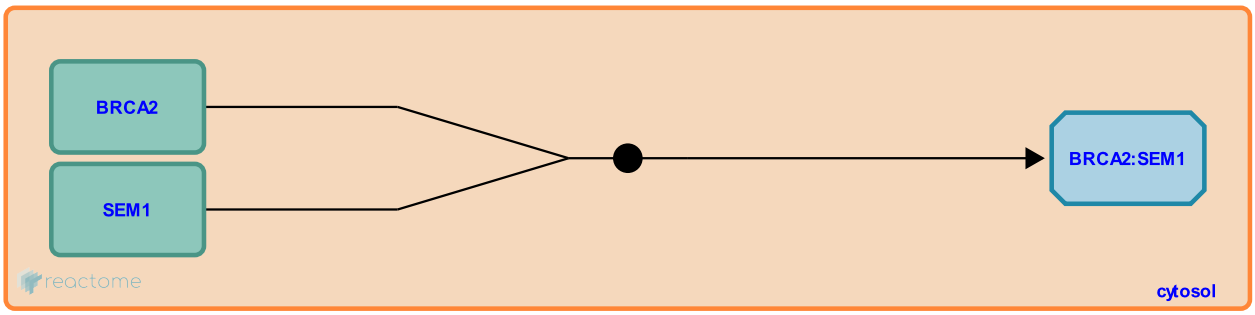
BRCA2 binds SEM1 (DSS1) ↗

Location: Presynaptic phase of homologous DNA pairing and strand exchange

Stable identifier: R-HSA-9763137

Type: binding

Compartments: cytosol



DSS1 is a critical and positive effector of BRCA2 function (Gudmundsdottir et al. 2004), which binds through the BRCA2 helical domain, OB1 and OB2 folds (Marston et al. 1999, Yang et al. 2002). Defective binding of DSS1 leads to reduced homologous recombination repair (HRR) activity of BRCA2 mutant constructs in mice (K2630A and K2630D) (Siaud et al. 2011). BRCA2 D2723H mutation is cancer-predisposing and leads to cytoplasmic mislocalization of BRCA2 due to wrongly exposed nuclear export signals as a consequence of defective DSS1 binding (Jeyasekhran et al. 2013). DSS1 controls the oligomeric state of BRCA2 and stabilizes the monomeric form of BRCA2 (Le et al. 2020). Defective DSS1 binding leads to intracellular oligomerization and cytosolic mislocalization (Le et al. 2020, Lee et al. 2021, reviewed in Le et al. 2021).

Followed by: BRCA2 translocates to the nucleus

Literature references

Shorthouse, D., Hall, BA., Venkitaraman, AR., Mahen, R., Lee, M. (2021). Cancer-causing BRCA2 missense mutations disrupt an intracellular protein assembly mechanism to disable genome maintenance. *Nucleic Acids Res, 49*, 5588-5604. ↗

Richards, WJ., Marston, NJ., Hughes, D., Ashworth, A., Marshall, CJ., Bertwistle, D. (1999). Interaction between the product of the breast cancer susceptibility gene BRCA2 and DSS1, a protein functionally conserved from yeast to mammals. *Mol Cell Biol, 19*, 4633-42. ↗

Ashworth, A., Tutt, AN., Gudmundsdottir, K., Witt, E., Lord, CJ. (2004). DSS1 is required for RAD51 focus formation and genomic stability in mammalian cells. *EMBO Rep, 5*, 989-93. ↗

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Siaud, N., Christ, N., Jasin, M., Xia, B., Barbera, MA., Egashira, A. et al. (2011). Plasticity of BRCA2 function in homologous recombination: genetic interactions of the PALB2 and DNA binding domains. *PLoS Genet, 7*, e1002409. ↗

Editions

2022-01-26	Reviewed	Liu, J., Heyer, WD., Le, HP.
2022-01-26	Authored	Orlic-Milacic, M.
2022-02-04	Edited	Orlic-Milacic, M.

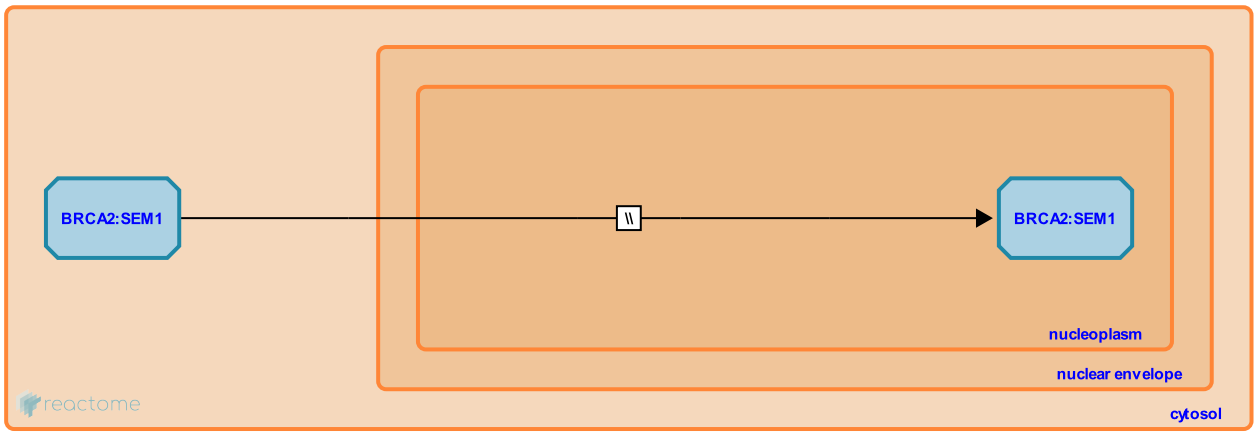
BRCA2 translocates to the nucleus ↗

Location: Presynaptic phase of homologous DNA pairing and strand exchange

Stable identifier: R-HSA-9709273

Type: omitted

Compartments: nucleoplasm, cytosol



BRCA2 possesses two functional nuclear localization signals (NLSs) in its C-terminal region. NLS1 has been mapped to amino acid residues 3263-3269, while NLS2 has been mapped to residues 3381-3385 (Spain et al. 1999, Yano et al. 2000). A third putative NLS maps to residues 3311-3317, however it was shown to be nonfunctional (Spain et al. 1999, Yano et al. 2000). NLS1 is essential for localization of BRCA2 to the nucleus, while NLS2 is not essential.

Binding of BRCA2 to SEM1 (DSS1) is needed for the translocation of BRCA2 to the nucleus and nuclear retention. SEM1 prevents BRCA2 oligomerization, which leads to cytosolic retention (Lee et al. 2021). Binding of SEM1 to BRCA2 masks the nuclear export signal of BRCA2, resulting in nuclear retention of BRCA2 (Jeyasekharan et al. 2013).

Preceded by: BRCA2 binds SEM1 (DSS1)

Followed by: CHEK1 phosphorylates BRCA2

Literature references

Shorthouse, D., Hall, BA., Venkitaraman, AR., Mahen, R., Lee, M. (2021). Cancer-causing BRCA2 missense mutations disrupt an intracellular protein assembly mechanism to disable genome maintenance. *Nucleic Acids Res, 49*, 5588-5604. ↗

Shihabuddin, LS., Spain, BH., Larson, CJ., Gage, FH., Verma, IM. (1999). Truncated BRCA2 is cytoplasmic: implications for cancer-linked mutations. *Proc Natl Acad Sci U S A, 96*, 13920-5. ↗

Yano, K., Miki, Y., Matsuo, F., Morotomi, K., Kato, M., Saito, H. (2000). Nuclear localization signals of the BRCA2 protein. *Biochem Biophys Res Commun, 270*, 171-5. ↗

Liu, Y., Rajendra, E., Jonsdottir, AB., Ayoub, N., Sato, K., Savill, J. et al. (2013). A cancer-associated BRCA2 mutation reveals masked nuclear export signals controlling localization. *Nat Struct Mol Biol, 20*, 1191-8. ↗

Editions

2020-12-11	Authored	Orlic-Milacic, M.
2022-01-26	Reviewed	Liu, J., Heyer, WD., Le, HP.
2022-02-04	Edited	Orlic-Milacic, M.

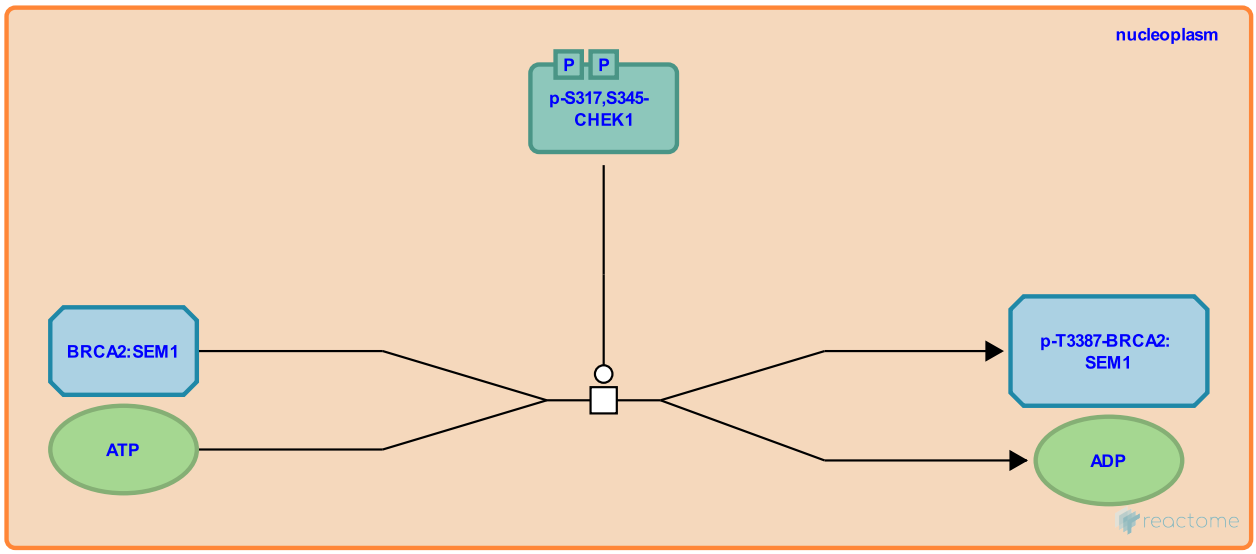
CHEK1 phosphorylates BRCA2 ↗

Location: Presynaptic phase of homologous DNA pairing and strand exchange

Stable identifier: R-HSA-5685242

Type: transition

Compartments: nucleoplasm



CHEK1 phosphorylates BRCA2 on threonine residue T3887, in the C-terminal region of BRCA2. CHEK1-mediated BRCA2 phosphorylation, as well as CHEK1 mediated RAD51 phosphorylation, promotes the association of BRCA2 with RAD51 (Bahassi et al. 2008).

Preceded by: BRCA2 translocates to the nucleus

Followed by: RAD51 binds BRCA2 at resected DNA DSBs

Literature references

Ovesen, JL., Riesenberg, AL., Bahassi, EM., Stambrook, PJ., Hasty, PE., Bernstein, WZ. (2008). The checkpoint kinases Chk1 and Chk2 regulate the functional associations between hBRCA2 and Rad51 in response to DNA damage. *Oncogene, 27*, 3977-85. ↗

Editions

2015-05-12	Authored, Edited	Orlic-Milacic, M.
2015-06-12	Reviewed	Borowiec, JA.
2022-01-31	Edited	Orlic-Milacic, M.

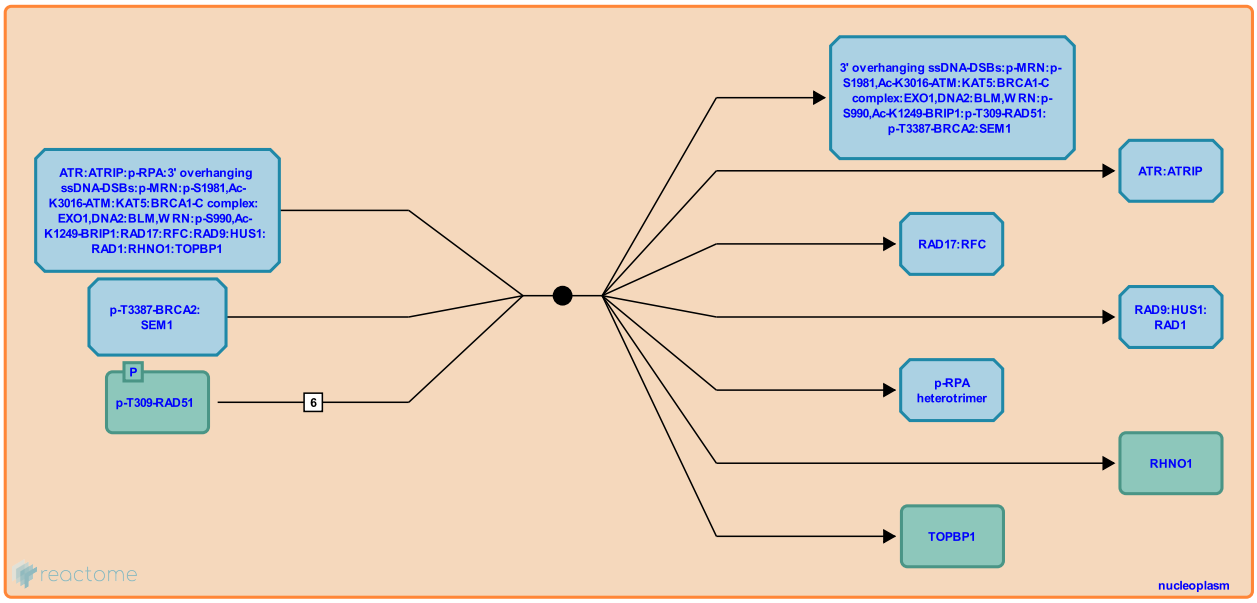
RAD51 binds BRCA2 at resected DNA DSBs ↗

Location: Presynaptic phase of homologous DNA pairing and strand exchange

Stable identifier: R-HSA-5693561

Type: binding

Compartments: nucleoplasm



BRCA2 and RAD51 interact directly through the highly conserved BRC repeats in BRCA2 (Venkitaraman 2002). CHEK1-mediated phosphorylation of BRCA2 (at threonine residue T3387) and RAD51 (at threonine residue T309) modulates their binding (Sorensen et al. 2005, Bahassi et al. 2008). One BRCA2 can bind up to six RAD51 molecules, thus playing an important role in RAD51 nucleation at the dsDNA-ssDNA junction created by resection of DNA double-strand breaks (DSBs) (Liu et al. 2010, Thorslund et al. 2010, Jensen et al. 2010). After the nucleation step, additional RAD51 molecules bind the ssDNA and multimerize, forming RAD51 nucleoprotein filaments (Yang et al. 2005). BRCA2-mediated RAD51 loading displaces the RPA complex from 3' overhanging ssDNA at DSBs (Liu et al. 2010, Jensen et al. 2010), possibly with other RPA-bound proteins, such as ATR:ATRIP and complexes involved in ATR catalytic activation.

BRCA2-binding partner SEM1 (DSS1) directly facilitates RPA release from ssDNA and its replacement with RAD51 (Zhao et al. 2015).

The interaction with DNA is critical for BRCA2 function, and the protein contains two different DNA binding regions. The amino-terminal DNA-binding domain (NTD) (residue 267-350) consists of a zinc finger-poly(adenosine diphosphate-ribose) polymerase (PARP)-like domain and binds to different DNA structures (ssDNA, dsDNA, tailed DNA, and gapped DNA) with similar affinity (von Nicolai et al. 2016). The carboxy-terminal DNA-binding domain CTD (residue 2,474-3,190) is significantly more complex with three OB-folds and an apical 3 bundle helix on top of a tower domain preceded by a helical domain (Yang et al. 2002). The proposed breast and ovarian cancer cluster regions (BCCR and OCCR) overlap with the CTD (Rebbeck et al. 2015, Gayther et al. 1997). Several common pathogenic mutations (ClinVar database) and loss-of-function mutations are found in this region including K2630A, K2630D, D2723H.

Preceded by: CHEK1 phosphorylates BRCA2, CHEK1 phosphorylates RAD51

Followed by: BCDX2 complex stabilizes RAD51 filament

Literature references

West, SC., Lekomtsev, S., Compton, SA., McIlwraith, MJ., Petronczki, M., Thorslund, T. et al. (2010). The breast cancer tumor suppressor BRCA2 promotes the specific targeting of RAD51 to single-stranded DNA. *Nat. Struct. Mol. Biol., 17*, 1263-5. ↗

Venkitaraman, AR. (2002). Cancer susceptibility and the functions of BRCA1 and BRCA2. *Cell, 108*, 171-82. ↗

Ovesen, JL., Riesenberg, AL., Bahassi, EM., Stambrook, PJ., Hasty, PE., Bernstein, WZ. (2008). The checkpoint kinases Chk1 and Chk2 regulate the functional associations between hBRCA2 and Rad51 in response to DNA damage. *Oncogene, 27*, 3977-85. ↗

Holloman, WK., Li, Q., Pavletich, NP., Fan, J., Yang, H. (2005). The BRCA2 homologue Brh2 nucleates RAD51 filament formation at a dsDNA-ssDNA junction. *Nature, 433*, 653-7. ↗

Lundin, C., Bartek, J., Dziegielewski, J., Helleday, T., Hansen, LT., Sørensen, CS. et al. (2005). The cell-cycle checkpoint kinase Chk1 is required for mammalian homologous recombination repair. *Nat. Cell Biol., 7*, 195-201. ↗

Editions

2003-11-23	Authored	Matthews, L.
2015-05-12	Edited, Revised	Orlic-Milacic, M.
2015-06-12	Reviewed	Borowiec, JA.
2022-01-26	Reviewed	Liu, J., Heyer, WD., Le, HP.
2022-01-31	Edited	Orlic-Milacic, M.

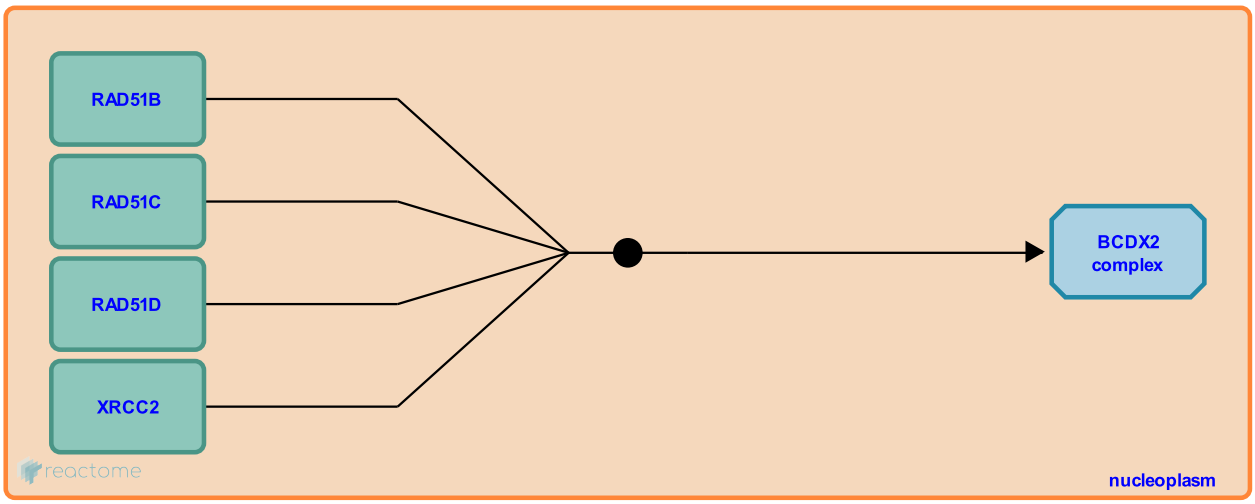
BCDX2 complex formation ↗

Location: Presynaptic phase of homologous DNA pairing and strand exchange

Stable identifier: R-HSA-5685318

Type: binding

Compartments: nucleoplasm



RAD51 paralogs RAD51B, RAD51C, RAD51D and XRCC2 form a complex named BCDX2 with 1:1:1:1 stoichiometry. In this complex, RAD51B directly interacts with RAD51C, which interacts with RAD51D, which interacts with XRCC2 (Masson et al. 2001, Chun et al. 2013).

Followed by: BCDX2 complex stabilizes RAD51 filament

Literature references

Buechelmaier, ES., Powell, SN., Chun, J. (2013). Rad51 paralog complexes BCDX2 and CX3 act at different stages in the BRCA1-BRCA2-dependent homologous recombination pathway. *Mol. Cell. Biol., 33*, 387-95. ↗

Benson, FE., Shah, R., Masson, JY., West, SC., Stasiak, AZ., Tarsounas, MC. et al. (2001). Identification and purification of two distinct complexes containing the five RAD51 paralogs. *Genes Dev., 15*, 3296-307. ↗

Editions

2015-05-12	Authored, Edited	Orlic-Milacic, M.
2015-06-12	Reviewed	Borowiec, JA.

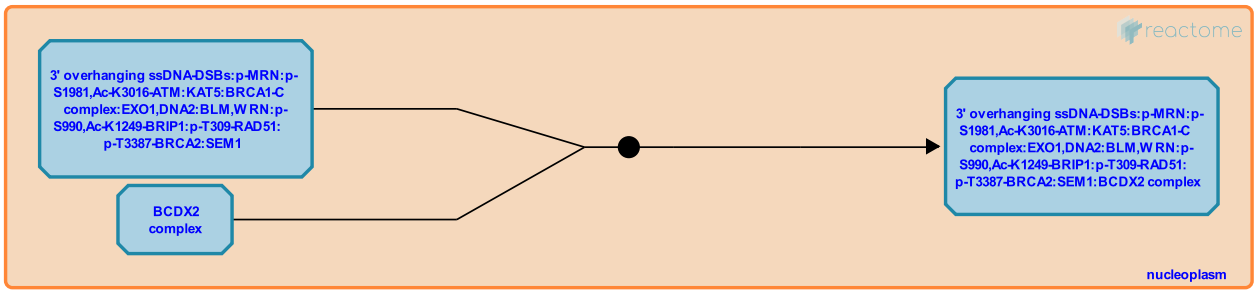
BCDX2 complex stabilizes RAD51 filament ↗

Location: Presynaptic phase of homologous DNA pairing and strand exchange

Stable identifier: R-HSA-5685341

Type: binding

Compartments: nucleoplasm



The BCDX2 complex, composed of RAD51 paralogs RAD51B, RAD51C, RAD51D and XRCC2, preferentially binds at the ends of 3' overhanging ssDNA created by resection of DNA double-strand breaks (DSBs). The BCDX2 complex stabilizes nucleoprotein filaments formed by BRCA2-mediated RAD51 loading onto ssDNA (Masson et al. 2001, Chun et al. 2013). The BCDX2 complex may act by inhibiting displacement of RAD51 by BLM helicase (Amunugama et al. 2013).

Preceded by: RAD51 binds BRCA2 at resected DNA DSBs, BCDX2 complex formation

Literature references

Buechelmaier, ES., Powell, SN., Chun, J. (2013). Rad51 paralog complexes BCDX2 and CX3 act at different stages in the BRCA1-BRCA2-dependent homologous recombination pathway. *Mol. Cell. Biol., 33*, 387-95. ↗

Benson, FE., Shah, R., Masson, JY., West, SC., Stasiak, AZ., Tarsounas, MC. et al. (2001). Identification and purification of two distinct complexes containing the five RAD51 paralogs. *Genes Dev., 15*, 3296-307. ↗

Fishel, R., Amunugama, R., Groden, J. (2013). The HsRAD51B-HsRAD51C stabilizes the HsRAD51 nucleoprotein filament. *DNA Repair (Amst.), 12*, 723-32. ↗

Editions

2015-05-12	Authored, Edited	Orlic-Milacic, M.
2015-06-12	Reviewed	Borowiec, JA.

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