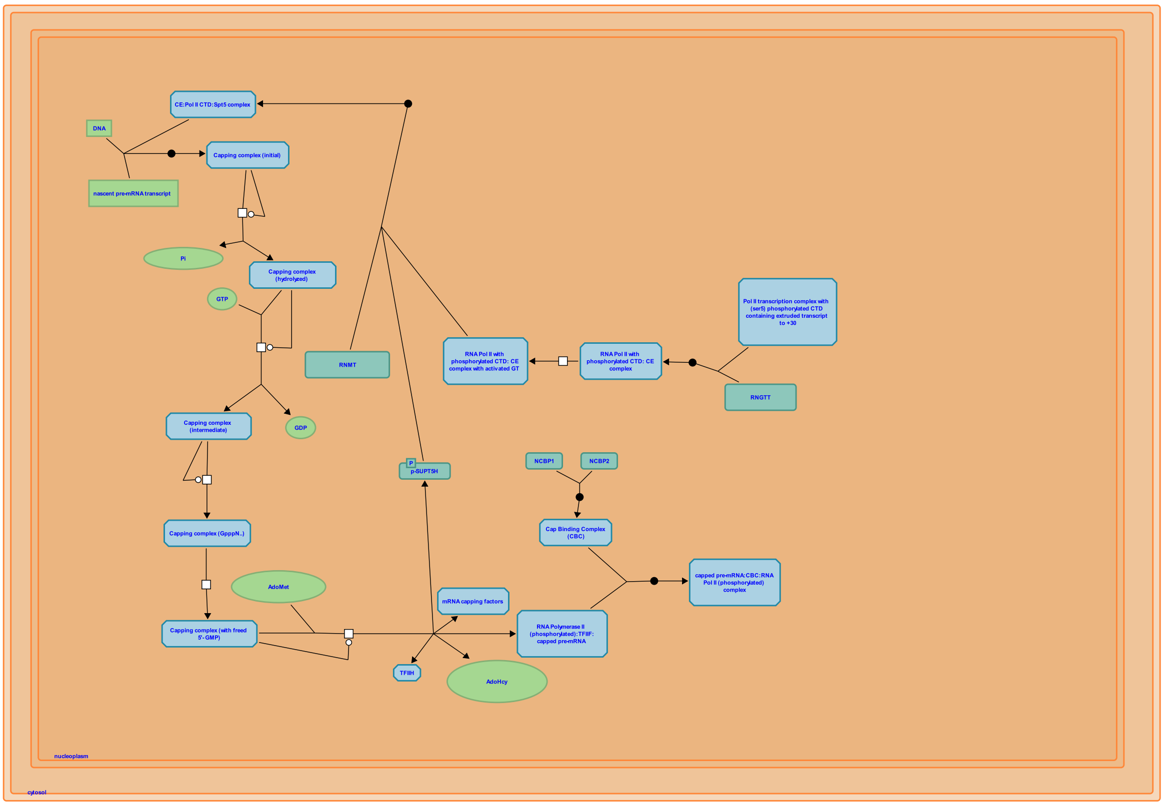
mRNA Capping



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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.

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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. ↗

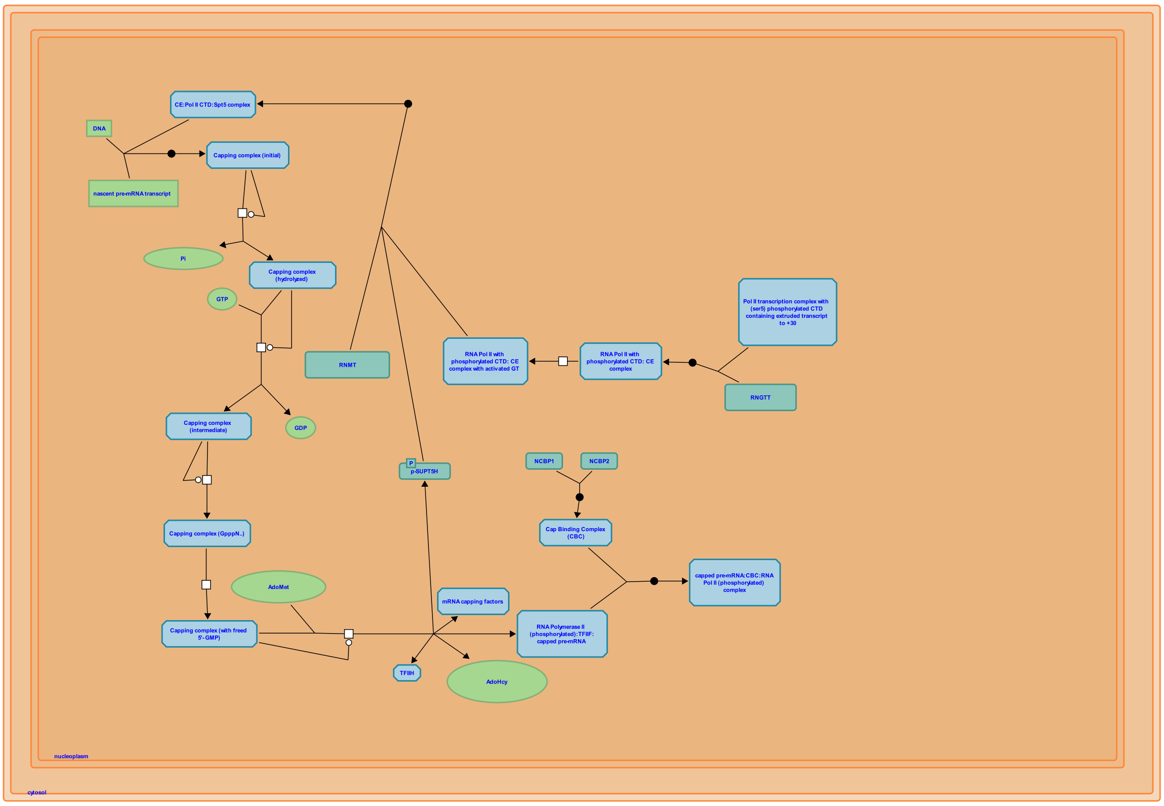
Reactome database release: 88

This document contains 1 pathway and 11 reactions (see Table of Contents)

mRNA Capping ↗

Stable identifier: R-HSA-72086

Compartments: nucleoplasm



The 5'-ends of all eukaryotic pre-mRNAs studied thus far are converted to cap structures. The cap is thought to influence splicing of the first intron, and is bound by 'cap-binding' proteins, CBP80 and CBP20, in the nucleus. The cap is important for translation initiation, and it also interacts with the poly(A)terminus, via proteins, resulting in circularization of the mRNA to facilitate multiple rounds of translation. The cap is also important for mRNA stability, protecting it from 5' to 3' nucleases, and is required for mRNA export to the cytoplasm.
The capping reaction usually occurs very rapidly on nascent transcripts; after the synthesis of only a few nucleotides by RNA polymerase II. The capping reaction involves the conversion of the 5'-end of the nascent transcript from a triphosphate to a diphosphate by a RNA 5'-triphosphatase, followed by the addition of a guanosine monophosphate by the mRNA guanylyltransferase, to form a 5'-5'-triphosphate linkage. This cap is then methylated by 2'-O-methyltransferases.

Literature references

Bentley, D. (1999). Coupling RNA polymerase II transcription with pre-mRNA processing. *Curr Opin Cell Biol, 11*, 347-51. ↗

Manley, JL., Shatkin, AJ. (2000). The ends of the affair: capping and polyadenylation. *Nat Struct Biol, 7*, 838-42. ↗

Mizumoto, K., Kaziro, Y. (1988). Messenger RNA capping enzymes from eukaryotic cells. *Prog Nucleic Acid Res Mol Biol, 34*, 1-28. ↗

Furger, A., Dye, MJ., Proudfoot, NJ. (2002). Integrating mRNA processing with transcription. *Cell, 108*, 501-12. ↗

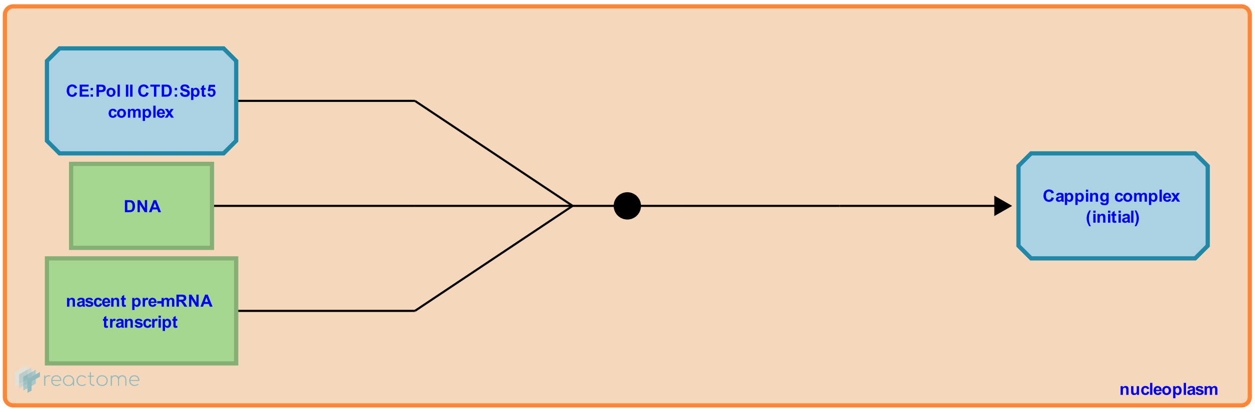
Capping complex formation ↗

Location: mRNA Capping

Stable identifier: R-HSA-77077

Type: binding

Compartments: nucleoplasm



The capping enzyme binds the 5'-end of the nascent transcript soon after it is synthesized on the DNA template, and results in the formation of the capping complex along with the C-terminal domain of RNA polymerase II, and Spt5 (Heidemann et al. 2013, Buratowski 2009, Schoenberg and Maquat 2009).

Preceded by: SPT5 subunit of Pol II binds the RNA triphosphatase (RTP)

Followed by: Hydrolysis of the 5'-end of the nascent transcript by the capping enzyme

Literature references

Voß, K., Eick, D., Heidemann, M., Hintermair, C. (2013). Dynamic phosphorylation patterns of RNA polymerase II CTD during transcription. *Biochim. Biophys. Acta, 1829*, 55-62. ↗

Maquat, LE., Schoenberg, DR. (2009). Re-capping the message. *Trends Biochem. Sci., 34*, 435-42. ↗

Buratowski, S. (2009). Progression through the RNA polymerase II CTD cycle. *Mol. Cell, 36*, 541-6. ↗

Gonatopoulos-Pournatzis, T., Cowling, VH. (2014). Cap-binding complex (CBC). *Biochem. J., 457*, 231-42. ↗

Editions

2003-10-15	Authored	Buratowski, S.
2024-03-06	Edited	Gopinathrao, G.

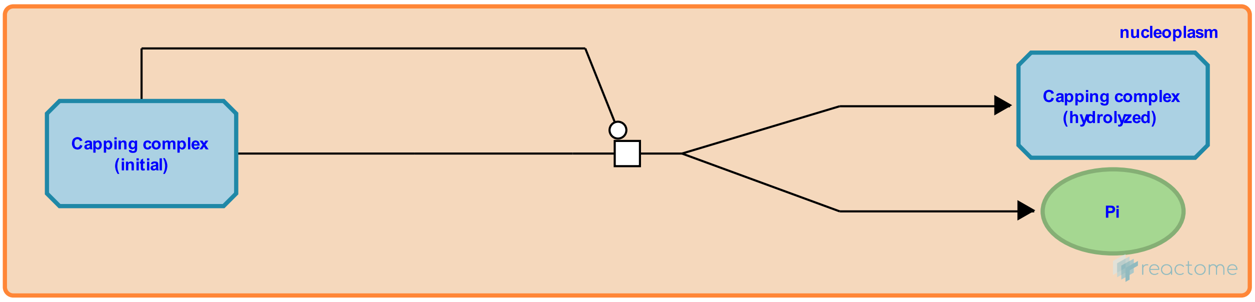
Hydrolysis of the 5'-end of the nascent transcript by the capping enzyme ↗

Location: mRNA Capping

Stable identifier: R-HSA-77078

Type: transition

Compartments: nucleoplasm



After the capping complex is formed, the RNA triphosphatase activity of the capping enzyme hydrolyzes the 5'-end phosphate group of the nascent mRNA transcript to a diphosphate.
The RNA triphosphatase (RTP) domain of mammalian capping enzyme is a member of a superfamily of phosphatases that include the protein tyrosine phosphatases, some lipid phosphatases, and several nucleic acid phosphatases. This family uses a conserved nucleophilic cysteine residue to attack the target phosphate. A transient phospho-cysteinyl enzyme intermediate is then hydrolyzed to regenerate the enzyme active site. It should be noted that while higher eukaryotic capping enzymes use PTP-like triphosphatase domains, the yeast triphosphatases are a completely different class of enzymes. The yeast RTPs are metal-dependent phosphatases. RNA 5'-triphosphatase (RTP) catalyzed first reaction can be represented as:pppN(pN)n + GTP -> ppN(pN)n + Pi; (n=20-25)

Preceded by: Capping complex formation

Followed by: Formation of the CE:GMP intermediate complex

Literature references

Yamada-Okabe, H., Shimmi, O., Arisawa, M., Yamada-Okabe, T., Doi, R. (1998). Isolation and characterization of a human cDNA for mRNA 5'-capping enzyme. *Nucleic Acids Res, 26*, 1700-6. ↗

Editions

2003-10-15	Authored	Buratowski, S.

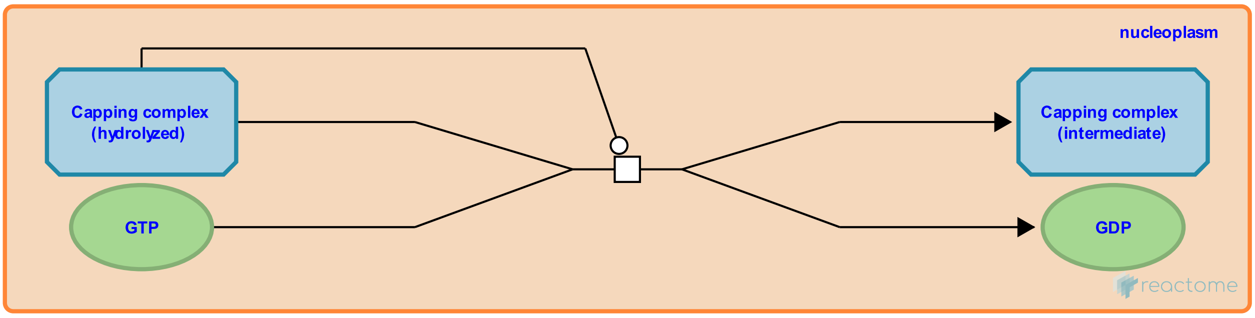
Formation of the CE:GMP intermediate complex ↗

Location: mRNA Capping

Stable identifier: R-HSA-77081

Type: transition

Compartments: nucleoplasm



A highly conserved lysine within the guanylyltransferase (GT) site of the mRNA capping enzyme attacks the alpha-phosphate of GTP. An enzyme-GMP covalent intermediate is formed.

Preceded by: Hydrolysis of the 5'-end of the nascent transcript by the capping enzyme

Followed by: Transfer of GMP from the capping enzyme GT site to 5'-end of mRNA

Literature references

Yamada-Okabe, H., Shimmi, O., Arisawa, M., Yamada-Okabe, T., Doi, R. (1998). Isolation and characterization of a human cDNA for mRNA 5'-capping enzyme. *Nucleic Acids Res, 26*, 1700-6. ↗

Editions

2003-10-15	Authored	Buratowski, S.

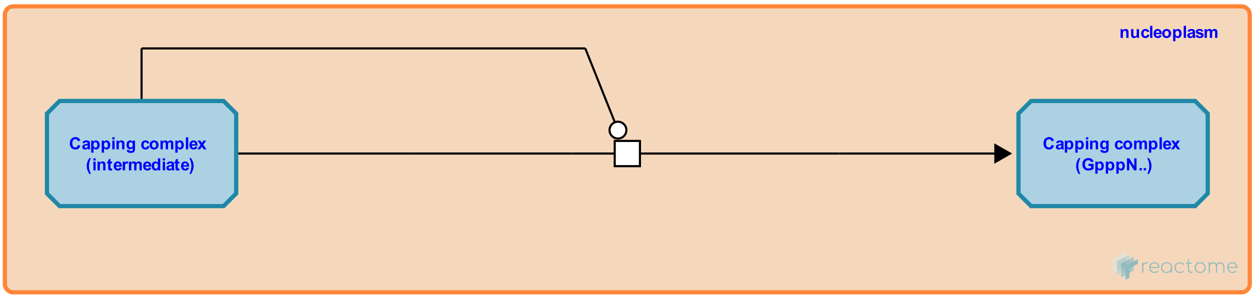
Transfer of GMP from the capping enzyme GT site to 5'-end of mRNA ↗

Location: mRNA Capping

Stable identifier: R-HSA-77083

Type: transition

Compartments: nucleoplasm



The diphosphate 5'-end of the mRNA is joined to the GMP, releasing it from the enzyme. At this time, it is unclear how the RNA diphosphate end is transferred from the active site of the triphosphatase to the guanylyltransferase site. The covalent enzyme-GMP complex can form in the absence of RNA.
Guanylyltransferase (GT) catalyzed second reaction can be represented as:ppN(pN)n + GTP -> GpppN(pN)n + PPi

(Yamada-Okabe et al. 1998).

Preceded by: Formation of the CE:GMP intermediate complex

Followed by: Dissociation of transcript with 5'-GMP from GT

Literature references

Yamada-Okabe, H., Shimmi, O., Arisawa, M., Yamada-Okabe, T., Doi, R. (1998). Isolation and characterization of a human cDNA for mRNA 5'-capping enzyme. *Nucleic Acids Res, 26*, 1700-6. ↗

Editions

2003-10-15	Authored	Buratowski, S.

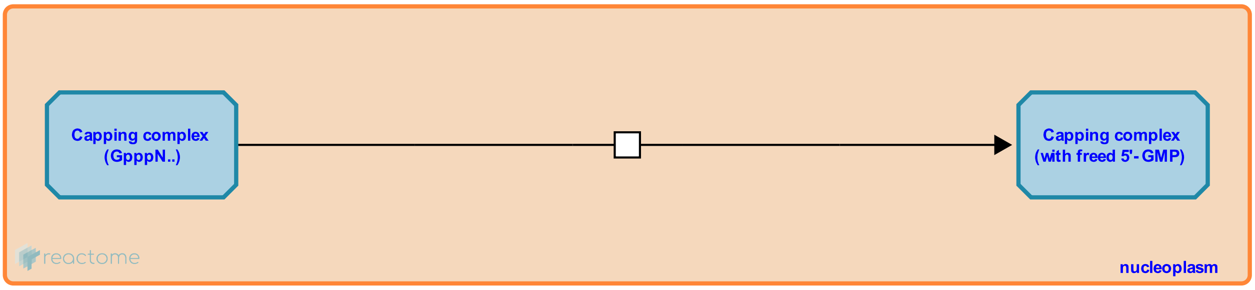
Dissociation of transcript with 5'-GMP from GT ↗

Location: mRNA Capping

Stable identifier: R-HSA-77085

Type: transition

Compartments: nucleoplasm



GMP capped mRNA transcript dissociates from GT for further modification (Yamada-Okabe et al. 1998).

Preceded by: Transfer of GMP from the capping enzyme GT site to 5'-end of mRNA

Followed by: Methylation of GMP-cap by RNA Methyltransferase

Literature references

Yamada-Okabe, H., Shimmi, O., Arisawa, M., Yamada-Okabe, T., Doi, R. (1998). Isolation and characterization of a human cDNA for mRNA 5'-capping enzyme. *Nucleic Acids Res, 26*, 1700-6. ↗

Editions

2003-10-15	Authored	Buratowski, S.

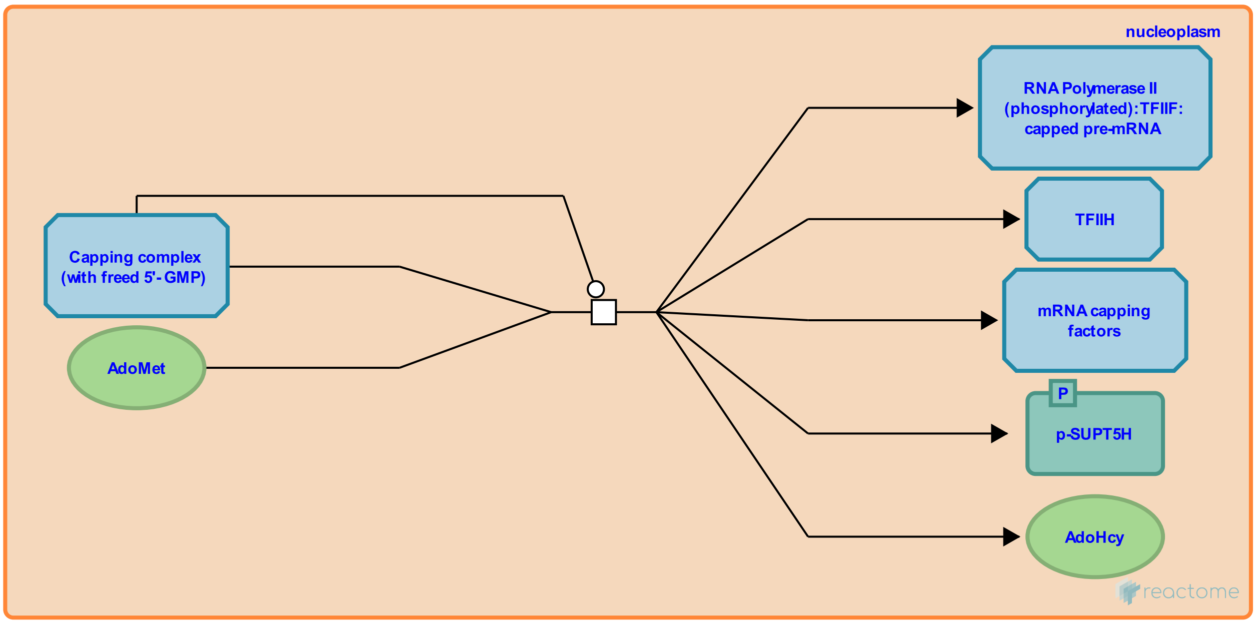
Methylation of GMP-cap by RNA Methyltransferase ↗

Location: mRNA Capping

Stable identifier: R-HSA-77090

Type: transition

Compartments: nucleoplasm



In the final step of the capping reaction, the methyltransferase takes a methyl group from S-adenosyl-methionine to the N7 position of the cap guanine. N7G-methyltransferase (MT) mediated reaction can be represented as: GpppN(pN)n + S-adenosylmethionine (Adomet) ->m7GpppN(pN)n + S-adenosylhomocysteine (Adohcy).

Preceded by: Dissociation of transcript with 5'-GMP from GT

Followed by: Recognition and binding of the mRNA cap by the cap-binding complex

Literature references

Shibagaki, Y., Niikura, Y., Tsukamoto, T., Mizumoto, K. (1998). Cloning and characterization of three human cDNAs encoding mRNA (guanine-7-)-methyltransferase, an mRNA cap methylase. *Biochem Biophys Res Commun, 251*, 27-34. ↗

Editions

2003-10-15	Authored	Buratowski, S.

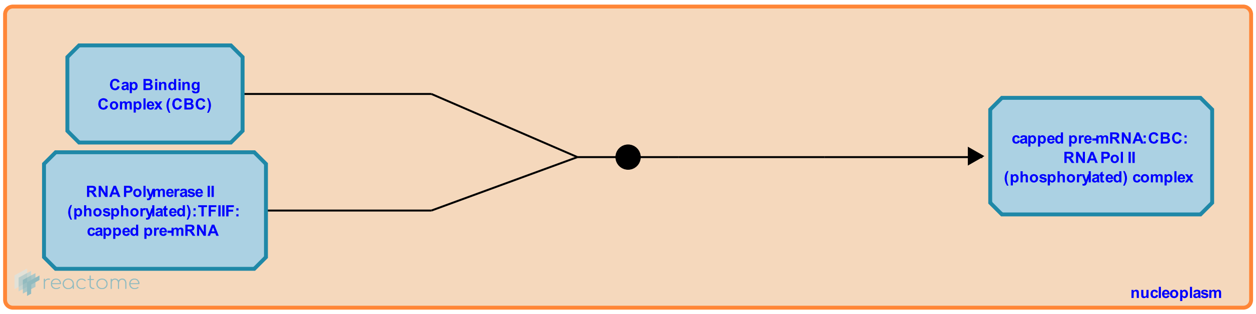
Recognition and binding of the mRNA cap by the cap-binding complex ↗

Location: mRNA Capping

Stable identifier: R-HSA-77095

Type: binding

Compartments: nucleoplasm



The cap binding complex binds to the methylated GMP cap on the nascent mRNA transcript (Gonatopoulos-Pournatzis & Cowling 2014).

Preceded by: Formation of cap binding complex (CBC), Methylation of GMP-cap by RNA Methyltransferase

Literature references

Gonatopoulos-Pournatzis, T., Cowling, VH. (2014). Cap-binding complex (CBC). *Biochem. J.*, *457*, 231-42. ↗

Editions

2003-10-15	Authored	Buratowski, S.
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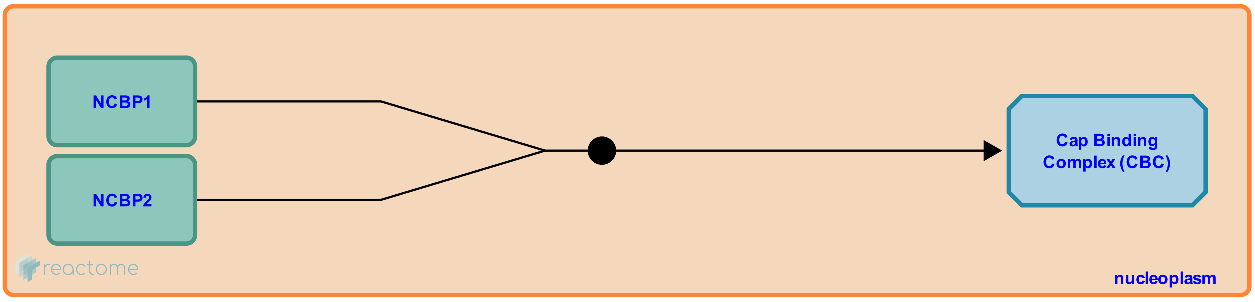
Formation of cap binding complex (CBC) ↗

Location: mRNA Capping

Stable identifier: R-HSA-77094

Type: binding

Compartments: nucleoplasm



At the beginning of this reaction, 1 molecule of 'CBP80', and 1 molecule of 'CBP20' are present. At the end of this reaction, 1 molecule of 'Cap Binding Complex (CBC)' is present (Glover-Cutter et al., 2008, Görnemann et al., 2005, Narita et al., 2007). This reaction takes place in the nucleus.

Followed by: Recognition and binding of the mRNA cap by the cap-binding complex

Literature references

Tanaka, K., Tanabe, H., Handa, H., Yung, TM., Narita, T., Yamaguchi, Y. et al. (2007). NELF interacts with CBC and participates in 3' end processing of replication-dependent histone mRNAs. *Mol. Cell, 26*, 349-65. ↗

Neugebauer, KM., Hujer, K., Kotovic, KM., Görnemann, J. (2005). Cotranscriptional spliceosome assembly occurs in a stepwise fashion and requires the cap binding complex. *Mol. Cell, 19*, 53-63. ↗

Schulze, WM., Giacometti, S., Kudla, G., Bertrand, E., Verheggen, C., Meola, N. et al. (2017). Mutually Exclusive CBC-Containing Complexes Contribute to RNA Fate. *Cell Rep, 18*, 2635-2650. ↗

Kim, S., Espinosa, J., Bentley, DL., Glover-Cutter, K. (2008). RNA polymerase II pauses and associates with pre-mRNA processing factors at both ends of genes. *Nat. Struct. Mol. Biol., 15*, 71-8. ↗

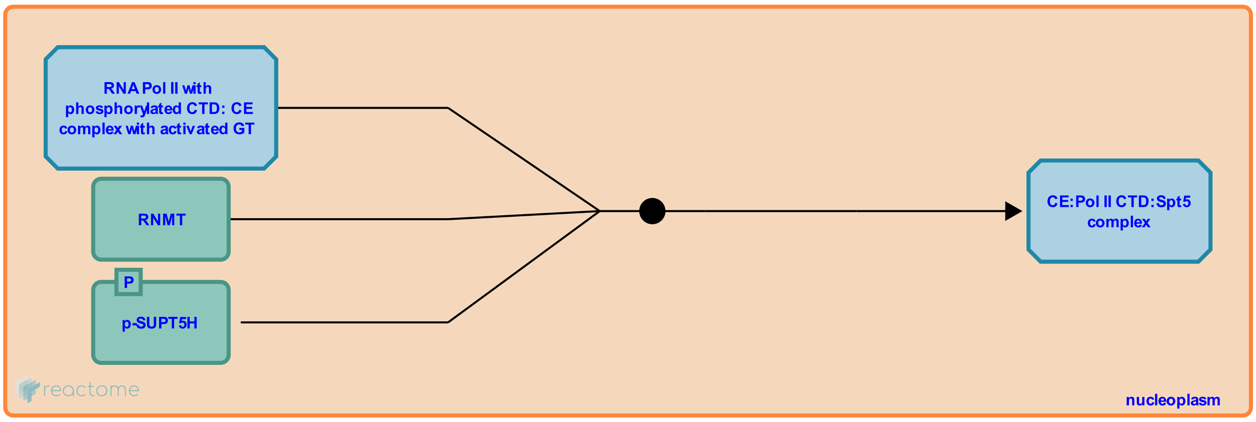
SPT5 subunit of Pol II binds the RNA triphosphatase (RTP) ↗

Location: mRNA Capping

Stable identifier: R-HSA-77073

Type: binding

Compartments: nucleoplasm



The capping enzyme interacts with the Spt5 subunit of transcription elongation factor DSIF. This interaction may couple the capping reaction with promoter escape or elongation, thereby acting as a "checkpoint" to assure that capping has occurred before the polymerase proceeds to make the rest of the transcript (Gonatopoulos-Pournatzis et al.2011).

Preceded by: Activation of GT

Followed by: Capping complex formation

Literature references

Gonatopoulos-Pournatzis, T., Cowling, VH. (2014). Cap-binding complex (CBC). *Biochem. J., 457*, 231-42. ↗

Editions

2003-10-15	Authored	Buratowski, S.

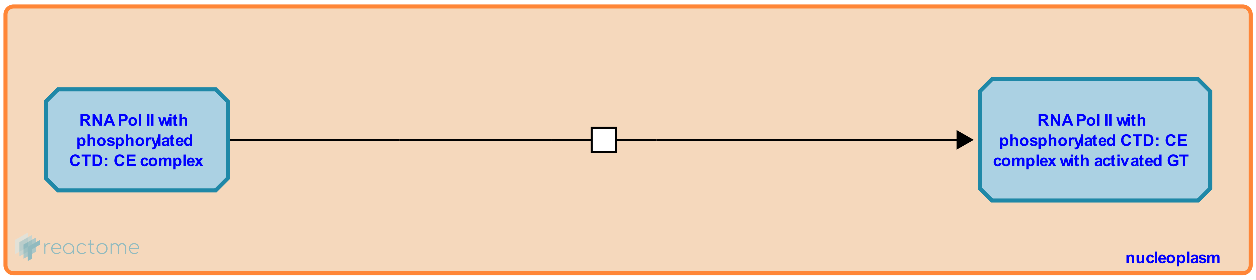
Activation of GT ↗

Location: mRNA Capping

Stable identifier: R-HSA-77068

Type: transition

Compartments: nucleoplasm



At the beginning of this reaction, 1 molecule of 'RNA Pol II with phosphorylated CTD: CE complex' is present. At the end of this reaction, 1 molecule of 'RNA Pol II with phosphorylated CTD: CE complex with activated GT' is present.

This reaction takes place in the 'nucleus'.

Preceded by: RNA Polymerase II CTD (phosphorylated) binds to CE

Followed by: SPT5 subunit of Pol II binds the RNA triphosphatase (RTP)

Literature references

Gonatopoulos-Pournatzis, T., Cowling, VH. (2014). Cap-binding complex (CBC). *Biochem. J.*, *457*, 231-42. ↗

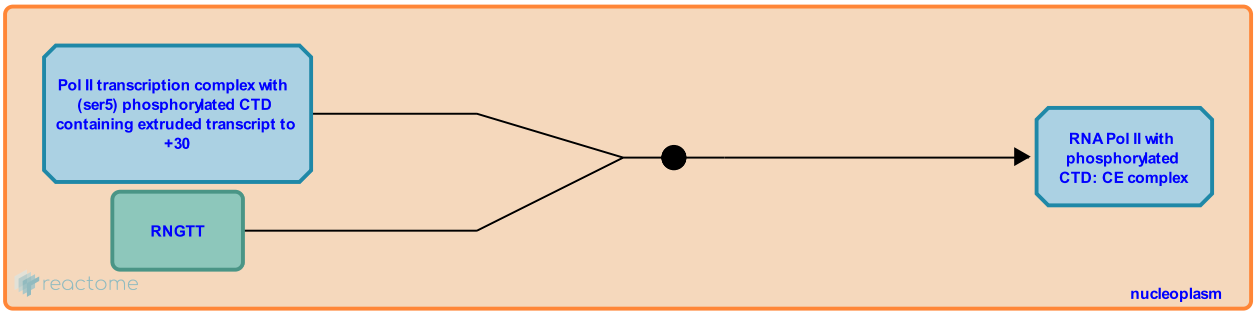
RNA Polymerase II CTD (phosphorylated) binds to CE ↗

Location: mRNA Capping

Stable identifier: R-HSA-77069

Type: binding

Compartments: nucleoplasm



At the beginning of this reaction, 1 molecule of 'mRNA capping enzyme', and 1 molecule of 'Pol II transcription complex with (ser5) phosphorylated CTD containing extruded transcript to +30' are present. At the end of this reaction, 1 molecule of 'RNA Pol II with phosphorylated CTD: CE complex' is present.

This reaction takes place in the 'nucleus'.

Followed by: Activation of GT

Literature references

Gonatopoulos-Pournatzis, T., Cowling, VH. (2014). Cap-binding complex (CBC). *Biochem. J.*, *457*, 231-42. ↗

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