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ABC Transporters and Immunity: Mechanism of Self-Defense

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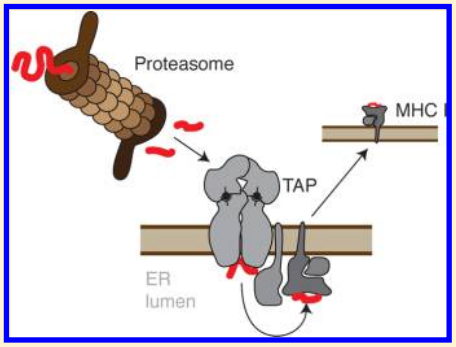
ABSTRACT: The transporter associated with antigen processing (TAP) is a prototype of an asymmetric ATP-binding cassette (ABC) transporter, which uses ATP binding and hydrolysis to translocate peptides from the cytosol to the lumen of the endoplasmic reticulum (ER). Here, we review molecular details of peptide binding and ATP binding and hydrolysis as well as the resulting allosteric cross-talk between the nucleotide-binding domains and the transmembrane domains that drive translocation of the solute across the ER membrane. We also discuss the general molecular architecture of ABC transporters and demonstrate the importance of structural and functional studies for a better understanding of the role of the noncanonical site of asymmetric ABC transporters. Several aspects of peptide binding and specificity illustrate details of peptide translocation by TAP. Furthermore, this ABC transporter forms the central part of the major histocompatibility complex class I (MHC I) peptide-loading machinery. Hence, TAP is confronted with a number of viral factors, which prevent antigen translocation and MHC I loading in virally infected cells. We review how these viral factors have been used as molecular tools to decipher mechanistic aspects of solute translocation and discuss how they can help in the structural analysis of TAP.



The clearance of infected or malignant cells by cytotoxic T-lymphocytes (CTLs) relies on the presentation of antigens on major histocompatibility complex class I (MHC I) molecules at the cell surface. CTLs recognize MHC I molecules in complex with antigenic peptides and finally eliminate the target cell. Antigen presentation critically depends on the translocation of proteasomal degradation products from the cytosol to the lumen of the endoplasmic reticulum (ER). The transporter associated with antigen processing (TAP) mediates peptide recognition and delivery to MHC I molecules as final acceptors. Peptide loading of MHC I molecules is catalyzed by several ER-resident chaperones (tapasin, ERp57, and calreticulin), resulting in the formation of the macromolecular MHC I peptide-loading complex (PLC). ER-associated amino peptidases (ERAAP1 and -2) finally trim the N-terminus of the peptide, so that they optimally fit into the MHC I binding pocket. After passing the ER quality control, kinetically stable peptide–MHC complexes are transported to the cell surface, where they present their antigenic cargo to CTLs.[1−3] In professional antigen-presenting cells such as dendritic cells (DCs), TAP is also involved in the cross-presentation of exogenous antigens on MHC I molecules in endosomal and phagosomal compartments.[4−6] Because TAP is a key player in the adaptive immune system, viruses have evolved several immune evasion strategies to interfere with peptide loading of MHC I. Members of the Herpesviridae family encode proteins that block the TAP function, leading to the downregulation of MHC I surface expression. Here, we summarize recent findings about the mechanism and viral modulation of the TAP complex as the cardinal component of the antigen processing pathway.

MHC I ASSEMBLY LINE: THE PEPTIDE-LOADING COMPLEX

Similar to an assembly line in the automotive industry, the peptide-loading complex (PLC) guarantees the efficient release of assembled peptide–MHC I complexes by linking the translocation complex TAP to the ER quality control machinery. Thus, the selection of kinetically stable peptide–MHC complexes occurs, which move via the secretory pathway to the cell surface where they are inspected by CTLs for their antigenic cargo.[3] The PLC consists of six different proteins that interact either temporarily or permanently with each other (Figure 1). The heterodimeric ATP-binding cassette (ABC) transport complex is responsible for the recognition of proteasomal degradation products in the cytosol and their subsequent translocation into the ER lumen. Each of the two TAP subunits consists of a transmembrane domain (TMD) followed by a nucleotide-binding domain (NBD). In the ER lumen, peptide loading of MHC I is catalyzed by the type I membrane glycoprotein tapasin (Tsn) that binds to a unique N-terminal domain (TMD_0) of each TAP subunit.[7−10] Tapasin, which is linked to the oxidoreductase ERp57 via an intermolecular disulfide bridge ($C95^{Tsn}$–$C57^{ERp57}$),[11,12] binds to preassembled MHC I composed of the heavy chain (hc) and β_2-microglobulin (β_2m).[13,14] MHC I molecules are further stabilized by the lectin-like chaperone calreticulin (Crt), which recognizes both monoglucosylated *N*-glycans of MHC I heavy chains via its lectin domain[15,16] and the β'-domain of ERp57 via its flexible proline-rich loop.[17] Within this transient subcomplex

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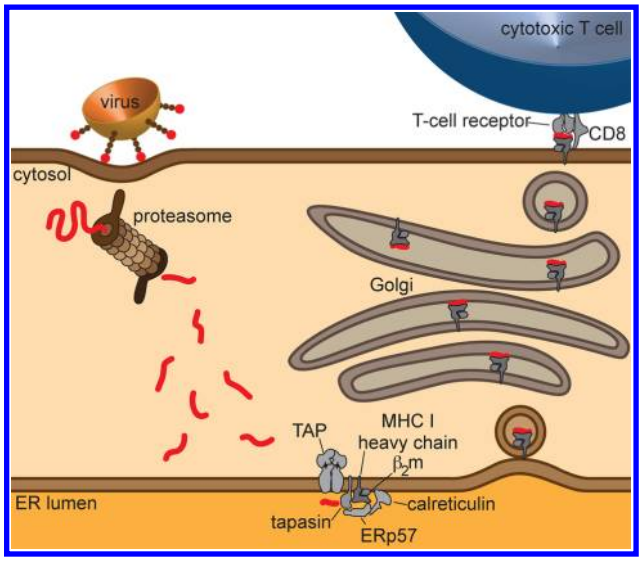

Figure 1. Antigen processing by the MHC I peptide-loading complex (PLC). Viral or tumor-associated antigens are degraded by the proteasome, and peptides are translocated into the ER lumen by TAP. MHC I molecules are loaded with high-affinity peptides and subsequently move via the Golgi apparatus to the cell surface. Cytotoxic T-cells recognize loaded MHC I complexes via their receptors, which leads to the apoptosis of the target cell.

·Crt·MHC I·Tsn−ERp57·, stabilized by multivalent, low-affinity interactions, the peptide association and dissociation rates are increased by putatively opening the binding pocket of MHC I.[18]

The peptide pool presented on MHC I is well balanced by (i) the proteolytic specificity of the proteasome and other proteases, (ii) peptide recognition and translocation by the TAP complex, and (iii) binding affinities of individual MHC I alleles, including the editing function of tapasin. TAP preferentially translocates peptides with a length of 8−12 amino acids, which matches the length of the peptide-binding pocket of MHC I.[19] The cleavage preference of the proteasome, yielding peptides with C-terminal residues that fit well with the binding specificities of TAP and MHC I. In contrast, the specificities of TAP and MHC I do not necessarily overlap for the N-terminal peptide residues, thus requiring editing in the ER lumen.[20] Thus, the ER-associated aminopeptidases (ERAAP1 and -2 in humans) are involved in the N-terminal trimming of peptides, optimizing their affinity for MHC I.[21−23]

Although the individual subunits and their contributions within the PLC are well characterized, their stoichiometry and spatiotemporal organization are unknown. After quantitative [^{35}S]methionine labeling, a model in which the heterodimeric TAP complex recruits four tapasin molecules was proposed.[24] On the other side, only two tapasin molecules per TAP were found using native polyacrylamide gel electrophoresis,[25] whereas another report proposed one tapasin associated with TAP.[26] Because the detergent is very critical for the integrity of the PLC, the structural organization of the PLC is very hard to determine, and its apparent size might be much larger, as reflected by the very slow lateral diffusion and immobile fraction of GFP-tagged TAP complexes in the ER membrane.[27,28] Thus, the stoichiometry of TAP and tapasin as well as the overall structural organization of all components within the PLC remains to be captured by biochemists, biophysicists, and cell biologists.

THE TAP COMPLEX: A PROTOTYPE OF AN ABC EXPORT SYSTEM

The two NBDs of the TAP complex convert the chemical energy of ATP into conformational changes, which are transmitted to the two TMDs to mediate peptide translocation. This four-domain architecture represents the general blueprint of an ABC transporter, although the gene organization of this protein family varies from single genes encoding all four domains, e.g., multidrug transporter P-glycoprotein (P-gp) or cystic fibrosis transmembrane conductance regulator (CFTR), to the expression of two half-transporters, e.g., TAP1 and -2, up to four polypeptides in most bacterial ABC import systems, e.g., $BtuC_2D_2$, $MalFGK_2$, or OppBCDF. Thus, despite the fact that ABC transporters deal with a very wide range of substrates, their domain architecture is conserved. The TMDs embedded in the membrane form the solute translocation pathway, whereas the NBDs facing the cytosol bind and hydrolyze ATP (Figure 2).

ABC transporters can be classified into at least three different types on the basis of the direction of transport as well as the overall architecture.[2,29,30] Type I import systems like the bacterial maltose permease and the molybdate transporter have a core of 2 × 5 TM helices with TM2−TM5 forming the substrate translocation pathway and TM1 wrapping up these helices.[31,32] Type II importers, including the bacterial vitamin B_{12} $BtuC_2D_2$ and the molybdate transporter HI14570/71, are composed of a set of 2 × 10 TMs with TM5 and TM10 dominating the interface between the TMDs and TM2−TM5 and TM7−TM10 arranged in a pseudorotational symmetry.[33,34] Eukaryotic ABC transporters are almost exclusively formed by ABC exporters (the structure of the type III transporter has been resolved), which typically expel solutes from the cytosol out of the cell or into subcellular compartments, such as the lumen of the ER, lysosomes, and peroxisomes. The TMDs form a wing-like structure by intertwining TM1 and TM2 from one subunit and TM3−TM6 from the other.[35−37] Long cytosolic loops (CLs) extending the transmembrane helices by ∼25 Å connect TM2 with TM3 and TM4 with TM5 by short coupling helices (CH1 and CH2). The CHs interact with both NBDs, forming the transition interface between the NBDs and the TMDs. CH1 contacts the Q-loop of the NBD in cis, sensing the nucleotide binding status during the catalytic cycle,[38] whereas CH2 interacts with the Q-loop in trans as shown for CFTR.[39] The X-loop is coordinated by both coupling helices in trans.[40]

Both TAP subunits can be dissected into two independent modules with an extra N-terminal domain (named TMD_0, composed of four transmembrane-spanning helices), recruiting tapasin and subsequently initiating the formation of the PLC,[7,9,10] and TM1−TM6 in combination with the NBDs, representing a minimal functional unit of TAP and therefore named coreTAP.[7,41] The sequences of coreTAP subunits are significantly similar with those of homodimeric ABC transporter Sav1866 from *Staphylococcus aureus*,[36] the bacterial lipid flippase MsbA,[37] the heterodimeric ABC transporter TM287/288 from *Thermotoga maritima*,[42] and mouse P-glycoprotein.[35] Using homology modeling, structural information for coreTAP can be derived, indicating residues located within the translocation pathway (Figure 2).

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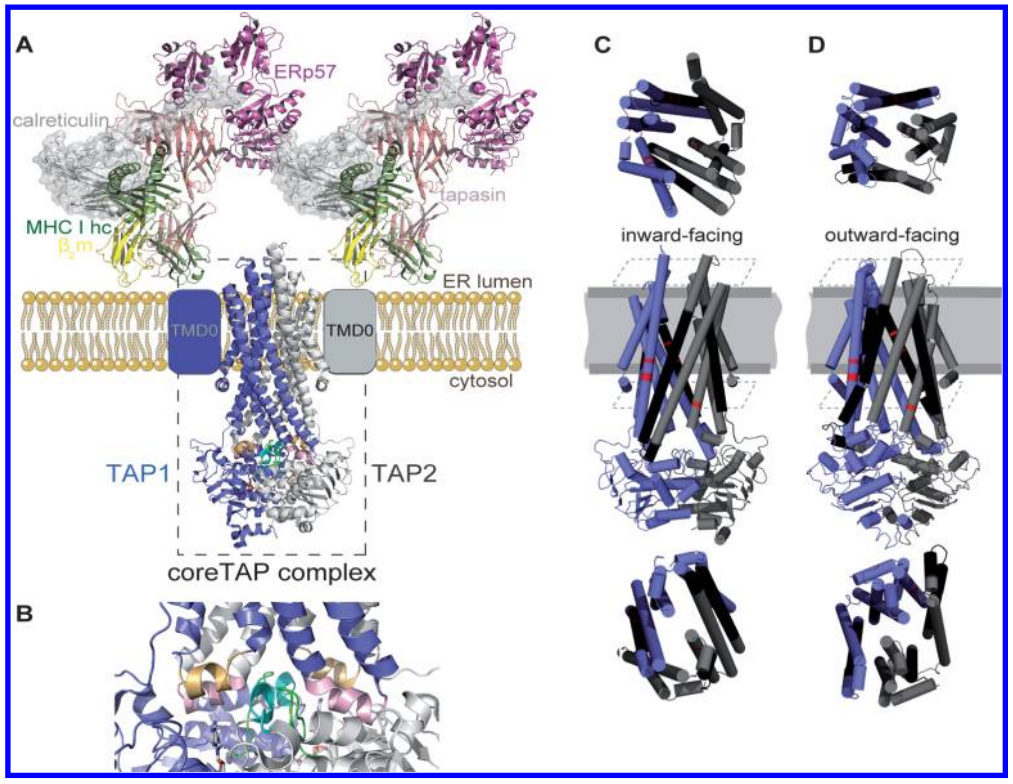

Figure 2. Structural organization of the peptide loading complex and the core TAP complex. (A) Cartoon representation of the components of the peptide-loading complex using a homology model of TAP based on the crystal structure of Sav1866 (PDB entry 2ONJ). The interactions are based on molecular docking simulations and indicate a potential macromolecular complex conformation. The color code is as follows: blue for TAP subunit 1, gray for TAP subunit 2, magenta for ERp57, green for the MHC I heavy chain, yellow for β_2-microglobulin, salmon for tapasin, and light gray for calreticulin. The N-terminal subdomains (TMD_0) of TAP are denoted with boxes. Structural elements involved in ATP hydrolysis and/or interdomain communication are color-coded within the coreTAP complex: green for the Q-loop, cyan for the X-loop, orange for coupling helix 1, and magenta for coupling helix 2. Two bound molecules of AMP-PNP are shown as balls and sticks. (B) Closeup of the interface region between the two NBDs. The color code is the same as in panel A. The coupling helices contact the Q- and X-loops of both NBDs. (C) Three-dimensional (3D) model of the coreTAP complex in the outward-facing conformation using Sav1866 (PDB entry 2ONJ) as a search model.[36,40] TAP1 and TAP2 are colored blue and gray, respectively. The peptide sensors are highlighted in black, and residues involved in peptide binding and transport are colored red. (D) 3D model of the coreTAP complex in the inward-facing conformation using MsbA (PDB entry 3B5X) as a search model.[37] Structural elements have the same color code as in panel B. 3D models are rotated by ±90° showing the translocation pathway from the lumenal (top) and the cytosolic (bottom) sides. Section views are indicated by dashed trapezoids. NBDs have been omitted for the sake of clarity.

In a unifying model of type I ABC importers and type III exporters, ATP binding and subsequent formation of a tight NBD dimer force the TMDs into an outward-facing ("catalytic") conformation, whereas ATP hydrolysis and disengagement of the NBD dimer reset the transporter in its inward-facing or "resting" conformation (Figure 3).

CONFORMATIONAL SWITCH BY ATP BINDING AND HYDROLYSIS

ABC proteins couple binding and hydrolysis of ATP with further enzymatic activities such as DNA repair and ribosome recycling, or with transport across membranes as in the case of ABC transporters.[43] The individual NBD, composed of a RecA fold and an α-helical subdomain,[44,45] binds ATP and ADP with similar affinities (0.1−0.5 mM), but has a very low ATPase activity. Upon ATP-induced dimerization, the helical domain rotates ~30° toward the RecA fold, arranging two NBDs in a head-to-tail orientation with two ATP molecules sandwiched between the NBDs.[46,47] This NBD dimer consists of two symmetric ATP-binding sites, which are aligned by Walker A/B motifs of one NBD and the ABC signature motif (C-loop) of the opposite NBD.[48,49]

The crystal structure of the vitamin B_{12} importer $BtuC_2D_2$ demonstrated that full-length ABC transporters have folds identical to that of the soluble NBD dimer.[33] Crystal structures of ABC transporters in either an inward-facing or outward-facing conformation visualize structural changes of the TMDs as a consequence of the catalytic state of the NBDs but are only snapshots of a complex allosteric mechanism, which may have distinct intermediates. A sequential hydrolysis of two ATP molecules during one cycle was demonstrated for mitochondrial transporter ABCB10 (Mdl1p) from *Saccharomyces cerevisiae*.[50]

Several ABC transporters of subfamilies B and C consist of two nonequivalent NBDs aligning one consensus (canonic) and one nonconsensus (noncanonic) ATP-binding site. It is unclear whether the noncanonical site is ATPase inactive or has a regulatory function with a largely reduced ATP hydrolysis rate as proposed for, e.g., TAP[51] and CFTR.[52] An allosteric coupling of the degenerate and the consensus site was recently shown for the heterodimeric TmrAB complex from *Thermus thermophilus*.[53]

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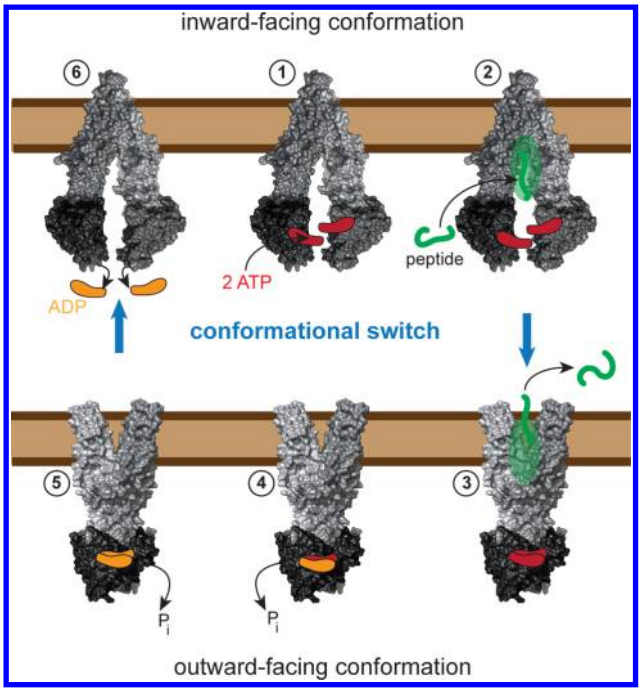

Figure 3. Model of the translocation cycle of TAP. Under physiological conditions, TAP is loaded with ATP (red) and rests in the inward-facing, so-called "open" or "receptive", conformation (1) represented by a surface model of P-gp (PDB entry 3G61). Peptide (green) binding induces a dimerization of the two NBDs sandwiching the two ATP molecules within the NBD interface (2). NBD dimerization induces a conformational switch of the ABC transporter that releases the peptide into the ER lumen (3) represented by a surface model of Sav1866 (PDB entry 2ONJ). The resulting solute free outward-facing conformation (4) is also termed the "closed" conformation. ATP is hydrolyzed, and inorganic phosphate (P_i) is released from the NBD sandwich (5). Unbinding of ADP (yellow) induces a conformational switch of TAP back into the inward-facing conformation (6). The NBD dimer opens and coordinates two ATP molecules for a new translocation cycle.

TUNNELING THROUGH THE MEMBRANE

Antigen processing is the result of an interplay among the immunoproteasome, TAP, and MHC I.[3,54,55] Peptides produced by the proteasomal degradation machinery display a large variability in sequence and length; the peptide-binding site of TAP must thus be quite promiscuous. Like multidrug transporters, TAP binds a wide range of solutes with relatively high (submicromolar) affinities. TAP nicely illustrates how nature has solved the problem of transporting a very broad spectrum of solutes using a common mechanism.

Binding of peptide to TAP is ATP-independent, but peptide translocation strictly requires ATP hydrolysis.[56−58] Notably, TAP does not exhibit a basal ATPase activity, although the NBDs are loaded with ATP under physiological conditions.[59] Peptide binding induces an allosteric cross-talk between the peptide-binding site in the TMDs and the NBDs and subsequently triggers peptide translocation and ATP hydrolysis.[60] Peptide association is a two-step process: a fast association phase followed by a peptide-induced slow structural reorganization of the transporter.[61] The heat capacity of TAP is temperature-dependent, influencing enthalpy and entropy of peptide binding. At low temperatures, peptide binding is exothermic, but it is endothermic at physiological temperature and thus driven by entropy. The increase in heat capacity reflects a structural rearrangement of almost one-fourth of all TAP residues.[62]

PEPTIDE SPECIFICITY

TAP is found in all higher (jawed) vertebrates; however, only forms of TAP from human, mouse, rat, and chicken have been characterized with respect to their peptide specificity.[20,63−66] TAP has a binding preference for peptides from 8 to 16 amino acids.[57] However, longer peptides of ≤40 amino acids can be translocated with greater selectivity[67] and reduced efficiency.[67,68] For rat TAP, the length restriction by the *cim*^{a} allele is more permissive than the rat *cim*^{b} allele and human TAP,[68] whereas the C-terminal sequence selection is similar between human and rat *cim*^{a} TAP, as well as mouse and rat *cim*^{b} TAP.[63,69]

Flanking residues of the peptide critically influence the efficiency of translocation. The C-terminal residue produced by proteasomal cleavage matches the restraints of both TAP and MHC I, but the N-termini of transported peptides often deviate from the sequence restraints of MHC I and thus require further proteolytic trimming in the lumen of the ER.[70,71] The C-terminal and three N-terminal residues have crucial effects on peptide translocation, and blocked N- or C-termini prevent peptide binding and transport.[56,72] Hydrophobic residues at position 3 are strongly favored by TAP, and position 2 must either be hydrophobic or charged. A negative effect on translocation has been assigned to aromatic or acidic residues at the very N-terminus and in particular to proline at position 1 or 2.[20,65,73] A specific demand on anchor residues as determined for binding of peptide to MHC I was not detected for human TAP,[65] although its preference for hydrophobic or basic residues at the C-terminus matches well with the properties of the binding groove of MHC I molecules.[20] For murine TAP, the peptide specificity at the N-terminus is much less pronounced; however, proline also has a negative effect at positions 1−3, whereas the C-terminus is critical as in human TAP with a similar specificity for hydrophobic (aliphatic or aromatic) residues.[73−75] The two rat TAP alleles, *cim*^{a} and *cim*^{b}, both prefer hydrophobic residues at the C-terminus, but the translocation of peptides with a basic C-terminus is unique to *cim*^{a}.[69]

A high degree of polymorphism is found in TAP of birds.[66,76,77] Each of the seven chicken MHC I haplotypes possesses different allelic variants of TAP1 and TAP2, leading to individual peptide specificities that are matched with the binding motif of the corresponding MHC I molecules. This might reflect the ancestral situation of the coevolution of TAP and MHC I.[66] The polymorphism of the TAP subunits is scattered throughout all of the genes, resulting in silent mutations to nonconservative amino acid exchanges in the putative peptide-binding region, the NBDs, and the tapasin-binding region (TMD_0). In contrast to, e.g., humans, one MHC I allele dominates all others in chicken, and the epitope diversity is most likely given by the allelic variations of TAP.

SUBSTRATE-BINDING POCKET OF TAP

The exact position and structural organization of the peptide-binding pocket are unknown, but biochemical studies have identified several residues involved in peptide binding (Figure 2). Residues within the cytosolic loops CL1 (Thr^{217} and Met^{218}) and CL2 (Ala^{374} and Arg^{380}) of coreTAP2 were shown to control the peptide repertoire.[78,79] CL2 and the linker region

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between TM6 and the NBD of coreTAP1 and coreTAP2 have been identified by peptide photo-cross-linking to be involved in peptide translocation.[80] Recently, two additional residues have been shown to act as a peptide sensor and/or conformational stabilizer: Val[288] within CL1 of coreTAP1 contacts the peptide as shown by disulfide cross-linking,[81] and Cys[213] (TAP2) is involved in the stabilization of the receptive conformation of TAP.[82] The structural flexibility of the binding pocket on TAP was addressed by site-specific spin labeling and electron paramagnetic resonance (EPR) studies of binding of peptides to TAP. In a manner independent of the length of the bound peptides, the distance from the N- and C-terminal residues is approximately 2.1 nm, leading to the conclusion that peptides bind to TAP like they do to MHC I in an extended kink conformation.[83]

VIRAL IMMUNE EVASION STRATEGIES

Antigen presentation is a fundamental principle in adaptive immunity for detecting and eliminating infected or malignant cells by CTLs. Because peptides derived from proteasomal degradation are the major source for loading of MHC I, TAP assumes an essential role in the delivery of these peptides from the cytosol to the lumen of the ER. Several viruses have evolved strategies for interfering with TAP function by inhibiting distinct steps in the translocation cycle, thereby blocking MHC I loading and antigen presentation.[84−86] On the molecular level, viral interference involves the inhibition of ATP binding or hydrolysis, peptide binding or translocation, degradation of TAP, or interference with the trafficking of MHC I molecules. Most of the inhibition of the MHC I antigen processing pathway has been characterized for members of the herpes virus family, but recently, proteins of the cowpox virus family have also been identified to interact with TAP[87−89] (Figure 4).

ICP47 encoded by herpes simplex virus (HSV type 1 and 2) was found as an inhibitor of peptide loading because MHC I molecules are retracted within the ER.[90,91] ICP47 competes with peptides for the binding pocket on TAP but does not interfere with ATP binding.[92,93] The active domain of ICP47 was mapped to cytosolic residues 2−34,[94,95] which adopts a helix−loop−helix motif upon binding to the membrane.[96−98] In contrast, human cytomegalovirus (HCMV) protein US6 inhibits ATP binding but does not influence peptide binding.[99−103] The ER-lumenal domain of US6 stabilizes TAP and prevents a conformational rearrangement necessary for peptide translocation.[103] US6 binds only to heterodimeric TAP1−TAP2 complexes but does not interact with nonassembled subunits. The inhibition of TAP by US6 involves several interactions between US6 and the two TAP subunits.[104] Upon incubation with soluble US6, cross-presentation is completely abrogated in dendritic cells, revealing the importance of TAP for cross-presentation.[4−6]

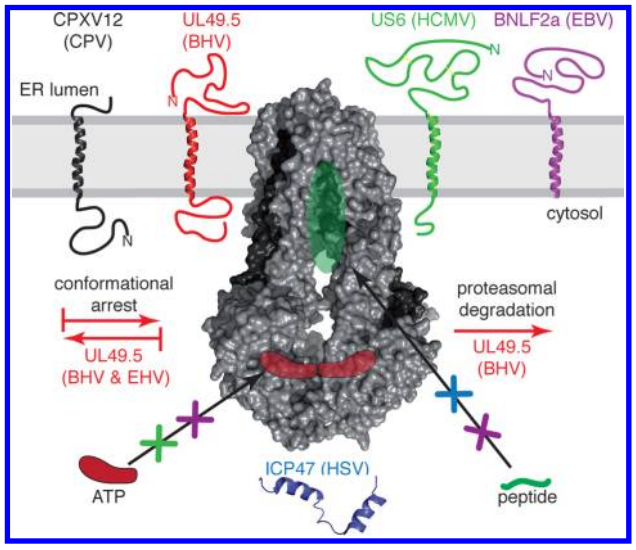

Figure 4. Viral factors that interfere with TAP function. ICP47 (HSV, blue) and tail-anchored BNLF2a (EBV, magenta) inhibit peptide binding. US6 (HCMV, green) and BNLF2a block ATP binding. UL49.5 (BHV & EHV, red) arrests TAP in a conformation incapable of peptide translocation; in addition, UL49.5 from BHV leads to the proteasomal degradation of TAP. The mechanism of inhibition of translocation by CPXV12 (CPV) is not known. The ATP and peptide binding sites of TAP are shaded in red and green, respectively.

Varicelloviruses have evolved sophisticated ways of inhibiting TAP. Forms of UL49.5 from various herpes viruses arrest TAP in various translocation incompetent conformations that, in case of bovine herpes virus-1 (BHV-1), lead to proteasomal degradation of TAP.[105,106] UL49.5 from BHV-1 inhibits binding of peptide to TAP by interacting with the core complex,[107] although it associates with glycoprotein M in later stages of infection.[108] In contrast, UL49.5 from equine herpes virus (EHV-1 and EHV-4) interferes with ATP binding.[109] Pseudorabies virus (PRV) UL49.5 does not affect ATP recruitment or TAP stability, although it arrests TAP in a translocation incompetent state.[109]

Epstein-Barr virus (EBV) protein BNLF2a competes with binding of peptide as well as ATP to TAP.[110−112] The tail-anchored BNLF2a targets the coreTAP complex, preventing a conformational switch, thus leaving TAP in a translocation incompetent state.[113] The recently discovered protein CPXV12 from cowpox virus (CPV) inhibits the translocation of peptides by binding to the peptide-loading complex and thereby interferes with loading of peptide onto empty MHC class I molecules.[87,88]

In summary, these viral factors represent very useful biochemical tools for characterizing different states of the translocation cycle of peptides from the cytosol to the ER lumen (Figure 4). Their combinatorial use will further explore mechanistic and structural details of the peptide translocation cycle of TAP.

OUTLOOK

Recent progress in the determination of the structure of ABC transporters has shed light on functional aspects of translocation of the solute across cellular membranes, including NBD dimerization and ATP hydrolysis, but important mechanistic details such as ATP:solute stoichiometry or the structural organization of the substrate-binding pocket remain unclear. TAP is well-characterized with regard to its physiological function and substrate specificity; it thus represents an ideal model for a heterodimeric ABC transporter for studying the mechanistic details mentioned above under important medical contexts, including infectious diseases and tumor development.

Although several mammalian ABC transport systems, including TAP, are well characterized, some very fundamental questions remain. Are these transporters indeed acting as a pump moving solutes against their gradient? Is this flux strictly unidirectional or mediated by a net bidirectional flow? In the case of TAP, how does the peptide concentration in the ER

lumen determine the peptide repertoire bound to MHC I molecules? Which concentration of peptides can be reached in the ER lumen, and how is this concentration counterbalanced with protein folding and quality control?

Because TAP is able to transport peptides with a length of up to ~40 amino acids, the conformational dynamics as well as the number of ATP molecules that are hydrolyzed during a translocation cycle are important issues to resolve. The architecture of the peptide-binding site will solve the problem of the broad solute specificity in TAP and many other ABC transporters. It will also demonstrate the correlation between the peptide-bound and occluded states during transport. Recently, the lipid environment of TAP has been characterized by mass spectrometry, and it was shown that some lipid components have a strong positive or negative effect on TAP function.[114] This study demonstrates the importance of determining the lipid environment of ABC transporters.

The assembly of the heterodimeric TAP complex calls into question the role of each subunit in the translocation cycle. The asymmetry of the two ATP-binding sites results in a noncanonical site that might serve in ATP hydrolysis and/or can have regulatory effects on ATP hydrolysis and NBD dimerization. The lack of detailed structural information about the TAP complex impedes the understanding of fundamental processes, such as peptide binding and translocation. This is why the structural characterization of TAP and other mammalian ABC transporters will be within the focus of many laboratories exploring the detailed mechanism of the translocation of the solute across membranes. Identifying new inhibitors (viral or endogenous) and deciphering their mode of action will help our understanding of the translocation mechanism of TAP.

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ABBREVIATIONS

CTL, cytotoxic T-lymphocyte; ER, endoplasmic reticulum; HCMV, human cytomegalovirus; HSV, herpes simplex virus; MHC I, major histocompatibility complex class I; NBD, nucleotide-binding domain; PDB, Protein Data Bank; PLC, peptide-loading complex; TAP, transporter associated with antigen processing; TM, transmembrane-spanning helix; TMD, transmembrane domain.

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