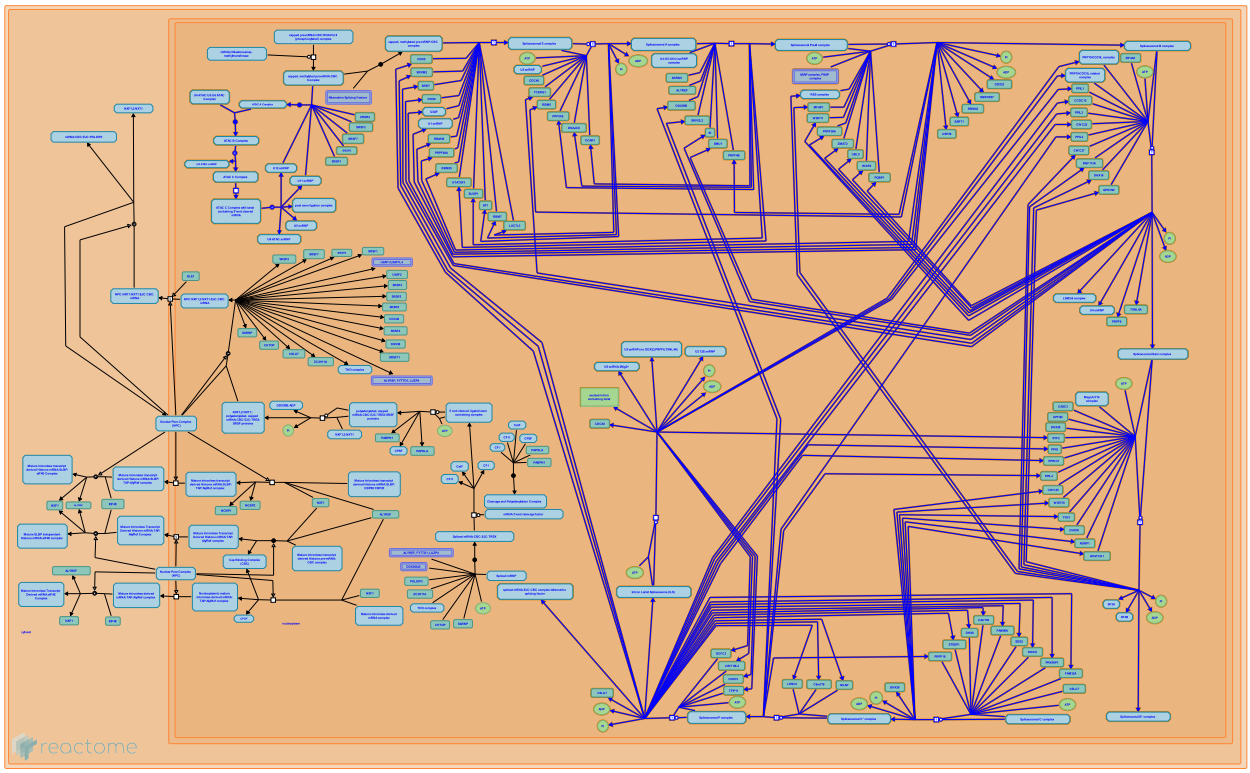
mRNA Splicing



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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.

05/11/2024

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.
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. ↗

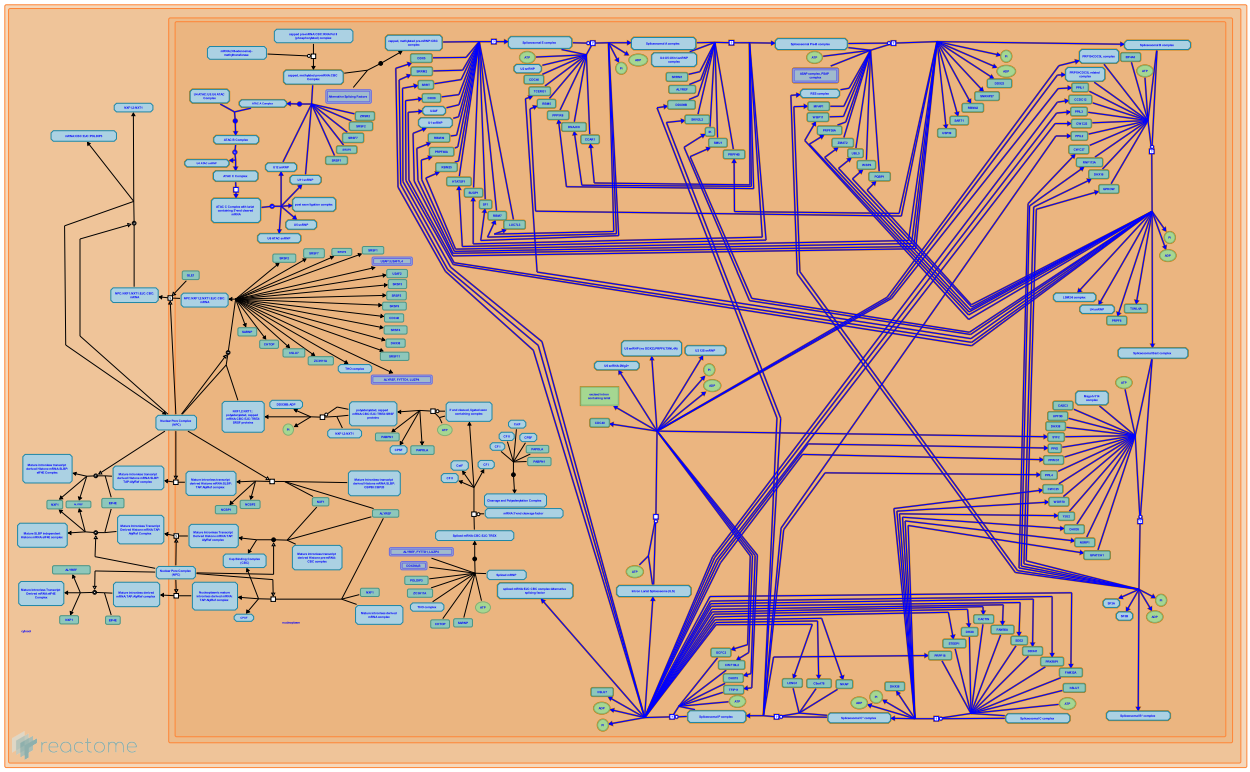
Reactome database release: 90

This document contains 3 pathways (see Table of Contents)

mRNA Splicing ↗

Stable identifier: R-HSA-72172

Compartments: nucleoplasm



The process in which excision of introns from the primary transcript of messenger RNA (mRNA) is followed by ligation of the two exon termini exposed by removal of each intron, is called mRNA splicing. Most of the mRNA is spliced by the major pathway, involving the U1, U2, U4, U5 and U6 snRNPs. A minor fraction, about 1 %, of the mRNAs are spliced via the U12 dependent pathway.

Literature references

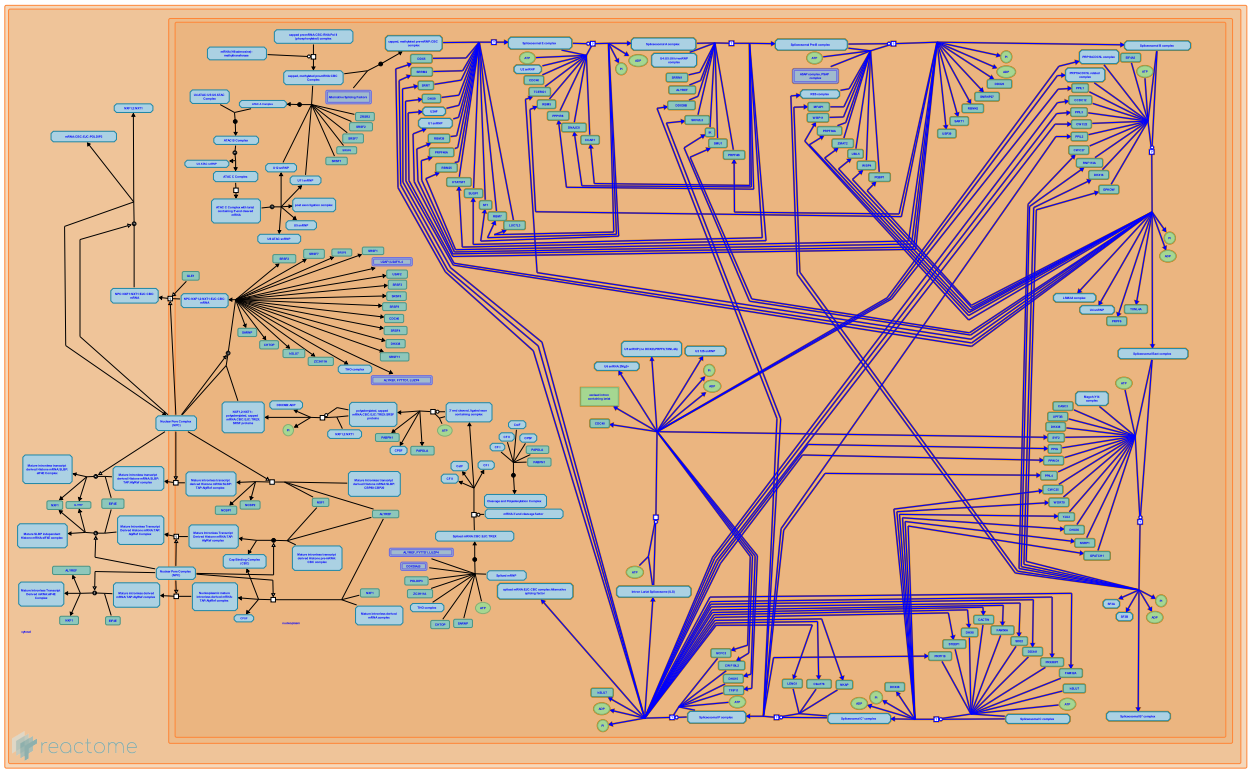
Reed, R., Gygi, SP., Licklider, LJ., Zhou, Z. (2002). Comprehensive proteomic analysis of the human spliceosome. *Nature, 419*, 182-5. ↗

mRNA Splicing - Major Pathway ↗

Location: mRNA Splicing

Stable identifier: R-HSA-72163

Compartments: nucleoplasm



Eukaryotic genes are transcribed to yield pre-mRNAs that are processed to add methyl guanosine cap structures and polyadenylate tails and to splice together segments of a pre-mRNA termed exons, thereby removing segments termed introns. More than 90% of human genes contain introns, with an average of 4.0 introns per gene and 3413 nucleotides per intron compared with 5.0 exons per gene and 50.9 nucleotides per exon (Deutsch and Long 1999). (Notable exceptions are the histone genes, which are intronless.)

Pre-mRNA splicing is performed by a large ribonucleoprotein complex, the spliceosome, which contains 5 small nuclear RNAs (snRNAs) and more than 150 proteins (reviewed in Will and Luhrmann 2011, Kastner et al. 2019, Yan et al. 2019, Fica et al. 2020, Wan et al. 2020, Wilkinson et al. 2020). The catalyst in the spliceosome comprises magnesium ions coordinated by the U6 snRNA that catalyze transesterification reactions between hydroxyl groups and phosphate groups from the pre-mRNA. The role of the U6 snRNA demonstrates that the spliceosome is a ribozyme hints at the origin of the spliceosome as a self-splicing group II intron.

The spliceosome is initially assembled cotranscriptionally on the pre-mRNA as the Spliceosomal E (Early) complex and then remodelled sequentially by association and dissociation of proteins and snRNAs to catalyze of the two reactions of splicing. First, a nucleophilic attack by the 2' hydroxyl group of a conserved adenine residue, the branch point, within the intron on the phosphate group of the 5' residue of the intron yields a lariat (looped) structure in the intron joined to the downstream (3') exon and a free upstream exon with a 3' hydroxyl group. Second, a nucleophilic attack by the 3' hydroxyl group of the upstream exon on the phosphate of the 5' residue of the downstream exon yields a spliced mRNA containing the upstream exon ligated to the downstream exon and a free intron containing a lariat structure.

The Spliceosomal E complex contains the U1 snRNP bound to the 5' splice site, SF1 bound to the branch point, and the U2AF complex bound to the polypyrimidine tract of the intron and the 3' splice site of the pre-mRNA (Zhuang and Weiner 1986, Hong et al. 1997, Das et al. 2000, Hartmuth et al. 2002, Rappsilber et al. 2002, Hegele et al. 2012, Makarov et al. 2012, Crisci et al. 2015, Kondo et al. 2015, Tan et al. 2016). SF1 and U2AF are displaced on the pre-mRNA and the U2 snRNP binds the branch region to yield the Spliceosomal A complex (Wu and Manley 1989, Fleckner et al. 1997, Neubauer et al. 1998, Hartmuth et al. 2002, Rappsilber et al. 2002, Xu et al. 2004, Behzadnia et al. 2007, Shen et al. 2008, Chen et al. 2017, Zhang et al. 2020). The U4/U6.U5 tri-snRNP, containing the U4 snRNA base-paired with the U6 snRNA plus the U5 snRNP and accessory proteins, binds the Spliceosomal A complex to form the Spliceosomal Pre-B complex (Hausner et al. 1990, Kataoka and Dreyfuss 2004, Chi et al. 2013, Mohlmann et al. 2014, Boesler et al. 2016, Zhan et al. 2018, Charenton et al. 2019, Kastner et al. 2019, Townsend et al. 2020). The U1 snRNP is replaced at the 5' splice site by the U6 snRNA and the spliceosome is remodelled to yield the Spliceosomal B complex (Ismaïli et al. 2001, Deckert et al. 2006, Bessonov et al. 2008, Wolf et al. 2009, Bessonov

et al. 2010, Schmidt et al. 2014, Boesler et al. 2016, Bertram et al. 2017, Zhang et al. 2018, Kastner et al. 2019). The Spliceosomal B complex is activated to form the Spliceosomal Bact complex by dissociation of the U4 snRNP and Lsm proteins from the U6 snRNA, freeing the U6 snRNA to form the active site of the spliceosome (Lamond et al. 1988, Laggerbauer et al. 1998, Ajuh et al. 2000, Bessonov et al. 2010, Agafonov et al. 2011, Haselbach et al. 2018, Zhang et al. 2018, Kastner et al. 2019, Busetto et al. 2020). Dissociation of the SF3A and SF3B subcomplexes of the U2 snRNP allows the intron branch point to dock near the 5' splice site, forming the B* Spliceosomal complex. Reaction of the branch point at the 5' splice site, yields the Spliceosomal C complex (Jurica et al. 2002, Makarov et al. 2002, Rappsilber et al. 2002, Reichert et al. 2002, Kataoka and Dreyfuss 2004, Bessonov et al. 2010, Gencheva et al. 2010, Agafonov et al. 2011, Alexandrov et al. 2012, Barbosa et al. 2012, Steckelberg et al. 2012, Schmidt et al. 2014, Zhan et al. 2018, Kastner et al. 2019, Busetto et al. 2020). The branch point is rotated to allow the 3' splice site to enter the active site, yielding the Spliceosomal C* complex (Ortlepp et al. 1998, Zhou and Reed 1998, Jurica et al. 2002, Makarov et al. 2002, Rappsilber et al. 2002, Ilagan et al. 2013, Bertram et al. 2017, Zhang et al. 2017, Kastner et al. 2019). Reaction of the 3' hydroxyl group of the upstream exon at the 3' splice site yields the Spliceosomal P (postcatalytic) complex (Zhou et al. 2000, Kataoka and Dreyfuss 2004, Tange et al. 2005, Zhang and Krainer 2007, Chi et al. 2013, Ilagan et al. 2013, Bertram et al. 2017, Zhang et al. 2017, Fica et al. 2019, Zhang et al. 2019). The Spliceosomal P complex then dissociates to yield an mRNP containing the spliced mRNA and associated proteins, including the exon junction complex (EJC) (Ohno and Shimura 1996, Merz et al. 2007, Yoshimoto et al. 2009, Zanini et al. 2017, Felisberto-Rodrigues et al. 2019, Zhang et al. 2019, EJC reviewed in Schlautmann and Gehring 2020), and the Intron Lariat Spliceosome (ILS), which contains the intron lariat. The ILS is then disassembled to free its components for further splicing reactions and the intron lariat is degraded (Wen et al. 2008, Yoshimoto et al. 2009, Yoshimoto et al. 2014, Zhang et al. 2019, Studer et al. 2020).

Literature references

Reed, R., Luo, MJ., Katahira, J., Hurt, E., Straesser, K., Zhou, Z. (2000). The protein Aly links pre-messenger-RNA splicing to nuclear export in metazoans. *Nature, 407*, 401-5.

Sander, B., Will, CL., Lührmann, R., Anokhina, M., Golas, MM., Krasauskas, A. et al. (2010). Characterization of purified human Bact spliceosomal complexes reveals compositional and morphological changes during spliceosome activation and first step catalysis. *RNA, 16*, 2384-403.

Reed, R., Das, R., Zhou, Z. (2000). Functional association of U2 snRNP with the ATP-independent spliceosomal complex E. *Mol Cell, 5*, 779-87.

Makarova, OV., Will, CL., Gentzel, M., Lührmann, R., Wilm, M., Makarov, EM. et al. (2002). Small nuclear ribonucleoprotein remodeling during catalytic activation of the spliceosome. *Science, 298*, 2205-8.

Komarov, I., Lührmann, R., Graf, B., Dybkov, O., Agafonov, DE., Haselbach, D. et al. (2018). Structure and Conformational Dynamics of the Human Spliceosomal B^{act} Complex. *Cell, 172*, 454-464.e11.

Editions

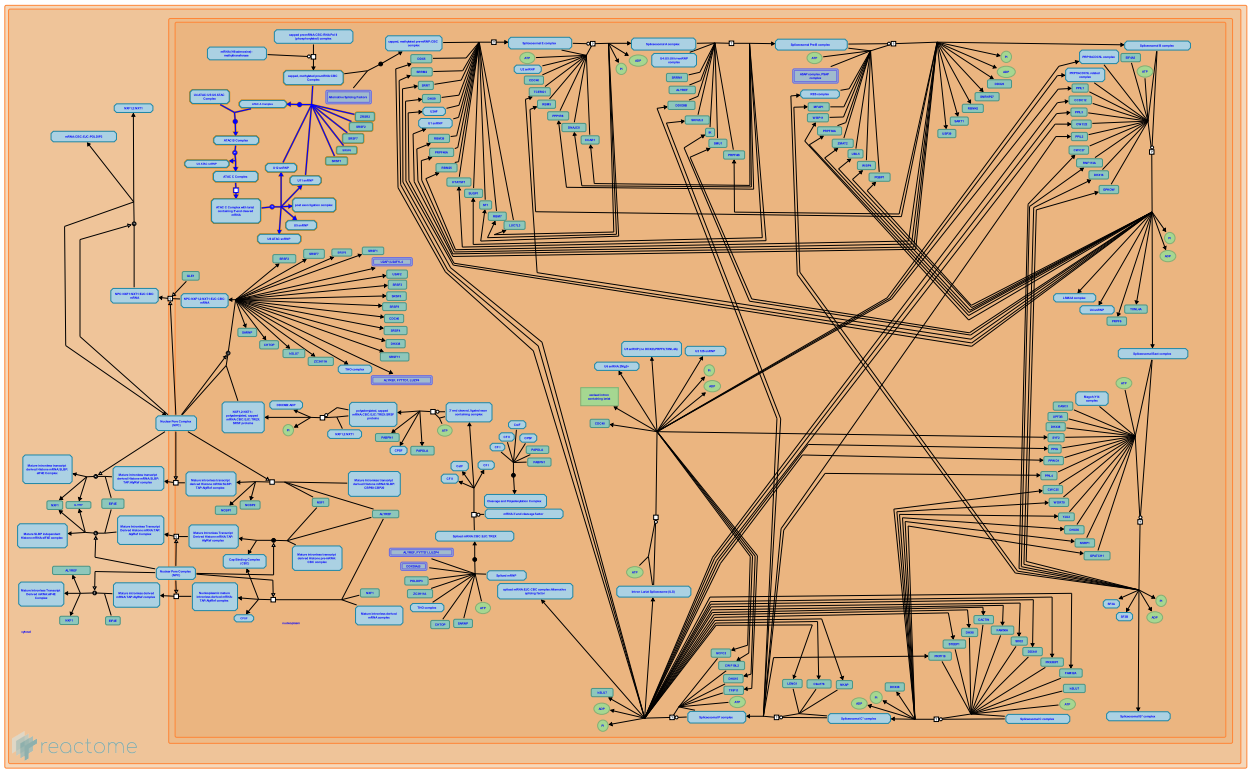
2003-06-05	Authored	Krainer, AR.
2003-08-22	Authored	Joshi-Tope, G.
2022-03-22	Authored, Edited, Revised	May, B.
2022-10-20	Reviewed	Charenton, C.

mRNA Splicing - Minor Pathway ↗

Location: mRNA Splicing

Stable identifier: R-HSA-72165

Compartments: nucleoplasm



The splicing of a subset of pre-mRNA introns occurs by a second pathway, designated the AT-AC or U12-dependent splicing pathway. AT-AC introns have highly conserved, non-canonical splice sites that are removed by the AT-AC spliceosome, which contains distinct snRNAs (U11, U12, U4atac, U6atac) that are structurally and functionally analogous to the major spliceosome. U5 snRNA as well as many of the protein factors appear to be conserved between the two spliceosomes.

Literature references

Krainer, AR., Wu, Q. (1999). AT-AC pre-mRNA splicing mechanisms and conservation of minor introns in voltage-gated ion channel genes. *Mol Cell Biol, 19*, 3225-36. ↗

Steitz, JA., Tarn, WY. (1997). Pre-mRNA splicing: the discovery of a new spliceosome doubles the challenge. *Trends Biochem Sci, 22*, 132-7. ↗

Editions

2024-08-27	Edited	Joshi-Tope, G.

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