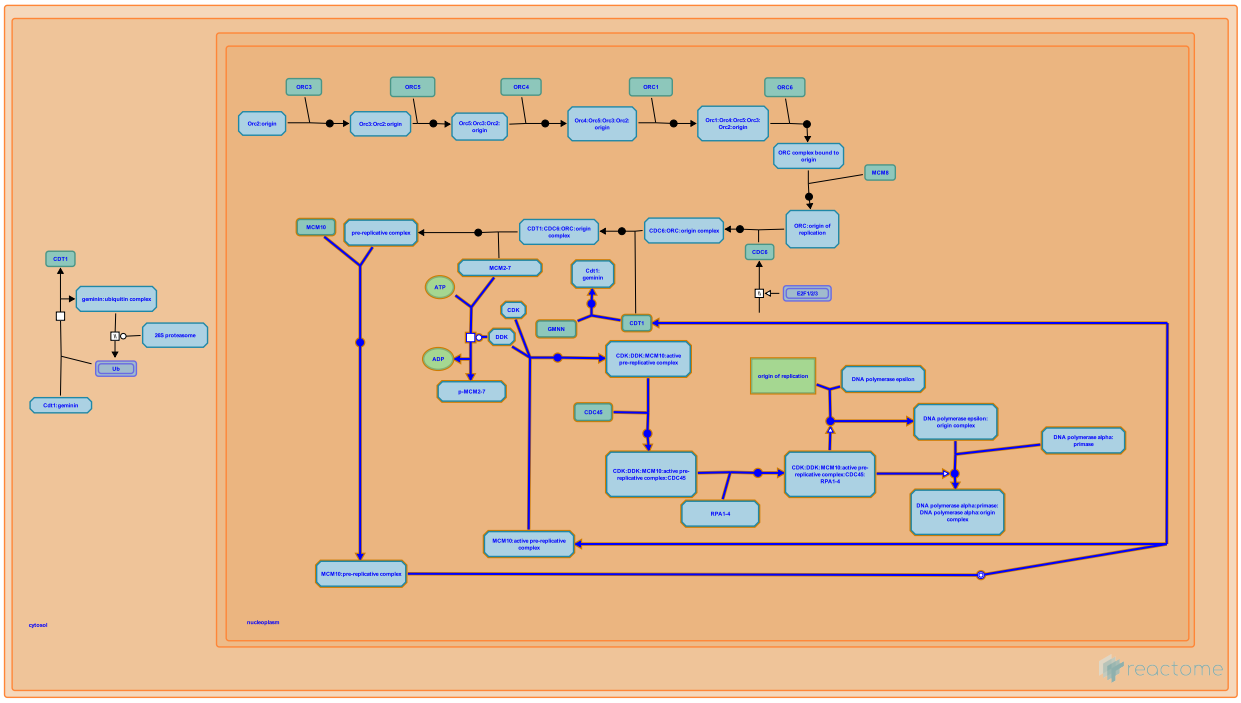
Activation of the pre-replicative complex



European Bioinformatics Institute, New York University Langone Medical Center, Ontario Institute for Cancer Research, Oregon Health and Science University.

20/01/2021

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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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. ↗

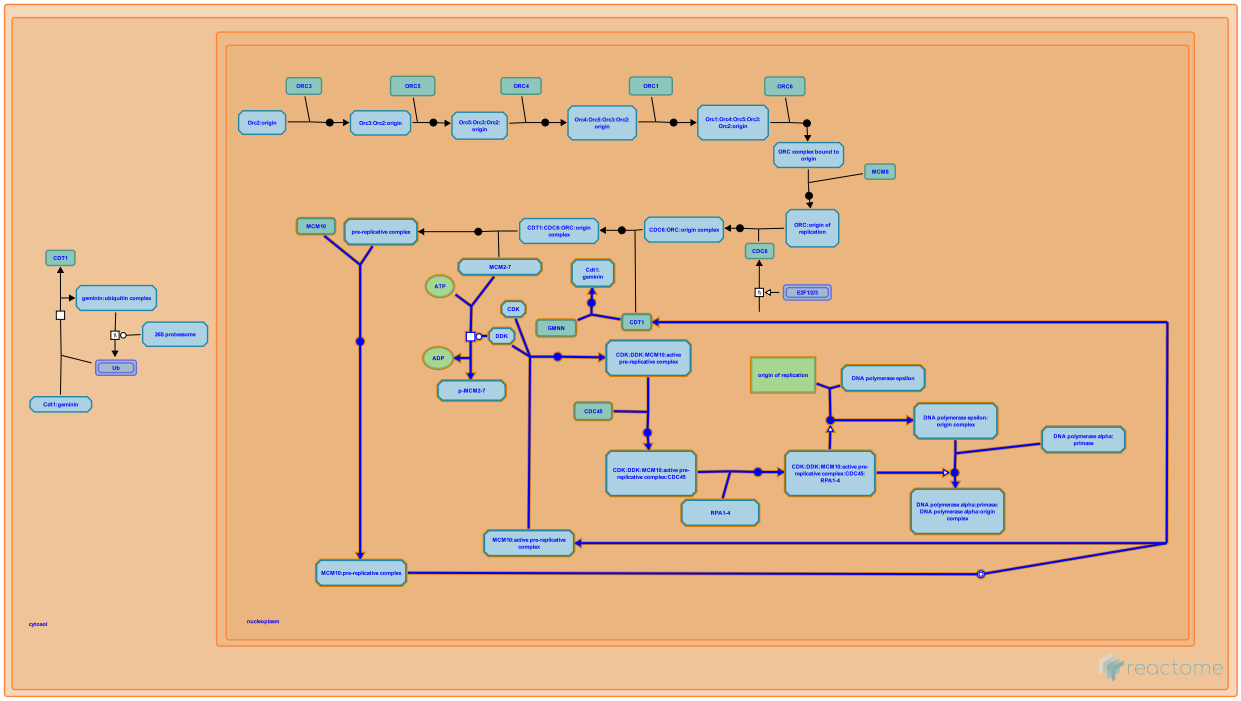
Fabregat, A., Jupe, S., Matthews, L., Sidiropoulos, K., Gillespie, M., Garapati, P. et al. (2018). The Reactome Pathway Knowledgebase. *Nucleic Acids Res, 46*, D649-D655. ↗

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. ↗

Activation of the pre-replicative complex ↗

Stable identifier: R-HSA-68962

Compartments: nucleoplasm



In S. cerevisiae, two ORC subunits, Orc1 and Orc5, both bind ATP, and Orc1 in addition has ATPase activity. Both ATP binding and ATP hydrolysis appear to be essential functions in vivo. ATP binding by Orc1 is unaffected by the association of ORC with origin DNA (ARS) sequences, but ATP hydrolysis is ARS-dependent, being suppressed by associated double-stranded DNA and stimulated by associated single-stranded DNA. These data are consistent with the hypothesis that ORC functions as an ATPase switch, hydrolyzing bound ATP and changing state as DNA unwinds at the origin immediately before replication. It is attractive to speculate that ORC likewise functions as a switch as human pre-replicative complexes are activated, but human Orc proteins are not well enough characterized to allow the model to be critically tested. mRNAs encoding human orthologs of all six Orc proteins have been cloned, and ATP-binding amino acid sequence motifs have been identified in Orc1, Orc4, and Orc5. Interactions among proteins expressed from the cloned genes have been characterized, but the ATP-binding and hydrolyzing properties of these proteins and complexes of them have not been determined.

Literature references

Klemm, RD., Austin, RJ., Bell, SP. (1997). Coordinate binding of ATP and origin DNA regulates the ATPase activity of the origin recognition complex. *Cell, 88*, 493-502. ↗

Tugal, T., Zou-Yang, XH., Gavin, K., Pappin, D., Canas, B., Kobayashi, R. et al. (1999). The Orc4p and Orc5p subunits of the Xenopus and human origin recognition complex are related to Orc1p and Cdc6p. *J Biol Chem, 273*, 32421-9. ↗

Klemm, RD., Bell, SP. (2001). ATP bound to the origin recognition complex is important for preRC formation. *Proc Natl Acad Sci U S A, 98*, 8361-7. ↗

Prasanth, SG., Prasanth, KV., Stillman, B. (2002). Orc6 involved in DNA replication, chromosome segregation, and cytokinesis. *Science, 297*, 1026-31. ↗

Quintana, DG., Hou, Zh., Thome, KC., Hendricks, M., Saha, P., Dutta, A. (1997). Identification of HsORC4, a member of the human origin of replication recognition complex. *J Biol Chem, 272*, 28247-51. ↗

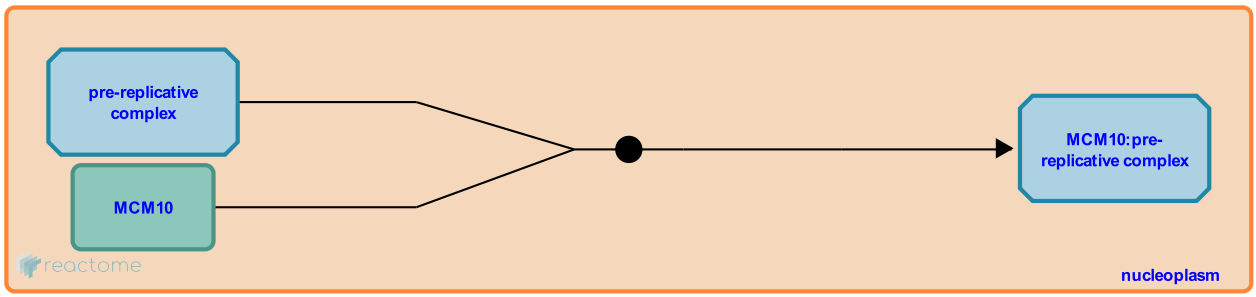
Mcm10 associates with the pre-replicative complex, stabilizing Mcm2-7 ↗

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-68919

Type: binding

Compartments: nucleoplasm



MCM10 is required for human DNA replication. In S. cerevisiae, Mcm10, like Mcm2-7, is required for minichromosome maintenance, but Mcm10 has no sequence homology with these other proteins (Merchant et al., 1997). Genetic studies have demonstrated that Mcm10 is required for DNA replication in S. pombe (Aves et al., 1998) and S. cerevisiae cells (Homesley et al., 2000) and immunodepletion of XlMcm10 interferes with DNA replication in Xenopus egg extracts (Wohlschlegel et al., 2002). Human Mcm10 interacts with chromatin in G1 phase and then dissociates during G2 phase. In S. cerevisiae, Mcm10 has been shown to localize to origins during G1 (Ricke and Bielinsky, 2004), and it may stabilize the association of Mcm2-7 with the pre-replicative complex (Sawyer et al., 2004). This timing of association is consistent with studies that demonstrate that, in Xenopus egg extracts, Mcm10 is required for association of Cdc45, but not Mcm2-7 with chromatin. Biochemical evidence that Mcm10 plays a direct role in the activation of the pre-replicative complex includes the requirement for SpMcm10 in the phosphorylation of the Mcm2-7 complex by DDK (Lee et al., 2004) and the fact that SpMcm10 binds and stimulates DNA polymerase alpha activity (Fien et al., 2004).

Followed by: Cdt1 is displaced from the pre-replicative complex.

Literature references

Izumi, M., Yanagi, K., Mizuno, T., Yokoi, M., Kawasaki, Y., Moon, KY. et al. (2000). The human homolog of Saccharomyces cerevisiae Mcm10 interacts with replication factors and dissociates from nuclease-resistant nuclear structures in G(2) phase. *Nucleic Acids Res, 28*, 4769-77. ↗

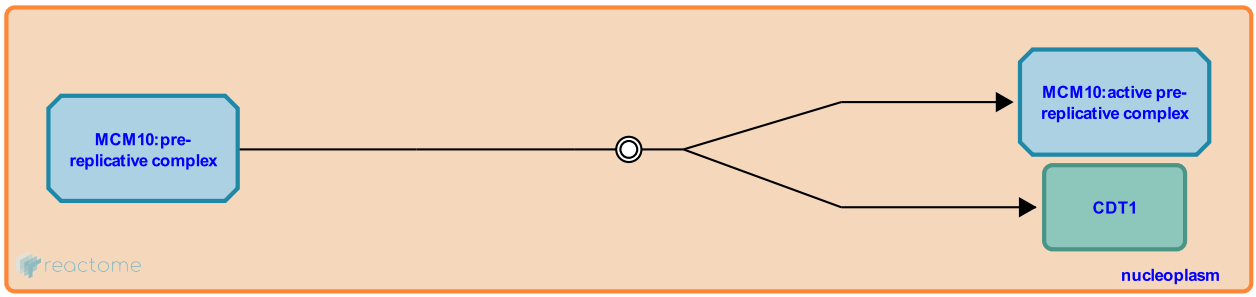
Cdt1 is displaced from the pre-replicative complex.

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-68940

Type: dissociation

Compartments: nucleoplasm



At the beginning of this reaction, 1 molecule of 'Mcm10:pre-replicative complex' is present. At the end of this reaction, 1 molecule of 'Mcm10:active pre-replicative complex', and 1 molecule of 'CDT1' are present.

This reaction takes place in the 'nucleus'.

Preceded by: Mcm10 associates with the pre-replicative complex, stabilizing Mcm2-7

Followed by: Cdt1 associates with geminin, CDK and DDK associate with the Mcm10:pre-replicative complex

Literature references

Bell, SP., Dutta, A. (2002). DNA replication in eukaryotic cells. *Annu Rev Biochem, 71*, 333-74.

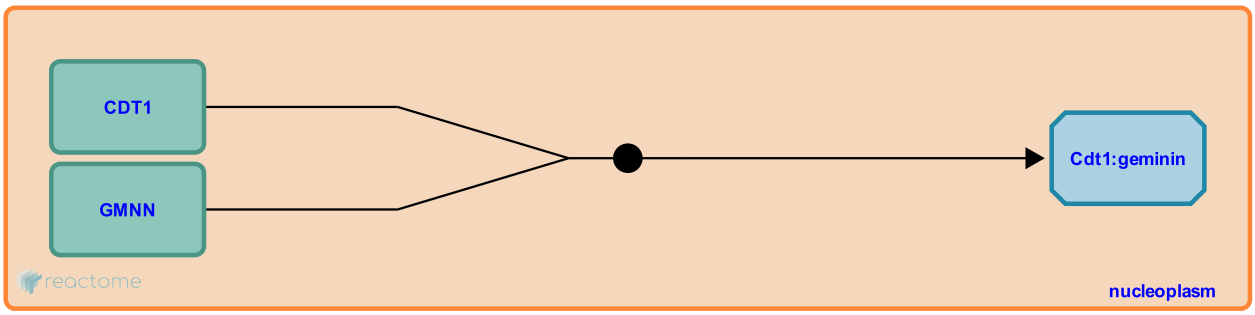
Cdt1 associates with geminin ↗

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-69299

Type: binding

Compartments: nucleoplasm



At the beginning of this reaction, 1 molecule of 'geminin', and 1 molecule of 'CDT1' are present. At the end of this reaction, 1 molecule of 'Cdt1:geminin' is present.

This reaction takes place in the 'nucleoplasm'.

Preceded by: Cdt1 is displaced from the pre-replicative complex.

Literature references

Wohlschlegel, JA., Dwyer, BT., Dhar, SK., Cvetic, C., Walter, JC., Dutta, A. (2000). Inhibition of eukaryotic DNA replication by geminin binding to Cdt1. *Science, 290*, 2309-12. ↗

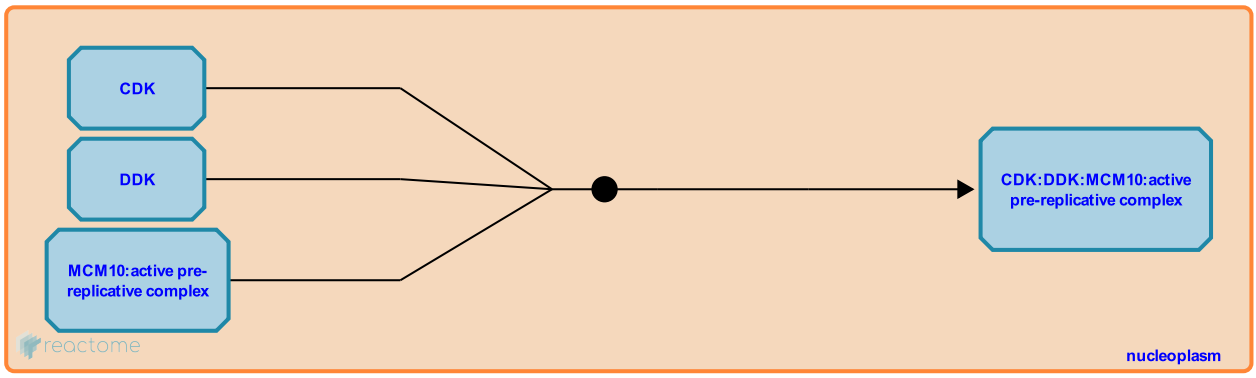
CDK and DDK associate with the Mcm10:pre-replicative complex ↗

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-68918

Type: binding

Compartments: nucleoplasm



At the beginning of this reaction, 1 molecule of 'Mcm10:active pre-replicative complex', 1 molecule of 'DDK', and 1 molecule of 'CDK' are present. At the end of this reaction, 1 molecule of 'CDK:DDK:Mcm10:pre-replicative complex' is present.

This reaction takes place in the 'nucleus'.

Preceded by: Cdt1 is displaced from the pre-replicative complex.

Followed by: Mcm2-7 is phosphorylated by DDK, Cdc45 associates with the pre-replicative complex at the origin

Literature references

Jiang, W., McDonald, D., Hope, TJ., Hunter, T. (1999). Mammalian Cdc7-Dbf4 protein kinase complex is essential for initiation of DNA replication. *EMBO J, 18*, 5703-13. ↗

Kumagai, H., Sato, N., Yamada, M., Mahony, D., Seghezzi, W., Lees, E. et al. (1999). A novel growth- and cell cycle-regulated protein, ASK, activates human Cdc7-related kinase and is essential for G1/S transition in mammalian cells. *Mol Cell Biol, 19*, 5083-95. ↗

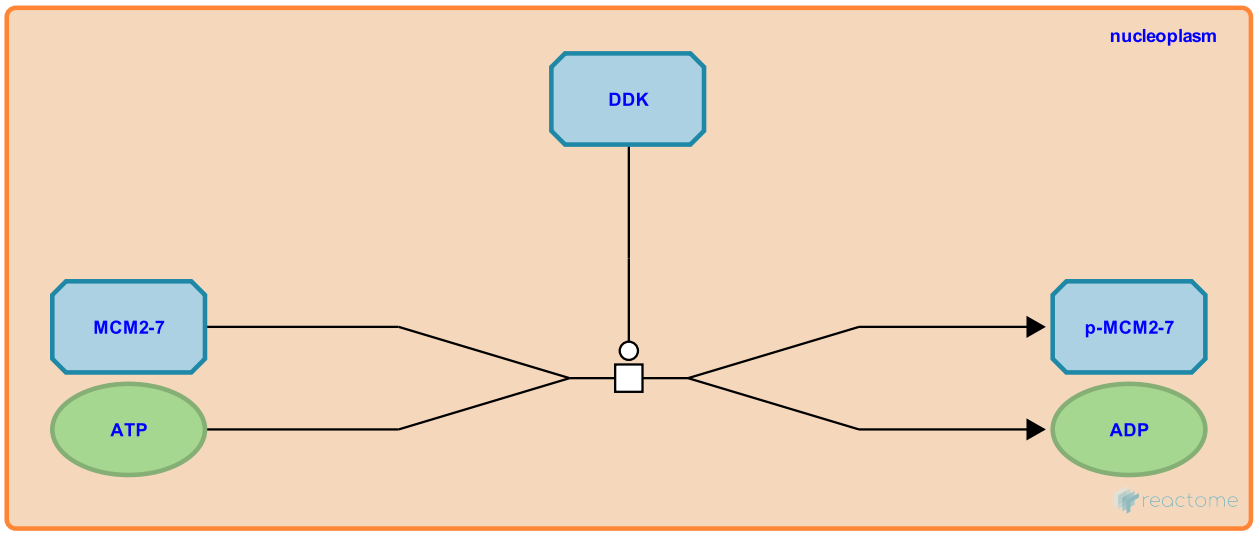
Mcm2-7 is phosphorylated by DDK ↗

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-68954

Type: transition

Compartments: nucleoplasm



At the beginning of this reaction, 1 molecule of 'Mcm2-7 complex', and 1 molecule of 'ATP' are present. At the end of this reaction, 1 molecule of 'phosphorylated Mcm2-7 complex', and 1 molecule of 'ADP' are present.

This reaction takes place in the 'nucleus' and is mediated by the 'kinase activity' of 'DDK'.

Preceded by: CDK and DDK associate with the Mcm10:pre-replicative complex

Literature references

Masai, H., Matsui, E., You, Z., Ishimi, Y., Tamai, K., Arai, K. (2000). Human Cdc7-related kinase complex. In vitro phosphorylation of MCM by concerted actions of Cdks and Cdc7 and that of a critical threonine residue of Cdc7 by Cdks. *J Biol Chem, 275*, 29042-52. ↗

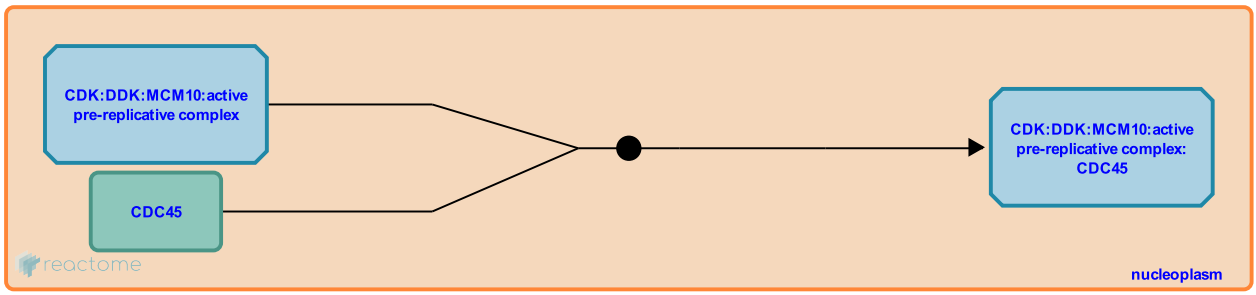
Cdc45 associates with the pre-replicative complex at the origin ↗

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-68917

Type: binding

Compartments: nucleoplasm



Once the Mcm2-7 complex has been assembled onto the origin of replication, the next step is the assembly of Cdc45, an essential replication protein, in late G1. The assembly of Cdc45 onto origins of replication forms a complex distinct from the pre-replicative complex, sometimes called the pre-initiation complex. The assembly of Cdc45 onto origins correlates with the time of initiation. Like the Mcm2-7 proteins, Cdc45 binds specifically to origins in the G1 phase of the cell cycle and then to non-origin DNA during S phase and is therefore thought to travel with the replication fork. Indeed, S. cerevisiae Cdc45 is required for DNA replication elongation as well as replication initiation. Cdc45 is required for the association of alpha DNA polymerase:primase with chromatin. Based on this observation and the observation that in S. cerevisiae, cCdc45 has been found in large complexes with some components of Mcm2-7 complex, it has been suggested that Cdc45 plays a scaffolding role at the replication fork, coupling Pol-alpha:primase to the replication fork through the helicase. Association of Cdc45 with origin DNA is regulated in the cell cycle and its association is dependent on the activity of cyclin-dependent kinases but not the Cdc7/Dbf4 kinase. In Xenopus egg extracts, association of Cdc45 with chromatin is dependent on Xmus101. TopBP1, the human homolog of Xmus1, is essential for DNA replication and interacts with DNA polymerase epsilon, one of the polymerases involved in replicating the genome. TopBP1 homologs have been found in S. cerevisiae and S. pombe. Sld3, an additional protein required for Cdc45 association with chromatin in S. cerevisiae and S. pombe, has no known human homolog.

Preceded by: CDK and DDK associate with the Mcm10:pre-replicative complex

Followed by: DNA Replication Factor A (RPA) associates with the pre-replicative complex at the origin

Literature references

Kukimoto, I., Igaki, H., Kanda, T. (1999). Human CDC45 protein binds to minichromosome maintenance 7 protein and the p70 subunit of DNA polymerase alpha. *Eur J Biochem, 265*, 936-43. ↗

Saha, P., Thome, KC., Yamaguchi, R., Hou, Z., Weremowicz, S., Dutta, A. (1998). The human homolog of Saccharomyces cerevisiae CDC45. *J Biol Chem, 273*, 18205-9. ↗

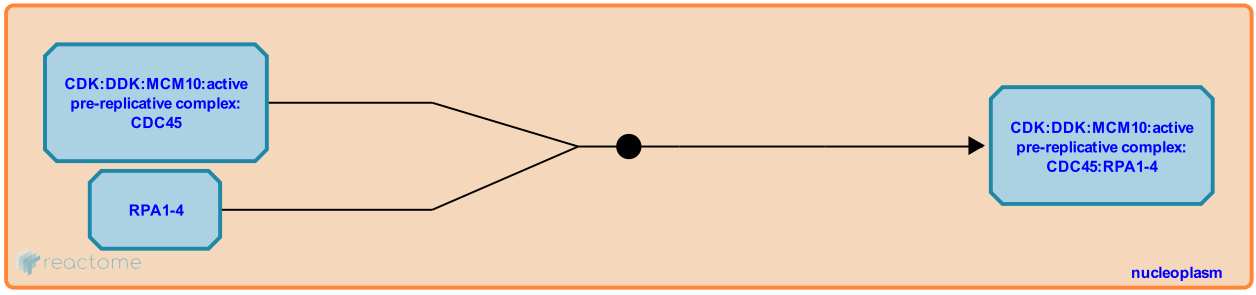
DNA Replication Factor A (RPA) associates with the pre-replicative complex at the origin ↗

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-68916

Type: binding

Compartments: nucleoplasm



After pre-RC assembly and Cdc45 association with the origin of replication, Replication Protein A (RPA) also associates with chromatin. RPA is a heterotrimeric complex containing p70, p34, and p11 subunits, and also is required for DNA recombination and DNA repair. The p70 subunit of RPA binds to the primase subunits of Pol alpha:primase. The p70 and p34 subunits of RPA are phosphorylated in a cell cycle-dependent manner. RPA is a single-strand DNA (ssDNA) binding protein and its association with chromatin at this stage suggests that DNA is partially unwound. This suggestion has been confirmed by detection of ssDNA in budding yeast origins of replication using chemical methods.

Preceded by: Cdc45 associates with the pre-replicative complex at the origin

Followed by: DNA polymerase alpha:primase binds at the origin, DNA polymerase epsilon binds at the origin

Literature references

Zou, L., Stillman, B. (2000). Assembly of a complex containing Cdc45p, replication protein A, and Mcm2p at replication origins controlled by S-phase cyclin-dependent kinases and Cdc7p-Dbf4p kinase. *Mol Cell Biol, 20*, 3086-96. ↗

Walter, J., Newport, J. (2000). Initiation of eukaryotic DNA replication: origin unwinding and sequential chromatin association of Cdc45, RPA, and DNA polymerase alpha. *Mol Cell, 5*, 617-27. ↗

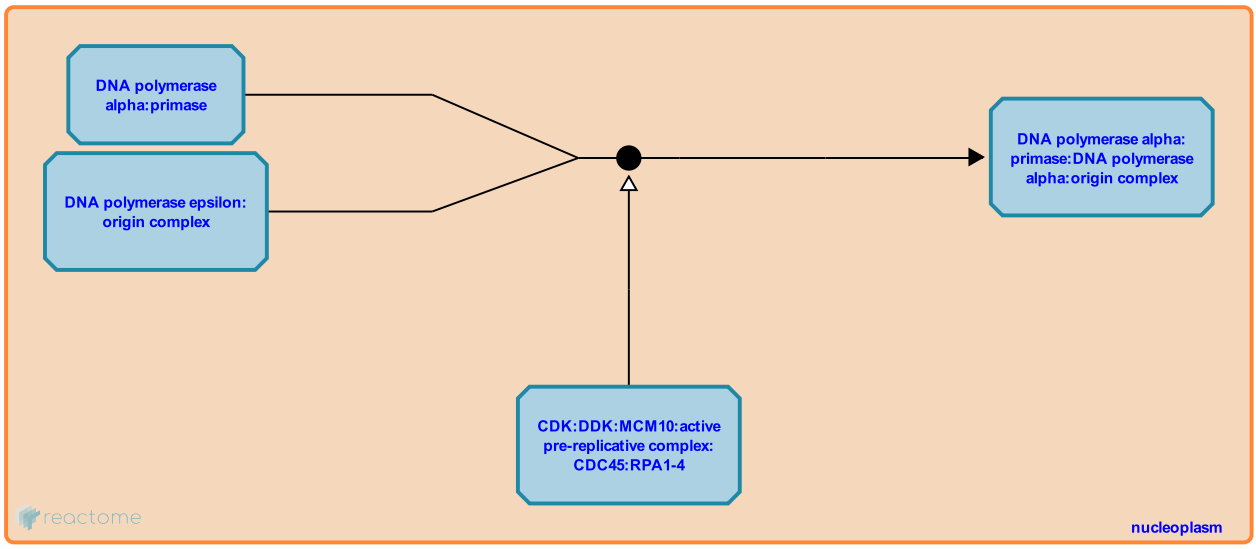
DNA polymerase alpha:primase binds at the origin ↗

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-68914

Type: binding

Compartments: nucleoplasm



DNA polymerase alpha:primase is comprised of four subunits, p180, p70, p58, and p49. The two primase subunits, p58 and p49, form a tight complex. The p49 subunit contains the DNA primase activity and one role of p58 appears to be tethering p49 to p180, the DNA polymerase catalytic subunit. The fourth subunit, p70, binds p180 and may tether the DNA polymerase alpha:primase complex to Cdc45.

Preceded by: DNA Replication Factor A (RPA) associates with the pre-replicative complex at the origin, DNA polymerase epsilon binds at the origin

Literature references

Zou, L., Stillman, B. (2000). Assembly of a complex containing Cdc45p, replication protein A, and Mcm2p at replication origins controlled by S-phase cyclin-dependent kinases and Cdc7p-Dbf4p kinase. *Mol Cell Biol, 20*, 3086-96. ↗

Walter, J., Newport, J. (2000). Initiation of eukaryotic DNA replication: origin unwinding and sequential chromatin association of Cdc45, RPA, and DNA polymerase alpha. *Mol Cell, 5*, 617-27. ↗

Bell, SP., Dutta, A. (2002). DNA replication in eukaryotic cells. *Annu Rev Biochem, 71*, 333-74. ↗

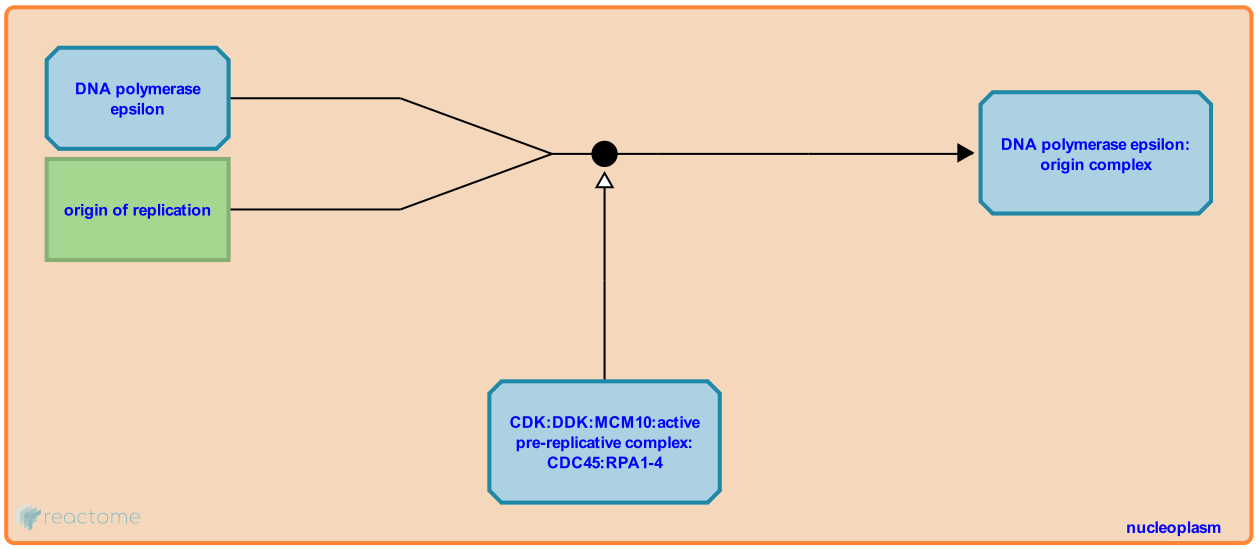
DNA polymerase epsilon binds at the origin ↗

Location: Activation of the pre-replicative complex

Stable identifier: R-HSA-68960

Type: binding

Compartments: nucleoplasm



At the beginning of this reaction, 1 molecule of 'origin of replication', and 1 molecule of 'DNA polymerase epsilon' are present. At the end of this reaction, 1 molecule of 'DNA polymerase epsilon:origin complex' is present.

Preceded by: DNA Replication Factor A (RPA) associates with the pre-replicative complex at the origin

Followed by: DNA polymerase alpha:primase binds at the origin

Literature references

Bell, SP., Dutta, A. (2002). DNA replication in eukaryotic cells. *Annu Rev Biochem, 71*, 333-74. ↗

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