Insertion of Precursor Polypeptides into the ER Membrane

Subjects: Biochemistry & Molecular Biology | Cell Biology

Contributor: Martin Jung , Richard Zimmermann

The protein import into the organelle termed the endoplasmic reticulum (ER) is the first step in the biogenesis of about one-third of the different soluble and membrane proteins (MPs) of human cells and, therefore, represents a central cell biological research topic.

gene expression | protein biogenesis | membrane proteins | endoplasmic reticulum

1. Introduction

Nucleated human cells are separated from the environment by the so-called plasma membrane and contain different subcellular compartments, called cell organelles (**Figure 1**). These organelles are surrounded and, thereby, separated from the aqueous, albeit gel-like, cytosol by at least one biological membrane and have to be distributed to daughter cells from the mother cell during cell division (with the exception of lipid droplets and peroxisomes). In the cytosol, the vast majority of the approximately 24,000 different polypeptides of human cells are synthesized by 80S ribosomes. Therefore, the distinct proteins of the various organelles and the plasma membrane have to, first, be targeted to the specific organelles and, subsequently, inserted into or translocated across the membrane(s) of the relevant organelles. The protein import into the organelle termed the endoplasmic reticulum (ER) is the first step in the biogenesis of about one-third of the different soluble and membrane proteins (MPs) of human cells and, therefore, represents a central cell biological research topic of the past fifty years as well as several years to come. In a second step, the non-ER proteins reach their functional location in either the extracellular space; one of the endocytotic or exocytotic organelles (ERGIC, Golgi apparatus, endosome, lysosome, or trafficking vesicles); the plasma, peroxisomal, and mitochondrial membrane or in lipid droplets by vesicular transport; direct budding of new organelles (peroxisomal precursors or lipid droplets); or the ER–SURF pathway (mitochondria) [1][2][3][4][5][6][7][8][9][10]. The first insights into ER protein import were gained about seventy years ago. From electron microscopic images, Palade and Potter concluded that the ER represents a ‘continuous, tridimensional reticulum’ and that ‘the surface appears to be dotted with small, dense granules that cover them in part or in entirety’, i.e., cytosolic 80S ribosomes [11][12].

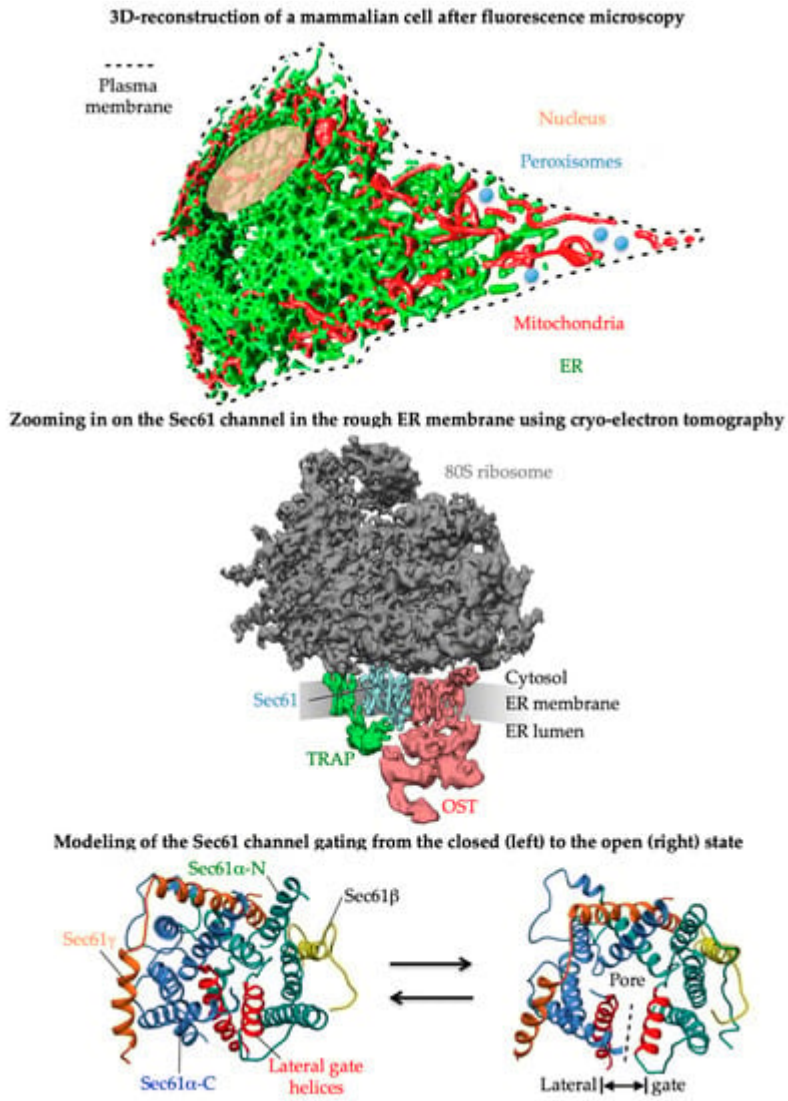

Figure 1. 3D reconstructions of a typical nucleated human cell and a ribosome-bound Sec61 translocon. The figure and its legend were adapted from Lang et al. and Sicking et al. [5][6]. The upper part shows an artist's view of a 3D reconstruction after live cell fluorescence imaging with ER-resident GFP and mitochondrial RFP and the central part a 3D reconstruction of the native ribosome-translocon complex in the human ER membrane after cryoelectron tomography. In human cells, the heterotrimeric Sec61 complex together with the ribosome form various large multicomponent complexes, e.g., the most abundant one comprising the multimeric membrane proteins translocon-associated protein (TRAP) and oligosaccharyltransferase (OSTA), which catalyzes N-linked glycosylation. This super-complex, now termed OSTA translocon, can insert into the membrane or translocate into the lumen a whole variety of topologically very different precursor polypeptides, i.e., type I-, type II-, glycosylphosphatidylinositol-, or GPI-anchored and type I multispanning membrane proteins, as well as soluble proteins, respectively (**Figure 2**). Membrane insertion and translocation are facilitated by either a cleavable amino-terminal SP or the TMH of the nascent precursor polypeptide, which acts as a non-cleavable SP substitute (**Figure 2**). The lowest part represents a modeling of reversible gating of the heterotrimeric Sec61 channel by SPs or TMHs. The fully open state of the Sec61 channel allows the translocation of hydrophilic domains of MPs or entire precursor polypeptides from the cytosol into the ER lumen (via the aqueous channel pore) and the insertion of transmembrane domains into the ER membrane (via the lateral gate), respectively.

The latter observation paved the way for the ‘signal hypothesis’ by G. Blobel and colleagues [1][2][13][14], which suggested that topogenic sequences in nascent precursor polypeptides guide the translating ribosomes to the ER membrane and that the subsequent membrane insertion or translocation occurs coupled to translation, i.e., cotranslationally (**Figure 1**). For membrane proteins, the beauty of this concept of cotranslational ER protein import is that their eventual hydrophobic transmembrane domain or domains do not face the problem of aggregation in the cytosol. Subsequent work in human cell-free systems and in the yeast *Saccharomyces cerevisiae* uncovered that the topogenic sequence, termed the amino-terminal signal peptide (SP) or SP-equivalent transmembrane helix (TMH), of a nascent precursor polypeptide is recognized and bound by the cytosolic signal recognition particle (SRP), which facilitates the association of the complex between the ribosome, nascent chain, and SRP with the heterodimeric SRP receptor in the ER membrane, termed the SR (**Figure 2** and **Figure 3**) [2][15][16][17][18][19][20][21][22][23][24][25][26][27][28]. Thus, the combined action of SRP plus SR represents an ER targeting pathway for nascent precursors of soluble and membrane proteins, as well as the corresponding mRNAs. Recently, proximity-based ribosome-profiling experiments confirmed the preference of SRP and SR for SPs and relatively amino-terminal TMHs of the nascent precursor polypeptide chains [29][30]. Typically, the interaction of SRP with SR leads to the cotranslational transfer of the ribosome-nascent chain complex (RNC) to the central component for both protein translocation and membrane insertion in the ER membrane, the translocon, or the heterotrimeric Sec61 complex (**Figure 1**) [31][32][33][34][35][36][37][38][39][40][41]. SPs and TMHs of nascent precursors may spontaneously interact with and trigger the opening of the Sec61 channel; i.e., both the central aqueous channel, as well as the lateral gate (**Figure 1**), or the productive Sec61 interaction may be facilitated by one of the auxiliary components of the ER membrane, i.e., the heterotetrameric translocon-associated protein (TRAP) [42][43][44][45][46][47][48], the heterodimeric Sec62/Sec63 complex with or without the help of the ER lumenal chaperone BiP [49][50][51][52][53][54][55][56][57][58][59][60][61][62], and the translocating chain-associated membrane protein 1 (TRAM1) (**Figure 1** and **Figure 3**) [63][64][65][66][67]. Notably, however, there is a human paralog of the α-subunit of the Sec61 complex, termed Sec61α2, i.e., a putative alternative Sec61 complex that is more or less uncharacterized but recently addressed with respect to its substrates or client spectrum in yeast [68], and there exist alternative components for the targeting (SND, TRC/GET, and PEX3) [69][70][71][72][73][74][75][76][77][78][79][80][81][82][83][84][85][86][87][88][89][90][91][92][93][94][95][96][97][98][99][100][101][102][103][104][105][106][107][108][109][110][111][112][113][114][115][116][117][118][119][120], as well as membrane insertion of precursors (EMC and the GEL-BOS-PAT complex) [121][122][123][124][125][126][127][128][129][130][131][132].

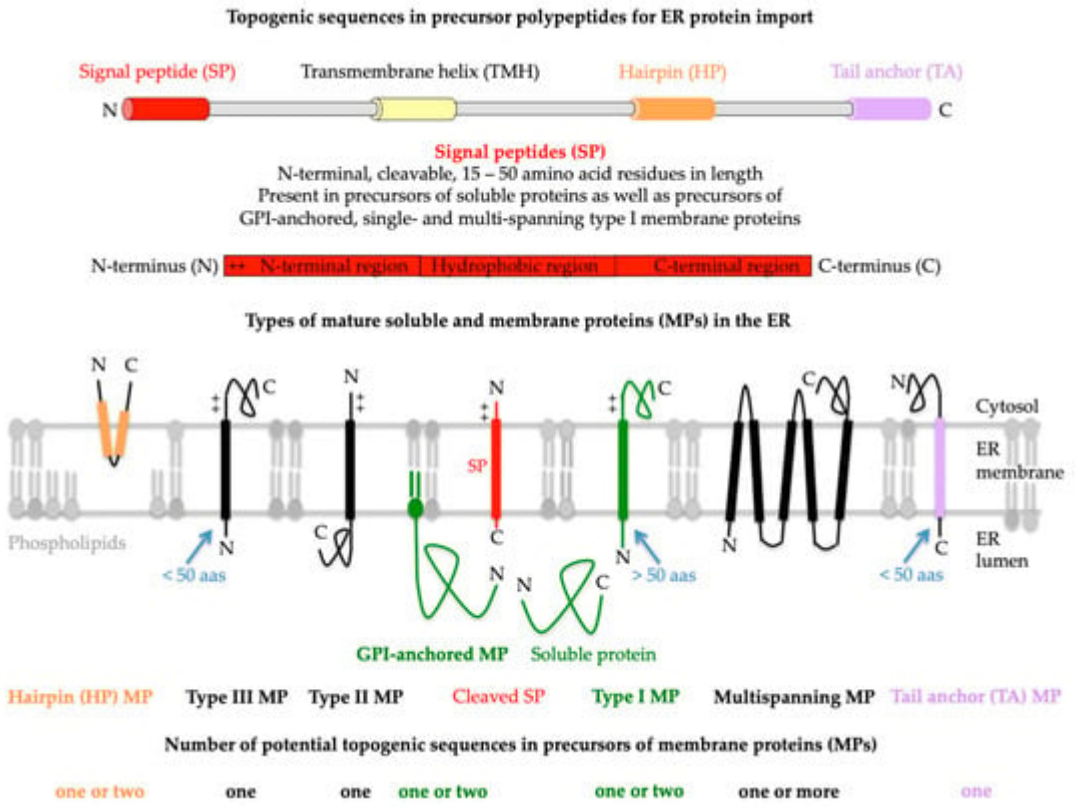

Figure 2. Topogenic sequences in precursors of soluble and membrane proteins for import into the human endoplasmic reticulum (ER). The cartoons depict signal peptides (SPs, shown in red) and seven classes of ER membrane proteins (MPs), plus their membrane protein types (in bold face) and potential topogenic sequences. Cleavable SPs have a tripartite structure (including a hydrophobic or H-region with 7–9 amino acid residues, or aas for short) and facilitate ER import of soluble proteins, GPI-anchored MPs, and single-spanning type I MPs (all shown in green). In addition, they mediate ER import of some multispanning MPs, but not of hairpin, single-spanning type II or III, TA, and other multispanning MPs (which can also be of type II or III; by definition, the shown multispanning MP is of type III). All the latter MPs depend on more or less amino-terminal transmembrane helices (TMHs, with a length of 15–25 amino acid residues) that serve as SP equivalents and facilitate membrane targeting, as well as membrane insertion. Notably, precursors of soluble proteins with SP and of type II or III and TA MPs contain only one topogenic sequence, whereas, in all other precursors, other TMDs than the TMHs may serve as alternative or additional topogenic sequences. Positively charged amino acid residues (+) play an important role in the orientation of SPs and MPs in the ER membrane, where the orientation follows the positive inside rule. Cleavable SPs are removed from the precursor polypeptides in transit by one of the two signal peptidase complexes (SPCs), which have their catalytic sites in the ER lumen. Following ER import and simultaneous cleavage of the SP, as well as the C-terminal GPI-attachment sequence, GPI-MPs become membrane-anchored via carboxy-terminal GPI attachment. Notably, HP proteins are also termed monotopic MPs, and type II and type III MPs are also referred to as MPs with N_{in} or N_{cyt} and N_{out} and N_{exo}, respectively, signal anchors [1][69][70]. The figure and its legend were adapted from Lang et al. and Sicking et al. [5][6]. C, carboxy-terminus; GPI, glycosylphosphatidylinositol; N, amino-terminus.

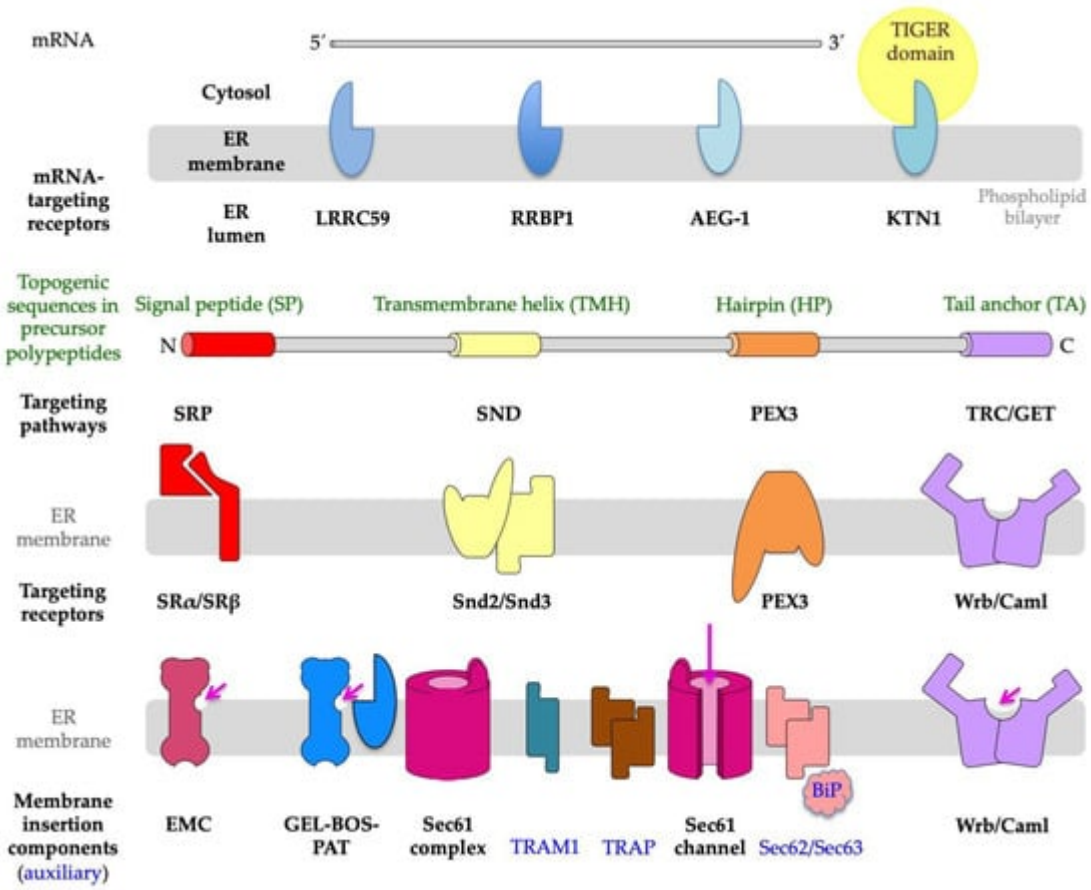

Figure 3. Components that are involved in protein import into the endoplasmic reticulum (ER) of human cells. ER protein import comprises a targeting step, plus a translocation or membrane insertion step, and may involve targeting of receptors for mRNAs or ribosomes with or without short nascent polypeptide chains to the ER (LRRC59, RRBP1, AEG-1, or KTN1). Alternatively, nascent or fully synthesized precursor polypeptides are targeted to the ER, depending on their topogenic sequences and targeting pathways, which involve cytosolic components, as well as their corresponding heterodimeric receptors in the ER membrane, such as SRα/SRβ, Snd2/Snd3, PEX3/PEX16, and Wrb/Caml. The membrane translocation is mediated by the heterotrimeric and polypeptide-conducting Sec61 channel, which may be supported by either the TRAP complex or the Sec62/Sec63 complex. Membrane insertion is mediated by several membrane protein insertases, i.e., (i) the Sec61 channel; (ii) the Sec61 channel with its partner complexes GEL, BOS, and PAT; (iii) the multimeric EMC; or (iv) the heterodimeric Wrb/Caml complex. The long arrow (in magenta) points to the open aqueous channel and open lateral gate, respectively, of the fully open Sec61 channel; the short arrows (in magenta) point to the characteristic hydrophilic vestibules of the MP insertases. Notably, (i) according to structural prediction tools, the yeast and human Snd2 may also be able to form a hydrophilic vestibule near the cytosolic surface of the ER membrane and, therefore, may also be able to facilitate membrane insertion [74][106]; (ii) the TIGER domain represents a cytosolic micro-domain, enriched in MP-encoding mRNAs with multiple AU-rich elements or AREs in their 3′UTRs in the vicinity of the ER [109][110]; and (iii) PEX3 is present in an ER subdomain which may be identical to the pre-peroxisomal ER [71][72][73]. The figure was adapted from Tirincsi et al. [106].

2. Proteins of the ER Membrane

The SPs, mentioned above, have a tripartite structure (including an amino-terminal or N-region, a hydrophobic or H-region, and a carboxy-terminal or C-region) and facilitate the ER targeting of soluble proteins, GPI-anchored MPs, and single- as well as multispanning type I MPs (**Figure 2**) [15][16][17][69][70]. In contrast, all the other MPs of the ER membrane (hairpin or HP proteins, and single- and multispanning type II or type III MPs, as well as TA proteins) depend on more or less amino-terminal or even carboxy-terminal TMHs (with a typical length of 15–25 amino acid residues) that serve as SP equivalents and facilitate membrane targeting as well as membrane insertion [15][16][17]. HP- and GPI-anchored proteins are special [71][72][73][74][75]: Following ER import and the simultaneous cleavage of the SP, as well as the carboxy-terminal GPI-attachment sequence, GPI-anchored MPs become membrane-anchored via carboxy-terminal GPI attachment. HP proteins do not contain a real transmembrane domain (TMD); instead, they comprise one or more hydrophobic stretches of amino acid residues, which are typically defined by databases as TMDs and, therefore, discussed here as such. By definition, precursors of soluble proteins with SP and of single-spanning type II or III and TA MPs contain only one topogenic sequence, whereas, in all other precursors, other TMDs (i.e., other than the most amino-terminal TMHs) may serve as alternative or additional topogenic sequences. Typically, SPs are removed from the precursor polypeptides in transit by signal peptidases with ER lumenal catalytic sites [76][77][78]. Furthermore, many of the polypeptides of the secretory pathway become N-glycosylated by one of the two oligosaccharyltransferases (OST), which also have their catalytic sites in the ER lumen but act either on precursors in transit (i.e., cotranslationally, OSTA, **Figure 1**) or posttranslationally (OSTB) [79][80][81].

3. Targeting of Precursor Polypeptides to the ER Membrane

The characterization of precursors capable of SRP-independent ER targeting, such as small presecretory proteins and TA proteins, first suggested the existence of SRP-independent ER targeting pathways to the ER [82][83][84][85][86][87]. In contrast to the SRP/SR pathway, these alternative targeting pathways can direct precursor polypeptides to the Sec61 complex co- as well as posttranslationally, and are named the TRC or GET, PEX19/PEX3, and Snd2/Snd3 pathway (**Figure 3**) [58][71][73][74][75][87][88][89][90][91][92][93][94][95][96][97][98][99][100][101][102][103][104][105][106]. Notably, there is also SRP-independent targeting of all sorts of mRNAs to the ER surface (**Figure 3**) [29][107][108][109][110]. Typically, this mRNA targeting to the ER depends on receptors for mRNAs (such as KTN1), or RNCs with nascent polypeptide chains that are not yet long enough to allow the interaction of the topogenic sequence with SRP (such as RRBP1, LRRC59, and AEG-1) (**Figure 3**) [29][111][112][113]. In the case of the ER targeting of mRNAs that code for cytosolic proteins, the nascent polypeptide-associated complex (NAC) can bind to the amino-terminus of the nascent polypeptides and trigger their release from the Sec61 complex [23][114][115][116].

4. Insertion of Precursor Polypeptides into the ER Membrane

As stated above, the insertion of precursors with SP or TMH into the Sec61 channel and the concomitant gating of the Sec61 channel to the open conformation occur spontaneously or involve client-specific auxiliary components of

the Sec61 channel (TRAP, Sec62/Sec63 complex) (**Figure 1** and **Figure 3**). Typically, the orientation of SP- and TMH in the Sec61 channel follows the positive inside rule; i.e., positively charged amino acid residues in the N-region support loop insertion (N_{in}-C_{out}) and positively charged residues downstream of the SP or TMH interfere with loop insertion and, therefore, favor head-on insertion (N_{out}-C_{in}) into the Sec61 channel (**Figure 2**) [117][118][119][120]. Next, TMDs can enter the phospholipid bilayer via the lateral gate of the fully open Sec61 channel by lateral movement and large hydrophilic domains (i.e., with more than 50 amino acid residues), or entire soluble proteins can be translocated into the ER lumen by vectorial movement through the channel (**Figure 1** and **Figure 2**). Alternatively, membrane insertion of some precursors of MPs can be facilitated co- or posttranslationally by evolutionarily conserved MP insertases that comprise a hydrophilic vestibule near the cytosolic surface of the ER membrane, but cannot form an aqueous channel through the ER membrane, and, therefore, cannot translocate hydrophilic domains with more than 50 amino acid residues across the ER membrane. These Oxa1-related insertases are the Wrb/Caml complex (in co-operation with cytosolic GET3 of the abovementioned TRC pathway), or the multimeric ER membrane protein complex (EMC, which may occasionally co-operate with the Sec61 complex), or the GEL-BOS-PAT complex, which depends on the interaction with but not the channel activity of the Sec61 complex (**Figure 3**) [121][122][123][124][125][126][127][128][129][130][131][132]. Notably, the first has its exclusive role in the membrane insertion of TA membrane proteins [92][95][98]. In the case of the EMC, a proteomic approach identified TA proteins, as well as multispanning MPs, as the predominant clients, the latter of which were also characterized as the main clients of the GEL-BOS-PAT-Sec61 supercomplex but proposed to have a preference for multispanning MPs with four or more TMDs, including those with marginal hydrophobicity [131][132]. There are two excellent reviews, plus several very recent original articles, on the structural and mechanistic details of the various MP insertases in the ER membrane [69][70][81][121][122][123][124][125][126][127][128][129][130][131][132].

5. A Single Proteomic Approach to Address the Client Spectra of Various Components for Targeting of Precursor Polypeptides to and Insertion into the Human ER Membrane

Following the pioneering work by G. Blobel and B. Dobberstein [13][14], protein import into the human ER was usually studied with the focus on single precursor polypeptides that were analyzed one-by-one in cell-free assays or intact cells. These classical studies led to the conclusions of whether and how the targeting and membrane insertion or translocation of a certain precursor is facilitated by a certain component. Recently, more global approaches were developed, such as the already mentioned proximity-specific ribosome-profiling [29][111][126][133][134] and quantitative proteomics approaches [5][106][112][126][135][136][137][138][139]. Typically, the proteomic approach employed siRNA-mediated knock-down or CRISPR/Cas9-mediated knock-out of components one-by-one in human cells, label-free quantitative proteomic analysis, and differential protein abundance analysis to characterize client specificities of components (**Figure 4, left part**). In contrast to the classical analyses, the quantitative proteomics approach addresses the question of which precursors use a certain pathway or component in intact

cells, i.e., under in-vivo-like conditions (**Figure 4, right part**).

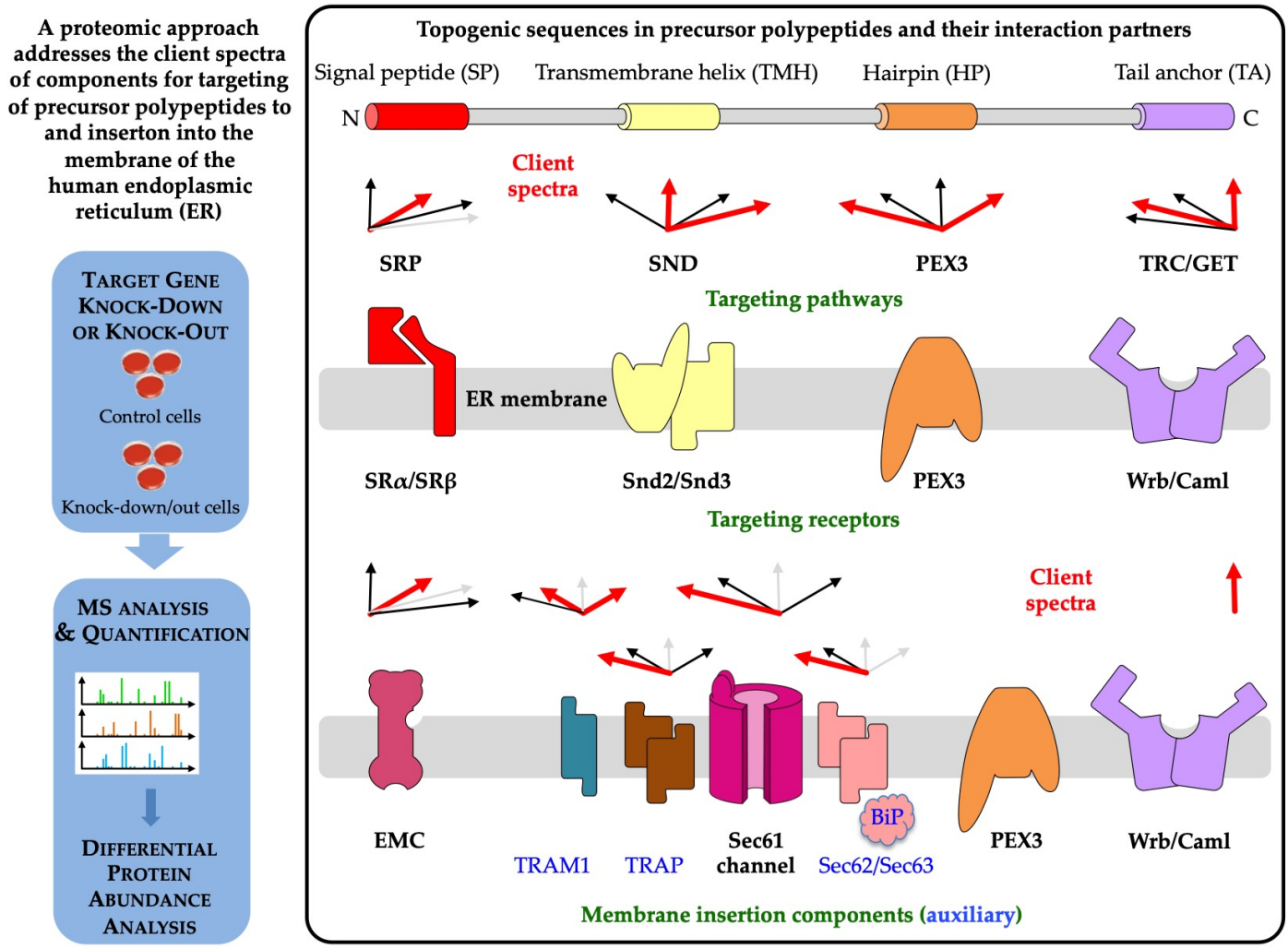

Figure 4. Components for protein targeting to and insertion into the human ER membrane with their client spectra and possible regulatory mechanisms. The first two percentages in each client spectrum refer to the percentage of precursors with SP and TMH (including HP and TA proteins), respectively, among the total number of clients, while the following two numbers refer to the percentage of precursors with HP or TA among the precursors with TMH. The red arrows highlight top scoring numbers > 10, light grey arrows represent the number 0 and are shown to indicate that fact.

References

1. Blobel, G. Intracellular protein topogenesis. Proc. Natl. Acad. Sci. USA 1980, 77, 1496–1500.
2. Egea, P.F.; Stroud, R.M.; Walter, P. Targeting proteins to membranes: Structure of the signal recognition particle. Curr. Opin. Struct. Biol. 2005, 15, 213–220.

3. Aviram, N.; Schuldiner, M. Targeting and translocation of proteins to the endoplasmic reticulum at a glance. J. Cell Sci. 2017, 130, 4079–4085.
4. Gemmer, M.; Förster, F. A clearer picture of the ER translocon complex. J. Cell Sci. 2020, 133, jcs231340.
5. Lang, S.; Nguyen, D.; Bhadra, P.; Jung, M.; Helms, V.; Zimmermann, R. Signal peptide features determining the substrate specificities of targeting and translocation components in human ER protein import. Front. Physiol. 2022, 13, 833540.
6. Sicking, M.; Lang, S.; Bochen, F.; Drenth, J.P.H.; Zacharia, M.; Zimmermann, R.; Roos, A.; Linxweiler, M. Complexity and specificity of Sec61 channelopathies: Human diseases affecting gating of the Sec61 complex. Cells 2021, 10, 1036.
7. Jansen, R.L.M.; van der Klei, I.J. The peroxisome biogenesis factors Pex3 and Pex19: Multitasking proteins with disputed functions. FEBS Lett. 2019, 593, 457–474.
8. Dhimann, R.; Caesar, S.; Thiam, A.R.; Schrul, B. Mechanisms of protein targeting to lipid droplets: A unified cell biological and biophysical perspective. Sem. Cell Dev. Biol. 2020, 108, 4–13.
9. Hansen, K.G.; Aviram, N.; Laborenz, J.; Bibi, C.; Meyer, M.; Spang, A.; Schuldiner, M.; Herrmann, J.M. An ER surface retrieval pathway safeguards the import of mitochondrial membrane proteins in yeast. Science 2018, 361, 1118–1122.
10. Koch, C.; Schuldiner, M.; Herrmann, J.M. ER-SURF: Riding the endoplasmic reticulum SURFace to mitochondria. Int. J. Mol. Sci. 2021, 22, 9655.
11. Palade, G. Intracellular aspects of protein synthesis. Science 1975, 189, 347–358.
12. Palade, G.; Porter, K.R. Studies on the endoplasmic reticulum. J. Exp. Med. 1954, 100, 641–656.
13. Blobel, G.; Dobberstein, B. Transfer of proteins across membranes: I. Presence of proteolytically processed and unprocessed nascent immunoglobulin light chains on membrane-bound ribosomes of murine myeloma. J. Cell Biol. 1975, 67, 835–851.
14. Blobel, G.; Dobberstein, B. Transfer of proteins across membranes: II. Reconstitution of functional rough microsomes from heterologous components. J. Cell Biol. 1975, 67, 852–862.
15. Von Heijne, G. Signal sequences. J. Mol. Biol. 1985, 184, 99–105.
16. Von Heijne, G.; Gavel, Y. Topogenic signals in integral membrane proteins. Eur. J. Biochem. 1988, 174, 671–678.
17. Hegde, R.S.; Bernstein, H. The surprising complexity of signal peptides. Trends Biochem. Sci. 2006, 31, 563–571.
18. Siegel, V.; Walter, P. Functional dissection of the signal recognition particle. Trends Biochem. Sci. 1988, 13, 314–316.

19. Ng, D.T.; Brown, J.D.; Walter, P. Signal sequences specify the targeting route to the endoplasmic reticulum membrane. J. Cell Biol. 1996, 134, 269–278.
20. Halic, M.; Beckmann, R. The signal recognition particle and its interactions during protein targeting. Curr. Opin. Struct. Biol. 2005, 15, 116–125.
21. Halic, M.; Blau, M.; Becker, T.; Mielke, T.; Pool, M.R.; Wild, K.; Sinning, I.; Beckmann, R. Following the signal sequence from ribosomal tunnel exit to signal recognition particle. Nature 2006, 444, 507–511.
22. Gamerdinger, M.; Hanebuth, M.A.; Frickey, T.; Deuerling, E. The principle of antagonism ensures protein targeting specificity at the endoplasmic reticulum. Science 2015, 348, 201–207.
23. Hsieh, H.-H.; Lee, J.H.; Chandrasekar, S.; Shan, S.-o. A ribosome-associated chaperone enables substrate triage in a cotranslational protein targeting complex. Nat. Commun. 2020, 11, 5840.
24. Jomaa, A.; Eitzinger, S.; Zhu, Z.; Chandrasekar, S.; Kobajashi, K.; Shan, S.-o.; Ban, N. Molecular mechanism of cargo recognition and handover by the mammalian signal recognition particle. Cell Rep. 2021, 36, 109350.
25. Jomaa, A.; Gamerdinger, M.; Hsieh, H.-H.; Wallisch, A.; Chandrasekaran, V.; Ulusoy, Z.; Scaiola, A.; Hegde, R.S.; Shan, S.-o.; Ban, N.; et al. Mechanism of signal sequence handover from NAC to SRP on ribosomes during ER-protein targeting. Science 2022, 375, 839–844.
26. Meyer, D.I.; Dobberstein, B. A membrane component essential for vectorial translocation of nascent proteins across the endoplasmic reticulum: Requirements for its extraction and reassociation with the membrane. J. Cell Biol. 1980, 87, 498–502.
27. Gilmore, R.; Blobel, G.; Walter, P. Protein translocation across the endoplasmic reticulum. I. Detection in the microsomal membrane of a receptor for the signal recognition particle. J. Cell Biol. 1982, 95, 463–469.
28. Tajima, S.; Lauffer, L.; Rath, V.L.; Walter, P. The signal recognition particle receptor is a complex that contains two distinct polypeptide chains. J. Cell Biol. 1986, 103, 1167–1178.
29. Jan, C.H.; Williams, C.C.; Weissman, J.S. Principles of ER cotranslational translocation revealed by proximity-specific ribosome profiling. Science 2014, 346, 1257521.
30. Chartron, J.W.; Hunt, K.C.L.; Frydman, J. Cotranslational signal-independent SRP preloading during membrane targeting. Nature 2016, 536, 224–228.
31. Görlich, D.; Prehn, S.; Hartmann, E.; Kalies, K.-U.; Rapoport, T.A. A mammalian homolog of SEC61p and SECYp is associated with ribosomes and nascent polypeptides during translocation. Cell 1992, 71, 489–503.
32. Görlich, D.; Rapoport, T.A. Protein translocation into proteoliposomes reconstituted from purified components of the endoplasmic reticulum membrane. Cell 1993, 75, 615–630.

33. Simon, S.M.; Blobel, G. A protein-conducting channel in the endoplasmic reticulum. Cell 1991, 65, 371–380.
34. Wirth, A.; Jung, M.; Bies, C.; Frien, M.; Tyedmers, J.; Zimmermann, R.; Wagner, R. The Sec61p complex is a dynamic precursor activated channel. Mol. Cell 2003, 12, 261–268.
35. Beckmann, R.; Spahn, C.M.; Eswar, N.; Helmers, J.; Penczek, P.A.; Sali, A.; Frank, J.; Blobel, G. Architecture of the protein-conducting channel associated with the translating 80S ribosome. Cell 2001, 107, 361–372.
36. Van den Berg, B.; Clemons, W.M.; Collinson, I.; Modis, Y.; Hartmann, E.; Harrison, S.C.; Rapoport, T.A. X-ray structure of a protein-conducting channel. Nature 2004, 427, 36–44.
37. Pfeffer, S.; Brandt, F.; Hrabe, T.; Lang, S.; Eibauer, M.; Zimmermann, R.; Förster, F. Structure and 3D arrangement of ER-membrane associated ribosomes. Structure 2012, 20, 1508–1518.
38. Pfeffer, S.; Dudek, J.; Gogala, M.; Schorr, S.; Linxweiler, J.; Lang, S.; Becker, T.; Beckmann, R.; Zimmermann, R.; Förster, F. Structure of the mammalian oligosaccharyltransferase in the native ER protein translocon. Nat. Commun. 2014, 5, 3072.
39. Voorhees, R.M.; Fernández, I.S.; Scheres, S.H.W.; Hegde, R.S. Structure of the mammalian ribosome-Sec61 complex to 3.4 Å resolution. Cell 2014, 157, 1632–1643.
40. Voorhees, R.M.; Hegde, R.S. Structure of the Sec61 channel opened by a signal peptide. Science 2016, 351, 88–91.
41. Pfeffer, S.; Burbaum, L.; Unverdorben, P.; Pech, M.; Chen, Y.; Zimmermann, R.; Beckmann, R.; Förster, F. Structure of the native Sec61 protein-conducting channel. Nat. Commun. 2015, 6, 8403.
42. Wiedmann, M.; Kurzchalia, T.V.; Hartmann, E.; Rapoport, T.A. A signal sequence receptor in the endoplasmic reticulum membrane. Nature 1987, 328, 830–833.
43. Fons, R.D.; Bogert, B.A.; Hegde, R.S. Substrate-specific function of the translocon-associated protein complex during translocation across the ER membrane. J. Cell Biol. 2003, 160, 529–539.
44. Menetret, J.F.; Hegde, R.S.; Aguiar, M.; Gygi, S.P.; Park, E.; Rapoport, T.A.; Akey, C.W. Single copies of Sec61 and TRAP associate with a nontranslating mammalian ribosome. Structure 2008, 16, 1126–1137.
45. Sommer, N.; Junne, T.; Kalies, K.-U.; Spiess, M.; Hartmann, E. TRAP assists membrane protein topogenesis at the mammalian ER membrane. Biochim. Biophys. Acta 2013, 1833, 3104–3111.
46. Pfeffer, S.; Dudek, J.; Ng, B.; Schaffa, M.; Albert, S.; Plitzko, J.; Baumeister, W.; Zimmermann, R.; Freeze, H.F.; Engel, B.D.; et al. Dissecting the molecular organization of the translocon-associatecd protein complex. Nat. Commun. 2017, 8, 14516.

47. Jaskolowski, M.; Jomaa, A.; Gamerdinger, M.; Shresta, S.; Leibundgut, M.; Deuerling, E.; Ban, N. Molecular basis of the TRAP complex function in ER protein biogenesis. Nat. Struct. Mol. Biol. 2023, 375, 839–844.
48. Pauwels, E.; Shewakramani, N.R.; De Wijngaert, B.; Camps, A.; Provinciael, B.; Stroobants, J.; Kalies, K.-U.; Hartmann, E.; Maes, P.; Vermeire, K.; et al. Structural insights into TRAP association with ribosome-Sec61 complex and translocon inhibition by a CADA derivative. Sci. Adv. 2023, 9, eadf0797.
49. Dierks, T.; Volkmer, J.; Schlenstedt, G.; Jung, C.; Sandholzer, U.; Zachmann, K.; Schlotterhose, P.; Neifer, K.; Schmidt, B.; Zimmermann, R. A microsomal ATP-binding protein involved in efficient protein transport into the mammalian endoplasmic reticulum. EMBO J. 1996, 15, 6931–6942.
50. Tyedmers, J.; Lerner, M.; Bies, C.; Dudek, J.; Skowronek, M.H.; Haas, I.G.; Heim, N.; Nastainczyk, W.; Volkmer, J.; Zimmermann, R. Homologs of the yeast Sec complex subunits Sec62p and Sec63p are abundant proteins in dog pancreas microsomes. Proc. Natl. Acad. Sci. USA 2000, 97, 7214–7219.
51. Mayer, H.-A.; Grau, H.; Kraft, R.; Prehn, S.; Kalies, K.-U.; Hartmann, E. Mammalian Sec61 is associated with Sec62 and Sec63. J. Biol. Chem. 2000, 275, 14550–14557.
52. Tyedmers, J.; Lerner, M.; Wiedmann, M.; Volkmer, J.; Zimmermann, R. Polypeptide chain binding proteins mediate completion of cotranslational protein translocation into the mammalian endoplasmic reticulum. EMBO Rep. 2005, 4, 505–510.
53. Lakkaraju, A.K.K.; Thankappan, R.; Mary, C.; Garrison, J.L.; Taunton, J.; Strub, K. Efficient secretion of small proteins in mammalian cells relies on Sec62-dependent posttranslational translocation. Mol. Biol. Cell 2012, 23, 2712–2722.
54. Lang, S.; Benedix, J.; Fedeles, S.V.; Schorr, S.; Schirra, C.; Schäuble, N.; Jalal, C.; Greiner, M.; Haßdenteufel, S.; Tatzelt, J.; et al. Different effects of Sec61α-, Sec62 and Sec63-depletion on transport of polypeptides into the endoplasmic reticulum of mammalian cells. J. Cell Sci. 2012, 125, 1958–1969.
55. Jadhav, B.; McKenna, M.; Johnson, N.; High, S.; Sinning, I.; Pool, M.R. Mammalian SRP receptor switches the Sec61 translocase from Sec62 to SRP-dependent translocation. Nat. Commun. 2015, 6, 10133.
56. Conti, B.J.; Devaraneni, P.K.; Yang, Z.; David, L.L.; Skach, W.R. Cotranslational stabilization of Sec62/63 within the ER Sec61 translocon is controlled by distinct substrate-driven translocation events. Mol. Cell 2015, 58, 269–283.
57. Haßdenteufel, S.; Johnson, N.; Paton, A.W.; Paton, J.C.; High, S.; Zimmermann, R. Chaperone-mediated Sec61 channel gating during ER import of small precursor proteins overcomes Sec61 inhibitor-reinforced energy barrier. Cell Rep. 2018, 23, 1373–1386.

58. Haßdenteufel, S.; Nguyen, D.; Helms, V.; Lang, S.; Zimmermann, R. Components and mechanisms for ER import of small human presecretory proteins. FEBS Lett. 2019, 593, 2506–2524.
59. Itskanov, S.; Park, E. Structure of the posttranslational Sec protein-translocation channel complex from yeast. Science 2019, 363, 84–87.
60. Wu, X.; Cabanos, C.; Rapoport, T.A. Structure of the post-translational protein translocation machinery of the ER membrane. Nature 2019, 566, 136–139.
61. Itskanov, S.; Kuo, K.M.; Gumbart, J.C.; Park, E. Stepwise gating of the Sec61 protein-conducting channel by Sec62 and Sec63. Nat. Struct. Mol. Biol. 2021, 28, 162–172.
62. Weng, T.-H.; Steinchen, W.; Beatrix, B.; Berninghausen, O.; Becker, T.; Bange, G.; Cheng, J.; Beckmann, R. Architecture of the active post-translational SEC translocon. EMBO J. 2021, 40, e105643.
63. Görlich, D.; Hartmann, E.; Prehn, S.; Rapoport, T.A. A protein of the endoplasmic reticulum involved early in polypeptide translocation. Nature 1992, 357, 47–52.
64. High, S.; Martoglio, B.; Görlich, D.; Andersen, S.S.L.; Ashford, A.A.; Giner, A.; Hartmann, E.; Prehn, S.; Rapoport, T.A.; Dobberstein, B.; et al. Site-specific photocross-linking reveals that Sec61p and TRAM contact different regions of a membrane-inserted signal sequence. J. Biol. Chem. 1993, 268, 26745–26751.
65. Hegde, R.S.; Voigt, S.; Rapoport, T.A.; Lingappa, V.R. TRAM regulates the exposure of nascent secretory proteins to the cytosol during translocation into the endoplasmic reticulum. Cell 1998, 92, 621–631.
66. Voigt, S.; Jungnickel, B.; Hartmann, E.; Rapoport, T.A. Signal sequence-dependent function of the TRAM protein during early phases of protein transport across the endoplasmic reticulum membrane. J. Cell Biol. 1996, 134, 25–35.
67. Sauri, A.; McCormick, P.J.; Johnson, A.E.; Mingarro, I. Sec61alpha and TRAM are sequentially adjacent to a nascent viral membrane protein during its ER integration. J. Mol. Biol. 2007, 366, 366–374.
68. Cohen, N.; Aviram, N.; Schuldiner, M. A systematic proximity ligation approach to studying protein-substrate specificity identifies the substrate spectrum of the Ssh1 translocon. EMBO J. 2023, 43, e113385.
69. O'Keefe, S.; Pool, M.R.; High, S. Membrane protein biogenesis at the ER: The highways and byways. FEBS J. 2022, 289, 6835–6862.
70. Hegde, R.S.; Keenan, R.J. The mechanism of integral membrane protein biogenesis. Nat. Rev. Mol. Cell Biol. 2022, 23, 107–124.

71. Schrul, B.; Kopito, R.R. Peroxin-dependent targeting of a lipid-droplet-destined membrane protein to ER subdomains. Nat. Cell Biol. 2016, 18, 740–751.
72. Schrul, B.; Schliebs, W. Intracellular communication between lipid droplets and peroxisomes: The Janus face of PEX19. Biol. Chem. 2018, 399, 741–749.
73. Yamamoto, Y.; Sakisaka, T. The peroxisome biogenesis factors posttranslationally target reticulon homology-domain containing proteins to the endoplasmic reticulum membrane. Sci. Rep. 2018, 8, 2322.
74. Aviram, N.; Ast, T.; Costa, E.A.; Arakel, E.; Chuartzman, S.G.; Jan, C.H.; Haßdenteufel, S.; Dudek, J.; Jung, M.; Schorr, S.; et al. The SND proteins constitute an alternative targeting route to the endoplasmic reticulum. Nature 2016, 540, 134–138.
75. Hirata, T.; Yang, J.; Tomida, S.; Tokoro, Y.; Kinoshita, T.; Fujita, M.; Kizuka, Y. ER entry pathway and glycosylation of GPI-anchored proteins are determined by N-terminal signal sequence and C-terminal GPI-attachment sequence. J. Biol. Chem. 2022, 298, 102444.
76. Kalies, K.-U.; Rapoport, T.A.; Hartmann, E. The beta-subunit of the Sec61 complex facilitates cotranslational protein transport and interacts with the signal peptidase during translocation. J. Cell Biol. 1998, 141, 887–894.
77. Chen, X.; Van Valkenburgh, C.; Liang, H.; Fang, H.; Green, N. Signal peptidase and oligosaccharyltransferase interact in a sequential and dependent manner within the endoplasmic reticulum. J. Biol. Chem. 2001, 276, 2411–2416.
78. Liaci, A.M.; Steigenberger, B.; Tamara, S.; de Souza, P.T.; Gröllers-Mulderij, M.; Ogrissek, P.; Marrink, S.-J.; Scheltema, R.A.; Förster, F. Structure of the human signal peptidase complex reveals the determinants for signal petide cleavage. Mol. Cell 2021, 81, 3934–3948.
79. Braunger, K.; Pfeffer, S.; Shrimal, S.; Gilmore, R.; Berninhausen, O.; Mandon, E.C.; Becker, T.; Förster, F.; Beckmann, R. Structural basis for coupling protein transport and N-glycosylation at the mammalian endoplasmic reticulum. Science 2018, 360, 215–219.
80. Wild, R.; Kowal, J.; Eyring, J.; Ngwa, E.M.; Aebi, M.; Locher, K.P. Structure of the yeast oligosaccharyltransferase complex gives insight into eukaryotic N-glycosylation. Science 2018, 359, 545–550.
81. Gemmer, M.; Chaillet, M.; van Loenhout, J.; Arenas, R.C.; Vismpas, D.; Gröllers-Mulderji, M.; Kohl, F.A.; Albanese, P.; Scheltema, R.A.; Howes, S.C.; et al. Visualization of translation and protein biogenesis at the ER membrane. Nature 2023, 614, 160–167.
82. Müller, G.; Zimmermann, R. Import of honeybee prepromelittin into the endoplasmic reticulum: Structural basis for independence of SRP and docking protein. EMBO J. 1987, 6, 2099–2107.

83. Müller, G.; Zimmermann, R. Import of honeybee prepromelittin into the endoplasmic reticulum: Energy requirements for membrane insertion. EMBO J. 1988, 7, 639–648.
84. Schlenstedt, G.; Zimmermann, R. Import of frog prepropeptide GLa into microsomes requires ATP but does not involve docking protein or ribosomes. EMBO J. 1987, 6, 699–703.
85. Schlenstedt, G.; Gudmundsson, G.H.; Boman, H.G.; Zimmermann, R. A large presecretory protein translocates both cotranslationally, using signal recognition particle and ribosome, and posttranslationally, without these ribonucleoparticles, when synthesized in the presence of mammalian microsomes. J. Biol. Chem. 1990, 265, 13960–13968.
86. Kutay, U.; Hartmann, E.; Rapoport, T.A. A class of membrane proteins with a C-terminal anchor. Trends Cell Biol. 1993, 3, 72–75.
87. Ast, T.; Cohen, G.; Schuldiner, M. A network of cytosolic factors targets SRP-independent proteins to the endoplasmic reticulum. Cell 2013, 152, 1134–1145.
88. Schuldiner, M.; Metz, J.; Schmid, V.; Denic, V.; Rakwalska, M.; Schmitt, H.D.; Schwappach, B.; Weissman, J.S. The GET complex mediates insertion of tail-anchored proteins into the ER membrane. Cell 2008, 134, 634–645.
89. Rabu, C.; Schmid, V.; Schwappach, B.; High, S. Biogenesis of tail-anchored proteins: The beginning for the end? J. Cell Sci. 2009, 122, 3605–3612.
90. Mariappan, M.; Li, X.; Stefanovic, S.; Sharma, A.; Mateja, A.; Keenan, R.J.; Hegde, R.S. A ribosome-associating factor chaperones tail-anchored membrane proteins. Nature 2010, 466, 1120–1124.
91. Leznicki, P.; Clancy, A.; Schwappach, B.; High, S. Bat3 promotes the membrane integration of tail-anchored proteins. J. Cell Sci. 2010, 123, 2170–2178.
92. Borgese, N.; Fasana, E. Targeting pathways of C-tail-anchored proteins. Biochim. Biophys. Acta 2011, 1808, 937–946.
93. Vilardi, F.; Lorenz, H.; Dobberstein, B. WRB is the receptor for TRC40/Asna1-mediated insertion of tail-anchored proteins into the ER membrane. J. Cell Sci. 2011, 124, 1301–1307.
94. Leznicki, P.; Warwicker, J.; High, S. A biochemical analysis of the constraints of tail-anchored protein biogenesis. Biochem. J. 2011, 436, 719–727.
95. Yamamoto, Y.; Sakisaka, T. Molecular machinery for insertion of tail-anchored membrane proteins into the endoplasmic reticulum membrane in mammalian cells. Mol. Cell 2012, 48, 387–397.
96. Vilardi, F.; Stephan, M.; Clancy, A.; Janshoff, A.; Schwappach, B. WRB and CAML are necessary and sufficient to mediate tail-anchored protein targeting to the ER membrane. PLoS ONE 2014, 9, e85033.

97. Wang, F.; Chan, C.; Weir, N.R.; Denic, V. The Get1/2 transmembrane complex is an endoplasmic-reticulum membrane protein insertase. Nature 2014, 512, 441–444.
98. Borgese, N.; Coy-Vergara, J.; Colombo, S.F.; Schwappach, B. The ways of tails: The GET pathway and more. Proteins 2019, 38, 289–305.
99. Leznicki, P.; High, S. SGTA associates with nascent membrane protein precursors. EMBO Rep. 2020, 21, e48835.
100. Leznicki, P.; Schneider, H.O.; Harvey, J.V.; Shi, W.Q.; High, S. Co-translational biogenesis of lipid droplet integral membrane proteins. J. Cell Sci. 2021, 132, jcs.259220.
101. Johnson, N.; Vilardi, F.; Lang, S.; Leznicki, P.; Zimmermann, R.; High, S. TRC-40 can deliver short secretory proteins to the Sec61 translocon. J. Cell Sci. 2012, 125, 3612–3620.
102. Casson, J.; McKenna, M.; Haßdenteufel, S.; Aviram, N.; Zimmermann, R.; High, S. Multiple pathways facilitate the biogenesis of mammalian tail-anchored proteins. J. Cell Sci. 2017, 130, 3851–3861.
103. Haßdenteufel, S.; Sicking, M.; Schorr, S.; Aviram, N.; Fecher-Trost, C.; Schuldiner, M.; Jung, M.; Zimmermann, R.; Lang, S. hSnd2 protein represents an alternative targeting factor to the endoplasmic reticulum in human cells. FEBS Lett. 2017, 591, 3211–3224.
104. Talbot, B.E.; Vandorpe, D.H.; Stotter, B.R.; Alper, S.L.; Schlondorff, J. Transmembrane insertases and N-glycosylation crtically determine synthesis, trafficking, and activity of the nonselective cation channel TRPC6. J. Biol. Chem. 2019, 294, 12655–12669.
105. Yang, J.; Hirata, T.; Liu, Y.-S.; Guo, X.-Y.; Gao, X.-D.; Kinoshita, T.; Fujita, M. Human SND2 mediates ER targeting of GPI-anchored proteins with low hydrophobic GPI attachment signals. FEBS Lett. 2021, 595, 1542–1558.
106. Tirincsi, A.; O'Keefe, S.; Nguyen, D.; Sicking, M.; Dudek, J.; Förster, F.; Jung, M.; Hadzibeganovic, D.; Helms, V.; High, S.; et al. Proteomics identifies substrates and a novel component in hSnd2-dependent ER protein targeting. Cells 2022, 11, 2925.
107. Seiser, R.M.; Nicchitta, C.V. The fate of membrane-bound ribosomes following the termination of protein synthesis. J. Biol. Chem. 2000, 275, 33820–33827.
108. Potter, M.D.; Seiser, R.M.; Nicchitta, C.V. Ribosome exchange revisited: A mechanism for translation-coupled ribosome detachment from the ER membrane. Trends Cell Biol. 2001, 11, 112–115.
109. Berkovits, B.D.; Mayr, C. Alternative 3′UTRs act as scaffolds to regulate membrane protein localization. Nature 2015, 522, 363–367.
110. Ma, W.; Mayr, C. A membraneless organelle associated with the endoplasmic reticulum enables 3′UTR-mediated protein-protein interactions. Cell 2018, 175, 1492–1506.

111. Hsu, J.C.-C.; Reid, D.W.; Hoffman, A.M.; Sarkar, D.; Nicchitta, C.V. Oncoprotein AEG-1 is an endoplasmic reticulum RNA-binding protein whose interactome is enriched in organelle resident protein-encoding mRNAs. RNA 2018, 24, 688–703.

112. Bhadra, P.; Schorr, S.; Lerner, M.; Nguyen, D.; Dudek, J.; Förster, F.; Helms, V.; Lang, S.; Zimmermann, R. Quantitative proteomics and differential protein abundance analysis after depletion of putative mRNA receptors in the ER membrane of human cells identifies novel aspects of mRNA targeting to the ER. Molecules 2021, 26, 3591.

113. Savitz, A.J.; Meyer, D.I. Identification of a ribosome receptor in the rough endoplasmic reticulum. Nature 1990, 346, 540–544.

114. Wiedmann, B.; Saki, H.; Davis, T.A.; Wiedmann, M. A protein complex required for signal-sequence-specific sorting and translocation. Nature 1994, 370, 434–440.

115. Gamerdinger, M.; Kobayashi, K.; Wallisch, A.; Kreft, S.G.; Sailer, C.; Schlömer, R.; Sachs, N.; Jomaa, A.; Stengel, F.; Ban, N.; et al. Early scanning of nascent polypeptides inside the ribosomal tunnel by NAC. Mol. Cell 2019, 75, 996–1006.

116. Moeller, I.; Jung, M.; Beatrix, B.; Levy, R.; Kreibich, G.; Zimmermann, R.; Wiedmann, M.; Lauring, B. A general mechanism for regulation of access to the translocon: Competition for a membrane attachment site on ribosomes. Proc. Natl. Acad. Sci. USA 1998, 95, 13425–13430.

117. Goder, V.; Spiess, M. Molecular mechanism of signal sequence orientation in the endoplasmic reticulum. EMBO J. 2003, 22, 3645–3653.

118. Goder, V.; Junne, T.; Spiess, M. Sec61p contributes to signal sequence orientation according to the positive-inside rule. Mol. Biol. Cell 2004, 15, 1470–1478.

119. Baker, J.A.; Wong, W.-C.; Eisenhaber, B.; Warwicker, J.; Eisenhaber, F. Charged residues next to transmembrane regions revisited: “Positive-inside rule” is complemented by the “negative inside depletion/outside enrichment rule”. BMC Biol. 2017, 15, 66.

120. Devaraneni, P.K.; Conti, B.; Matsumara, Y.; Yang, Z.; Johnson, A.E.; Skach, W.R. Stepwise insertion and inversion of a type II signal anchor sequence in the ribosome-Sec61 translocon complex. Cell 2011, 146, 134–147.

121. Meacock, S.L.; Lecomte, F.J.L.; Crawshaw, S.G.; High, S. Different transmembrane domains associate with distinct endoplasmic reticulum components during membrane integration of a polytopic protein. Mol. Biol. Cell 2002, 13, 4114–4129.

122. Ismail, N.; Crawshaw, S.G.; High, S. Active and passive displacement of transmembrane domains both occur during opsin biogenesis at the Sec61 translocon. J. Cell Sci. 2006, 119, 2826–2836.

123. Anghel, S.A.; McGilvray, P.T.; Hegde, R.S.; Keenan, R.J. Identification of Oxa1 homologs operating in the eukaryotic endoplasmic reticulum. Cell Rep. 2017, 21, 3708–3716.

124. Chitwood, P.J.; Juszkiewicz, S.; Guna, A.; Shao, S.; Hegde, R.S. EMC is required to initiate accurate membrane protein topogenesis. Cell 2018, 175, 1507–1519.
125. Shurtleff, M.J.; Itzhak, D.N.; Hussmann, J.A.; Schirle Oakdale, N.T.; Costa, E.A.; Jonikas, M.; Weibezahn, J.; Popova, K.D.; Jan, C.H.; Sinitcyn, P.; et al. The ER membrane protein complex interacts cotranslationally to enable biogenesis of multipass membrane proteins. eLife 2018, 7, e37018.
126. McGilvray, P.T.; Anghel, S.A.; Sundaram, A.; Zhong, F.; Trnka, M.J.; Fuller, J.R.; Hu, H.; Burlingame, A.L.; Keenan, R.J. An ER translocon for multi-pass mambrane protein biogenesis. eLife 2020, 9, e56889.
127. O'Donnel, J.P.; Philips, B.P.; Yagita, Y.; Juszkiewicz, S.; Wagner, A.; Malinverni, D.; Keenan, R.J.; Mille, E.A.; Hegde, R.S. The architecture of EMC reveals a path for membrane protein nsertion. eLife 2020, 9, e57887.
128. Pleiner, T.; Tomaleri, G.P.; Januszyk, K.; Inglis, A.J.; Hazu, M.; Voorhees, R.M. Structural basis for membrane insertion by the human ER membrane protein complex. Science 2020, 369, 433–436.
129. Bai, L.; You, Q.; Feng, X.; Kovach, A.; Li, H. Structure of the ER membrane complex, a transmembrane insertase. Nature 2020, 584, 475–478.
130. Wu, H.; Hegde, R.S. Mechanism of signal-anchor triage during early steps of membrane protein insertion. Cell 2023, 83, 961–973.
131. Sundaram, A.; Yamsek, M.; Zhong, F.; Hooda, Y.; Hegde, R.S.; Keenan, R.J. Substrate-driven assembly of a translocon for multipass membrane proteins. Nature 2022, 611, 167–172.
132. Samlinskaite, L.; Kim, M.K.; Lewis, A.J.O.; Keenan, R.J.; Hegde, R.S. Mechanism of an intramembrane chaperone for multipass membrane proteins. Nature 2022, 611, 161–166.
133. Reid, D.W.; Nicchitta, C.V. Primary role for endoplasmic reticulum-bound ribosomes in cellular translation identified by ribosome profiling. J. Biol. Chem. 2012, 287, 5518–5527.
134. Hannigan, M.M.; Hoffman, A.M.; Thompson, J.W.; Zheng, T.; Nicchitta, C.V. Quantitative proteomics links the LRRC59 interactome to mRNA translation on the ER membrane. Mol. Cell. Proteom. 2020, 19, 1826–1849.
135. Nguyen, D.; Stutz, R.; Schorr, S.; Lang, S.; Pfeffer, S.; Freeze, H.F.; Förster, F.; Helms, V.; Dudek, J.; Zimmermann, R. Proteomics reveals signal peptide features determining the client specificity in human TRAP-dependent ER protein import. Nat. Commun. 2018, 9, 37639.
136. Schorr, S.; Nguyen, D.; Haßdenteufel, S.; Nagaraj, N.; Cavalié, A.; Greiner, M.; Weissgerber, P.; Loi, M.; Paton, A.W.; Paton, J.C.; et al. Proteomics identifies signal peptide features determining the substrate specificity in human Sec62/Sec63-dependent ER protein import. FEBS J. 2020, 287, 4612–4640.

137. Klein, M.-C.; Lerner, M.; Nguyen, D.; Pfeffer, S.; Dudek, J.; Förster, F.; Helms, V.; Lang, S.; Zimmermann, R. TRAM1 protein may support ER protein import by modulating the phospholipid bilayer near the lateral gate of the Sec61 channel. Channels 2020, 14, 28–44.

138. Tian, S.; Wu, Q.; Zhou, B.; Choi, M.Y.; Ding, B.; Yang, W.; Dong, M. Proteomic analysis indentifies membrane proteins dependent on the ER membrane protein complex. Cell Rep. 2019, 28, 2517–2526.

139. Zimmermann, R.; Lang, S.; Lerner, M.; Förster, F.; Nguyen, D.; Helms, V.; Schrul, B. Quantitative proteomics and differential protein abundance ananalysis after depletion of PEX3 from human cells identifies additional aspects of protein targeting tot he ER. Int. J. Mol. Sci. 2021, 22, 13028.