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A Functional-Phylogenetic Classification System for Transmembrane Solute Transporters

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"To know truly is to know by causes."

Francis Bacon

"To me life consists simply in this, in the fluctuation between two poles, in the hither and thither between the two foundation pillars of the world."

Herman Hesse

INTRODUCTION

Transport systems serve the cell in numerous capacities (118–123). First, they allow entry of all essential nutrients into the cytoplasmic compartment and subsequently into organelles, allowing metabolism of exogenous sources of carbon, nitrogen, sulfur, and phosphorus. Second, they provide a means for the regulation of metabolite concentrations by catalyzing the excretion of end products of metabolic pathways from organelles and cells. Third, they mediate the active extrusion of drugs and other toxic substances from either the cytoplasm or the plasma membrane. Fourth, they mediate uptake and efflux of ionic species that must be maintained at concentrations that differ drastically from those in the external milieu. The maintenance of conditions conducive to life requires a membrane potential, requisite ion concentration gradients, and appropriate cytoplasmic concentrations of all essential trace minerals that participate as cofactors in metabolic processes. Such conditions are required for the generation of bioelectricity as well as for the maintenance of enzymatic activities. Fifth, transporters participate in the secretion of proteins, complex carbohydrates, and lipids into and beyond the cytoplasmic membrane, and these macromolecules serve a variety of biologically important roles in protection against environmental insult and predation, in communication with members of the same and different species, and in pathogenesis. Sixth, transport systems allow the transfer of nucleic acids across cell membranes, allowing genetic exchange between organisms and thereby promoting species diversification. Seventh, transporters facilitate the uptake and release of pheromones, alarmones, hormones, neurotransmitters, and a variety of other signaling molecules that allow a cell to participate in the biological experience of multicellularity. Finally, transport proteins allow living organisms to conduct biological warfare, secreting, for example, antibiotics, antiviral agents, antifungal agents, and toxins of humans and other animals that may confer upon the organism producing such an agent a selective advantage for survival purposes. Many of these toxins are themselves channel-forming proteins or peptides that serve a cell-disruptive transport function. Thus, from a functional standpoint, the importance of molecular transport to all facets of life cannot be overestimated.

The importance of transport processes to biological systems was recognized more than half a century ago (43, 82). Thanks largely to concerted efforts on the part of Jacques Monod and his coworkers at the Pasteur Institute in Paris, who studied the mechanism of action of the *Escherichia coli* lactose permease, the involvement of specific carrier proteins in transport became established (22, 113). Since these early studies, tremendous progress has been made in understanding the molecular bases of transport phenomena, and the *E. coli* lactose permease has frequently been at the forefront (45, 60, 143). Initially, transport processes were characterized from physiological standpoints using intact cells. Cell "ghosts" in which the cytoplasmic contents had been released by osmotic shock proved useful, particularly as applied to human red blood cells and later to bacteria. Work with such systems provided detailed kinetic descriptions of transport processes, and by analogy with chemical reactions catalyzed by enzymes, the proteinaceous nature of all types of permeases became firmly established (reviewed by Kaback [58]).

With the advent of gene-sequencing technologies, the primary structures of permeases first became available. Hydrophobicity analyses of these sequences revealed the strikingly hydrophobic nature of various types of integral membrane transporters (19, 68, 70, 95). Current multidisciplinary approaches are slowly yielding three-dimensional structural information about transport systems. However, since only a few such systems have yielded to X-ray crystallographic analyses (see, for example, references 26, 140, and 142 as well as Table 21 below), we still base our views of solute transport on molecular models that provide reasonable pictures of transport systems and the processes they catalyze without providing absolute assurance of accuracy (45, 59, 143).

It is well recognized that any two proteins that can be shown to be homologous (i.e., that exhibit sufficient primary and/or secondary structural similarity to establish that they arose from a common evolutionary ancestor) will in general prove to exhibit strikingly similar three-dimensional structures (32), although a few exceptions have been noted (127). Furthermore, the degree of tertiary structural similarity correlates well with the degree of primary structural similarity. For this reason, phylogenetic analyses allow application of modeling techniques to a large number of related proteins and additionally allow reliable extrapolation from one protein member of a family of known structure to others of unknown structure. Thus, once three-dimensional structural data are available for any one family member, these data can be applied to all other members within limits dictated by their degrees of sequence similarity. The same cannot be assumed for members of two independently evolving families or for any two proteins for which common descent has not been established.

Similar arguments apply to mechanistic considerations. Thus, the mechanism of solute transport is likely to be similar for all members of a permease family, and variations on a specific mechanistic theme will be greatest when the sequence divergence is greatest. By contrast, for members of any two independently evolving permease families, the transport mechanisms may be strikingly different. Knowledge of these considerations allows unified mechanistic deductive approaches to be correctly applied to the largest numbers of transport systems, even when evidence is obtained piecemeal from the study of different systems.

The capacity to deduce and extrapolate structural and mechanistic information illustrates the value of phylogenetic data. However, another benefit that may result from the study of molecular phylogeny is to allow an understanding of the mechanistic restrictions that were imposed upon an evolving family due to architectural constraints. Specific architectural features may allow one family to diversify in function with respect to substrate specificity, substrate affinity, velocity of transport, polarity of transport, and even mechanism of energy coupling. By contrast, the architectural constraints imposed on a second family may not allow functional diversification. Knowledge of the architectural constraints imposed on a permease family provides a clear clue as to the reliability of functional predictions for uncharacterized but related gene products revealed, for example, by genome sequencing. Conversely, the functional diversity of the members of a permease family must be assumed to reflect architectural constraints, and thus phylogenetic and functional analyses lead to architectural predictions.

Finally, phylogenetic analyses provide valuable information about the evolutionary process itself. One can sometimes glean clues regarding the time of appearance of a family, the organ-

ismal type in which the family arose, and the pathway taken for the emergence of the family during evolutionary history. Occasionally, it is also possible to ascertain whether or not two distinct families arose independently of each other.

Over the past decade, my laboratory has devoted considerable effort to the phylogenetic characterization of permease families (118–120). This work has led us to formulate a novel classification system superficially similar to that implemented years ago for enzymes by the Enzyme Commission. The transporter classification (TC) system has been reviewed and recommended for adoption by a panel of experts chaired by A. Kotyk of the International Union of Biochemistry and Molecular Biology (IUBMB). In contrast to the Enzyme Commission, which based its classification system solely on function, we have chosen to classify permeases on the basis of both function and phylogeny. In this review, I describe our proposal, point out some of its strengths, and emphasize its flexibility for the future inclusion of yet-to-be-discovered transporters. We hope that the TC classification system will prove to be as useful as the enzyme classification system. Earlier treatises concerning the TC system and transport protein evolution have appeared (121–123, 127).

A detailed description of the TC system can be found on our World Wide Web site (http://www-biology.ucsd.edu/~msaier/transport/). This site will be continuously updated as new relevant physiological, biochemical, genetic, biophysical, and sequence data become available. Thanks to the participation of Andrei Lupas and the SmithKline-Beecham bioinformatics group (5), the TC system is being automated so that new sequences will automatically appear in multiple alignments and phylogenetic trees with minimal human intervention. The system will also provide a user-friendly search tool, called TransBase, so that the TC system can be readily accessed by keyword, TC number, gene name, protein name, sequence, and sequence motif. These advances will render the TC system increasingly accessible to the entire scientific community worldwide. In return, members of the scientific community are strongly encouraged to communicate novel findings and corrections to me by E-mail, phone, fax, or snail mail.

TRANSPORT NOMENCLATURE

Communication of concepts relevant to transmembrane transport phenomena generally depends upon the use of a uniform, well-defined and accepted, universally understood set of terms that can be used by the international community of scientists regardless of national origin or discipline of training. In this section I therefore present the terms currently in use in the field and mention which of these terms have been recommended for adoption by the TC panel of the IUBMB. It is anticipated that the acceptance of these terms will greatly facilitate the interchange of information by scientists and students of transport internationally.

Almost all transmembrane transport processes are mediated by integral membrane proteins, sometimes functioning in conjunction with extracytoplasmic receptors or receptor domains as well as with cytoplasmic energy-coupling and regulatory proteins or protein domains (51, 112, 130, 139). Each such complex of these proteins and/or protein domains is referred to as a transport system, transporter, porter, permease system, or permease. These are all equivalent terms that are in general use by members of the transport community. A permease (porter) is a protein or protein complex that catalyzes a vectorial reaction, irrespective of whether or not it also catalyzes a chemical or electron transfer reaction that drives the vectorial process. Thus, many transport systems can be thought of as catalytic proteins or protein complexes analogous to enzymes or enzyme complexes. By definition, transporters facilitate vectorial rather than, or in addition to, chemical reactions. The preferred terms for these transport systems are transporters or porter.

Permease-mediated transport can occur by any one of three distinct but related processes. First and simplest is facilitated, equilibrative, or protein-mediated diffusion, a process that is not coupled to metabolic energy and therefore cannot give rise to concentration gradients of the transported substrate across the membrane. Two primary modes of facilitated transport have been recognized in biological systems: channel type and carrier type (Fig. 1). In channel-type facilitated diffusion, the solute passes in a diffusion-limiting process from one side of the membrane to the other via a channel or pore that is lined by appropriately hydrophilic (for hydrophilic substrates), hydrophobic (for hydrophobic substrates), or amphipathic (for amphipathic substrates) amino acyl residue moieties of the constituent protein(s). The structures of several such channel proteins have now been examined and elucidated by X-ray crystallographic techniques (see below). In carrier-type facilitated diffusion, some part of the transporter is classically presumed to pass through the membrane together with the substrate (143, 151). Whether or not this presumption is correct is not known, as no classical carrier has yet yielded to the analytical tools of the X-ray crystallographer.

Carriers usually exhibit rates of transport that are several orders of magnitude lower than those of channels. Moreover, in contrast to most channels, they exhibit stereospecific substrate specificities. Although both channels and carriers may exhibit the phenomenon of saturation kinetics, this is a more common characteristic of carriers. Very few carriers have been shown to be capable of functioning by a channel-type mechanism, and the few that exhibit this capacity generally do so only after the protein has been modified, either by covalent or noncovalent ligand binding or by imposition of a large membrane potential. Moreover, while most channels are oligomeric complexes, many carriers can function as monomeric proteins. These observations led to the suggestion that channels and carriers are fundamentally, not superficially, different.

If energy expenditure is coupled to transmembrane solute translocation, then a system catalyzing facilitated diffusion can become an active transporter. Such a system is considered to be a primary active transporter if a primary source of energy (i.e., a chemical reaction, light absorption, or electron flow) is coupled to the process. It is considered to be a secondary active transporter if a secondary source of energy (i.e., an ion electrochemical gradient, termed the proton motive force [PMF] in the case of protons or the sodium motive force [SMF] in the case of sodium ions), generated at the expense of a primary energy source, is coupled to the process. The transport panel considered all of these terms to be acceptable.

Active transporters (or porters) can function by uniport, symport, or antiport. Uniporters (the preferred term), also called single-species transporters or facilitated diffusion carriers (the less-preferred terms), catalyze the transport of a single molecular species, and transport therefore occurs independently of the movement of other molecular species. Symporters (the preferred term), also classically called cotransporters, catalyze the transport of two or more molecular species in the same direction. The fact that a single point mutation in a symporter can convert a carrier into a uniporter (41, 62, 66, 75, 147) emphasizes the superficial distinction between these two types of carriers. Antiporters (the preferred term), also called countertransporters, exchange transporters, and exchangers, catalyze the exchange of one or more molecular species for another. Antiport processes can be subdivided into two cate-

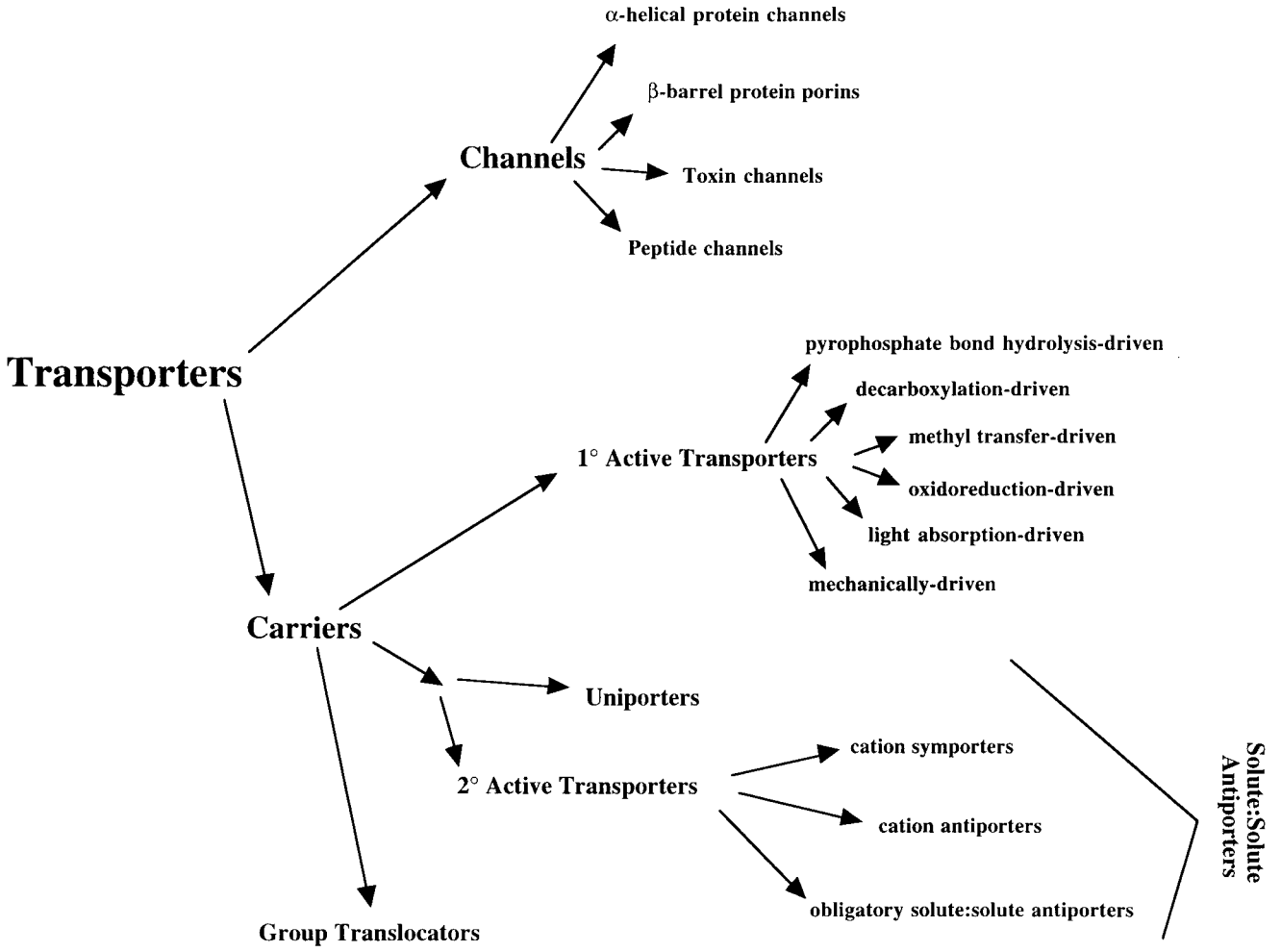

FIG. 1. Scheme illustrating the currently recognized primary types of transporters found in nature. These proteins are initially divided into channels and carriers. Channels are subdivided into α-helical protein channels, β-barrel protein porins (mostly in the outer membranes of gram-negative bacteria and eukaryotic organelles), toxin channels, and peptide channels. Carriers are subdivided into primary active carriers, secondary active carriers (including uniporters), and group translocators that modify their substrates during transport. Primary sources of chemical energy that can be coupled to transport include pyrophosphate bond (i.e., ATP) hydrolysis, decarboxylation, and methyl transfer. Oxidation-reduction reactions, light absorption, and mechanical devices can also be coupled to transport (see text). Secondary active transport is driven by ion and other solute (electro)chemical gradients created by primary active transport systems. The only well-established group-translocating system found in nature is the bacterial phosphoenolpyruvate:sugar PTS, which phosphorylates its sugar substrates during transport.

gories: antiport of like molecules (i.e., solute-solute antiport) and antiport of unlike molecules (i.e., solute-cation antiport). Many uniporters and symporters also catalyze solute-solute antiport, sometimes at rates that are substantially greater than those of uniport or symport. Some carriers catalyze solute-solute antiport at rates that exceed those of uniport or symport by 10^3- to 10^5-fold, and uniport via these carriers is of little or no physiological consequence (110). Such systems are said to be obligatory antiporters or exchangers.

Accelerative solute-solute antiport or countertransport has long been considered to be a diagnostic characteristic of carriers. Early transport kineticists concluded that its demonstration eliminated the possibility that a transporter functions by a channel-type mechanism and suggested that clear boundaries exist between carriers and channels (79, 135). Subsequent observations that certain "carriers" could apparently be converted into "channels" by chemical treatment (16, 17, 28, 29, 56), by imposition of large membrane potentials (131, 132, 149), or by ligand binding (13) led many students of transport to consider these boundaries indistinct. Our in silico phylogenetic and protein structural analyses suggest that these examples may be special cases and tend to reemphasize the importance of the channel-versus-carrier distinction (123, 127).

A few carriers modify their substrates during transport. The best-characterized such system is the bacterial phosphotransferase system (PTS), which phosphorylates its sugar substrates using phosphoenolpyruvate as the phosphoryl donor. Sugars taken up from the external milieu via the PTS are thus released into the cytoplasm as sugar-phosphates. Any process in which the substrate is modified during transport is termed group translocation. Although originally proposed in different form by Peter Mitchell as a general mechanism, its occurrence appears to be highly restricted in nature.

CONSIDERATIONS FOR THE SYSTEMATIC CLASSIFICATION OF TRANSMEMBRANE SOLUTE TRANSPORTERS

The introduction of Linnaeus of a universal classification system for living organisms allowed the rationalization of the

tremendous complexity of biological relationships into an evolutionary framework. Similarly, the introduction by the international Enzyme Commision of a universal enzyme classification system greatly increased our conception of the functional relationships of these proteins. Although protein-domain classification systems have been suggested, no comparable classification system has yet been proposed for proteins that catalyze vectorial reactions rather than (or in addition to) chemical reactions. In this section I describe the proposal for a universal system of classification for transporters based on both function and phylogeny.

As noted above, enzymes have long been classified in accordance with the directives and recommendations of the Enzyme Commission (31). The commission developed its directives decades ago, long before protein sequence data became available. Their system of classification was based solely on function. It was tacitly assumed that proteins of similar catalytic function would be closely related and that they therefore should be grouped together. We now know, however, that two different enzymes catalyzing exactly the same reaction sometimes exhibit completely different amino acid sequences and three-dimensional structures, function by entirely different mechanisms, and apparently evolved independently of each other, converging only with respect to the final reactions catalyzed. The enzyme classification system is thus limited in that it reflects only the reactions catalyzed by and the substrate specificities of the enzymes. It does not recognize the phylogenetic origins of these proteins and therefore does not reflect structural or mechanistic features.

As has been extensively documented, molecular phylogeny provides a reliable guide to protein structure and mechanism of action. It also provides an indication (albeit less definitive) of the specific process catalyzed and the substrate acted upon (127). Since the former characteristics are fundamental traits of a protein while the latter characteristics are more superficial traits, sometimes merely reflective of single amino acyl residue changes in a protein, it would be reasonable to suggest that as more and more sequence and phylogenetic data become available, these data should be used to provide the most reliable basis for protein classification. Since single amino acyl residue substitutions in permeases can give rise to different substrate-binding specificities (15, 23, 44, 94), these characteristics should be used in the final level of classification rather than in a primary level. We conclude that recognition of the evolutionary process provides a reliable guide to structure, mechanism, and function, although a few exceptions may exist (102, 127). If molecular phylogenetic studies can accurately retrace the evolutionary process, they should be used as a basis for any rational system of protein classification.

Some of the enzymes classified within the enzyme classification system are asymmetrically situated within an anisotropic, hydrophobic lipid membrane that separates two aqueous compartments. The resultant asymmetry allows these enzymes to catalyze vectorial as well as chemical modification reactions, as clearly enunciated decades ago by Peter Mitchell (83–85). Some of these integral membrane enzymes do, in fact, catalyze transmembrane transport of ions or other small solutes. However, most currently recognized solute permeases do not catalyze a chemical reaction and consequently are not included within the enzyme classification system. The comprehensive system of permease classification proposed here has the potential to encompass all types of transporters, both those that are currently recognized and those that are yet to be discovered.

TABLE 1. Classes and subclasses of transporters in the TC system[a]

1. Channels and pores
   - 1.A α-Type channels
   - 1.B β-Barrel porins
   - 1.C Pore-forming toxins (proteins and peptides)
   - 1.D Non-ribosomally synthesized channels

2. Electrochemical potential-driven transporters
   - 2.A Porters (uniporters, symporters, and antiporters)
   - 2.B Nonribosomally synthesized porters
   - 2.C Ion gradient-driven energizers

3. Primary active transporters
   - 3.A Diphosphate bond hydrolysis-driven transporters
   - 3.B Decarboxylation-driven transporters
   - 3.C Methyl transfer-driven transporters
   - 3.D Oxidoreduction-driven transporters
   - 3.E Light absorption-driven transporters

4. Group translocators
   - 4.A Phosphotransfer-driven group translocators

8. Accessory factors involved in transport
   - 8.A Auxiliary transport proteins

9. Incompletely characterized transport systems
   - 9.A Recognized transporters of unknown biochemical mechanism
   - 9.B Putative but uncharacterized transport proteins
   - 9.C Functionally characterized transporters lacking identified sequences

[a] This system of classification was approved by the transporter nomenclature panel of the International Union of Biochemistry and Molecular Biology in Geneva, 28–30 November 1999. No assignment has been made for categories 5 to 7. These will be reserved for novel types of transporters, yet to be discovered, that do not fall within categories 1 to 4.

THE TC SYSTEM

Early studies revealed that transport proteins could be grouped into families based exclusively on the degrees of similarity observed for their amino acid sequences (118). The significance of family assignment remained questionable until the study of internal gene duplications that had occurred during the evolution of some of these families established that these families had arisen independently of each other, at different times in evolutionary history, following different routes (119). In this section I will evaluate and utilize both function and molecular phylogeny for the purpose of conceptualizing transport protein characterization and classification (see also reference 120).

According to the proposed classification system, now recommended by the transport nomenclature panel of the IUBMB, transporters are grouped on the basis of five criteria, and each of these criteria corresponds to one of the five entries within the TC number for a particular permease. Thus, a permease-specific TC number has five components, V, W, X, Y, and Z. V corresponds to the transporter class, while W corresponds to the subclass (see Table 1). X specifies the permease family (or superfamily), while Y represents the subfamily in a family (or the family in a superfamily) in which a particular permease is found. Finally, Z delineates the substrate or range of substrates transported as well as the polarity of transport (in or out). Any two transport proteins in the same subfamily of a permease family that transport the same substrate(s) using the same mechanism are given the same TC number, regardless of whether they are orthologs (i.e., arose in distinct organisms by speciation) or paralogs (i.e., arose within a single organism by gene duplication). The mode of regulation proves not to cor-

relate with phylogeny and was probably superimposed on permeases late in the evolutionary process. Regulation is therefore not used as a basis for classification.

There are four recognized classes of transporters: channels, porters, primary active transporters, and group translocators (Table 1). Sequenced homologs of unknown function or mechanism and functionally characterized permeases for which sequence data are not available are included in a distinct class, class 9. Deficiencies in our knowledge will presumably be eliminated with time as more sequenced permeases become characterized biochemically and as sequences become available for the functionally but not molecularly characterized permeases. One additional class (class 8) is reserved for auxiliary transport proteins. It should be noted that each subclass of transporters has a two-digit TC number (V.W); each family has a three-digit TC number (V.W.X); each subfamily has a four-digit TC number (V.W.X.Y); and each permease type has a five-digit TC number (V.W.X.Y.Z).

As mentioned above, the primary level of classification in the TC system is based on mode of transport and energy-coupling source. The classes and subclasses of transporters currently recognized are listed below.

Category 1: Channels and Pores

1.A. α-Type channels. Transmembrane channel proteins of this class are ubiquitously found in the membranes of all types of organisms from bacteria to higher eukaryotes. These transporters usually catalyze the movement of solutes by an energy-independent process by passage through a transmembrane aqueous pore without evidence for a carrier-mediated mechanism. These channel proteins consist largely of α-helical spanners, although β-strands may be present and may even contribute to the channel. Outer membrane porin-type channel proteins are excluded from this class and are instead included in class 1.B.

1.B. β-Barrel porins. These proteins form transmembrane pores that usually allow the energy-independent passage of solutes across a membrane. The transmembrane portions of these proteins consist exclusively of β-strands that usually form β-barrels. Porin-type proteins are found in the outer membranes of gram-negative bacteria, mitochondria, plastids, and possibly acid-fast gram-positive bacteria.

1.C. Pore-forming toxins. These proteins and peptides are synthesized by one cell and secreted for insertion into the membrane of another cell, where they form transmembrane pores. They may exert their toxic effects by allowing the free flow of electrolytes and other small molecules across the membrane, or they may allow entry into the target cell cytoplasm of a toxin protein that ultimately kills or controls the cell. Both protein (large) and ribosomally synthesized peptide (small) toxins are included in this category.

1.D. Non-ribosomally synthesized channels. These molecules, often chains of L- and D-amino acids as well as other small molecular building blocks such as hydroxy acids (i.e., lactate and β-hydroxybutyrate), form oligomeric transmembrane ion channels. Voltage may induce channel formation by promoting assembly of the oligomeric transmembrane pore-forming structure. These "depsipeptides" are often made by bacteria and fungi as agents of biological warfare. Other substances, completely lacking amino acids, may also be capable of channel formation.

Category 2: Electrochemical Potential-Driven Porters

2.A. Porters (uniporters, symporters, and antiporters). Transport systems are included in this subclass if they utilize a carrier-mediated process to catalyze uniport (a single species is transported either by facilitated diffusion or in a membrane potential-dependent process if the solute is charged), antiport (two or more species are transported in opposite directions in a tightly coupled process, not coupled to a direct form of energy other than chemiosmotic energy), and/or symport (two or more species are transported together in the same direction in a tightly coupled process, not coupled to a direct form of energy other than chemiosmotic energy).

2.B. Non-ribosomally synthesized porters. These substances, like non-ribosomally synthesized channels, may be depsipeptides or non-peptide-like substances. Such a porter complexes a solute such as a cation in its hydrophilic interior and facilitates translocation of the complex across the membrane by exposing its hydrophobic exterior and moving from one side of the bilayer to the other. If the free porter can cross the membrane in the uncomplexed form, the transport process can be electrophoretic (the charged molecule moves down its electrochemical gradient), but if only the complex can cross the membrane, transport may be electroneutral, because one charged substrate is exchanged for another.

2.C. Ion gradient-driven energizers. Normally, outer membrane porins (1.B) of gram-negative bacteria catalyze passive transport of solutes across the membrane, but coupled to "energizers," they may accumulate their substrates in the periplasm against large concentration gradients. These energizers use the PMF across the cytoplasmic membrane, probably by allowing the electrophoretic transport of protons and conveying conformational change to the outer membrane receptors or porins. Homologous energizers drive bacterial flagellar motility (A. Lupas et al., unpublished results). The mechanism is poorly understood, but these energizers undoubtedly couple proton (H^+) or sodium (Na^+) fluxes through themselves in order to energize the process.

Category 3: Primary Active Transporters

These transporters use a primary source of energy to drive active transport of a solute against a concentration gradient. A secondary ion gradient is not considered a primary energy source because it is created by the expenditure of a primary energy source. Primary energy sources known to be coupled to transport are chemical, electrical, and solar.

3.A. Diphosphate bond hydrolysis-driven transporters. Transport systems are included in this subclass if they hydrolyze the diphosphate bond of inorganic pyrophosphate, ATP, or another nucleoside triphosphate to drive the active uptake and/or extrusion of a solute(s). The transport protein may or may not be transiently phosphorylated, but the substrate is not phosphorylated. These transporters are found universally in all living organisms.

3.B. Decarboxylation-driven transporters. Transport systems that drive solute (e.g., ion) uptake or extrusion by decarboxylation of a cytoplasmic substrate are included in this subclass. These transporters are currently thought to be restricted to prokaryotes.

3.C. Methyl transfer-driven transporters. A single characterized multisubunit protein family currently falls into this subclass, the Na^+-transporting methyltetrahydromethanopterin: coenzyme M methyltransferase. These transporter complexes are currently thought to be restricted to members of the *Archaea*.

3.D. Oxidoreduction-driven transporters. Transport systems that drive transport of a solute (e.g., an ion) energized by the exothermic flow of electrons from a reduced substrate to an oxidized substrate are included in this subclass. These trans-

porters are universal, although some families are restricted to one domain or another.

3.E. Light absorption-driven transporters. Transport systems that utilize light energy to drive transport of a solute (e.g., an ion) are included in this subclass. One family (fungal and archaeal rhodopsin) is found in archaea and eukaryotes, but the other (photosynthetic reaction center) is found only in bacteria and chloroplasts of eukaryotes.

Category 4: Group Translocators

4.A. Phosphotransfer-driven group translocators. Transport systems of the bacterial phosphoenolpyruvate:sugar PTS are included in this class. The product of the reaction, derived from extracellular sugar, is a cytoplasmic sugar-phosphate. No porters of the PTS have been identified in the archaeal or eukaryotic domain.

Category 8: Accessory Factors Involved in Transport

8.A. Auxiliary transport proteins. Proteins that in some way facilitate transport across one or more biological membranes but do not themselves participate directly in transport are included in this class. These proteins always function in conjunction with one or more established transport systems. They may provide a function connected with energy coupling to transport, play a structural role in complex formation, serve a biogenic or stability function, or function in regulation.

Category 9: Incompletely Characterized Transport Proteins

9.A. Transporters of unknown biochemical mechanism. Transport protein families of unknown classification are grouped in this subclass and will be classified elsewhere when the transport mode and energy-coupling mechanism have been characterized. These families include at least one member for which a transport function has been established, but either the mode of transport or the energy-coupling mechanism is not known.

9.B. Putative but uncharacterized transport proteins. Putative transport protein families are grouped in this subclass and will either be classified elsewhere when the transport function of a member becomes established or be eliminated from the TC system if the proposed transport function is disproven. These families include a member(s) for which a transport function has been suggested, but evidence for such a function is not yet compelling.

9.C. Functionally characterized transport proteins with unidentified sequences. Transporters of particular physiological significance will be included in this category even though a family assignment cannot be made. When their sequences are identified, they will be assigned to an established family. This is the only protein subclass that includes individual proteins rather than protein families.

FAMILIES OF TRANSPORTERS

The current index of transport system families is presented in Table 2. There are more than 250 entries, each of which usually describes a single family. Some of these families are actually large superfamilies with more than a thousand currently sequenced members (e.g., the voltage-gated ion channel (VIC) family (TC 1.A.1) (88); the major facilitator superfamily (MFS) (TC 2.A.1) (96, 125), and the ATP-binding cassette (ABC) superfamily (TC 3.A.1) (130, 139)). Others are very small families with only one or a few currently sequenced members. Most families, however, are currently of intermediate sizes, with between 5 and 500 sequenced members.

All of the families included in Table 2 will undoubtedly expand with time, and new families will be identified. The availability of new protein sequences will occasionally allow two or more currently recognized families to be placed together under a single TC number. In a few cases, two families are already known for which some evidence is available suggesting that they are related, e.g., the monovalent cation:proton antiporter-1 (CPA1) and CPA2 families (TC 2.A.36 and 2.A.37), the nucleobase-cation symporter-1 (NCS1) and NCS2 families (TC 2.A.39 and 2.A.40), as well as the L-lysine exporter, resistance to homoserine/threonine, and cadmium resistance families (TC 2.A.75, 2.A.76, and 2.A.77, respectively) (124, 148). Such evidence is usually based on limited sequence and/or sequence motif similarities, common function, and/or similar protein size, topology, and structure. When "missing link" sequences or three-dimensional structural data become available so that proteins of two families can be unequivocally grouped together within a single family, the lower TC number will be adopted for all of the family members, and the higher TC number will be abandoned.

The rigorous criteria used to delimit a family have been defined previously (121, 122). Briefly, in order for two proteins to belong to the same family, they must exhibit a region of 60 residues or more, in comparable portions of the two proteins, that have a comparison score in excess of 9 standard deviations (27). At this value, the probability that the degree of sequence similarity observed for these two proteins occurred by chance is less than 10^{-19} (25). It is considered that this degree of sequence similarity could not have arisen either by chance or by a convergent evolutionary process (32, 118). A minimum of 60 residues was arbitrarily selected because many protein domains in water-soluble proteins are of about this size.

The complete TC system is available on our web site (http://www-biology.ucsd.edu/~msaier/transport/), where the descriptions, primary references, and list of functionally characterized protein members of all families are provided. The whole-genome analysis data upon which this classification system was initially based are found on an included subportion of this web site, which was constructed under the guidance of Ian Paulsen (100, 101). This site will be updated continuously as new information becomes available. Anyone noting errors or incomplete listings is encouraged to contact me to provide the missing information and references.

As noted above, members of a transporter family generally utilize a single mode of transport and energy-coupling mechanism, thus justifying the use of these functional categories as the primary basis for classification. However, a few exceptions have been noted. First, the arsenite efflux permease (ArsAB; TC 3.A.4) of *E. coli* consists of two proteins, ArsA and ArsB. ArsB is an integral membrane protein that presumably provides the transport pathway for the extrusion of arsenite and antimonite (134, 153). ArsA is an ATPase that energizes ArsB-mediated transport. However, when ArsB alone is present, as in the case of the arsenical resistance pump of *Staphylococcus aureus*, transport is driven by the PMF (14). Expression of the *E. coli arsB* gene in the absence of the *arsA* gene similarly gives rise to PMF-driven transport. The presence or absence of the ArsA protein thus determines the mode of energy coupling.

The ArsB protein is a member of a large superfamily of ion transporters, the ion transporter superfamily, in which at least two families exhibit the unusual capacity of being able to incorporate auxiliary constituents that alter the transport characteristics of the carrier (107, 127). Such promiscuous use of energy is exceptionally rare and has been documented in only

TABLE 2. Complete index of families of transport proteins in the TC system[a]

TC no. (subclass)	Family
1.A. α-Type channels	
1.A.1	Voltage-gated ion channel (VIC) superfamily
1.A.2	Animal inward rectifier K^+ channel (IRK-C) family
1.A.3	Ryanodine-inositol 1,4,5-triphosphate receptor Ca^{2+} channel (RIR-CaC) family
1.A.4	Transient receptor potential Ca^{2+} channel (TRP-CC) family
1.A.5	Polycystine cation channel (PCC) family
1.A.6	Epithelial Na^+ channel (ENaC) family
1.A.7	ATP-gated cation channel (ACC) family
1.A.8	Major intrinsic protein (MIP) family
1.A.9	Ligand-gated ion channel (LIC) family of neurotransmitter receptors
1.A.10	Glutamate-gated ion channel (GIC) family of neurotransmitter receptors
1.A.11	Chloride channel (CIC) family
1.A.12	Organellar chloride channel (O-CIC) family
1.A.13	Epithelial chloride channel (E-CIC) family
1.A.14	Nonselective cation channel-1 (NSCC1) family
1.A.15	Nonselective cation channel-2 (NSCC2) family
1.A.16	Yeast stretch-activated, cation-selective, Ca^{2+} channel, Mid1 (Mid1) family
1.A.17	Chloroplast outer envelope solute channel (CSC) family
1.A.18	Chloroplast envelope anion channel-forming Tic110 (Tic110) family
1.A.19	Influenza virus matrix-2 channel (IVC) family
1.A.20	$gp91^{phox}$ phagocyte NADPH oxidase-associated cytochrome b_{558} (CybB) H^+ channel family
1.A.21	Bcl-2 (Bcl-2) family
1.A.22	Large conductance mechanosensitive ion channel (MscL) family
1.A.23	Small conductance mechanosensitive ion channel (MscS) family
1.A.24	Gap junction-forming connexin (connexin) family
1.A.25	Gap junction-forming innexin (innexin) family
1.A.26	Symbiotic ammonium transporter (SAT) family
1.A.27	Phospholemman (PLM) family
1.A.28	P21 holin S (P21 holin) family
1.A.29	λ holin S (λ holin) family
1.A.30	P2 holin TM (P2 holin) family
1.A.31	LydA holin (LydA holin) family
1.A.32	PRD1 holin M (PRD1 holin) family
1.A.33	T7 holin (T7 holin) family
1.A.34	HP1 holin (HP1 holin) family
1.A.35	T4 holin (T4 holin) family
1.A.36	T4 immunity holin (T4 immunity holin) family
1.A.37	ϕ29 holin (ϕ29 holin) family
1.A.38	ϕ11 holin (ϕ11 holin) family
1.A.39	ϕAdh holin (ϕAdh holin) family
1.A.40	ϕU53 holin (ϕU53 holin) family
1.A.41	LrgA holin (LrgA holin) family
1.A.42	ArpQ holin (ArpQ holin) family
1.A.43	Cph1 holin (Cph1 holin) family
1.A.44	Urea transporter (UT) family
1.A.45	H^+- or Na^+-translocating bacterial flagellar motor (Mot) family
1.B. β-Barrel porins	
1.B.1	General bacterial porin (GBP) family
1.B.2	Chlamydial porin (CP) family
1.B.3	Sugar porin (SP) family
1.B.4	*Brucella-Rhizobium* porin (BRP) family
1.B.5	*Pseudomonas* OprP porin (POP) family
1.B.6	OmpA-OmpF porin (OOP) family
1.B.7	*Rhodobacter* PorCa porin (RPP) family
1.B.8	Mitochondrial and plastid porin (MPP) family
1.B.9	FadL outer membrane protein (FadL) family
1.B.10	Nucleoside-specific channel-forming outer membrane porin (Tsx) family
1.B.11	Outer membrane fimbrial usher porin (FUP) family
1.B.12	Autotransporter (AT) family
1.B.13	Alginate export porin (AEP) family
1.B.14	Outer membrane receptor (OMR) family
1.B.15	Raffinose porin (RafY) family
1.B.16	Short-chain amide and urea porin (SAP) family
1.B.17	Outer membrane factor (OMF) family
1.B.18	Outer membrane auxiliary (OMA) protein family
1.B.19	Glucose-selective OprB porin (OprB) family
1.B.20	Bacterial toxin export channel (TEC) family
1.B.21	OmpG porin (OmpG) family

Continued on following page

TABLE 2—*Continued*

TC no. (subclass)	Family
1.B.22	Outer bacterial membrane secretin (secretin) family
1.B.23	Cyanobacterial porin (CBP) family
1.B.24	Mycobacterial porin (MBP) family
1.B.25	Outermembrane porin (Opr) family
1.B.26	Cyclodextrin porin (CDP) family
1.C. Pore-forming toxins	
1.C.1	Channel-forming colicin (colicin) family
1.C.2	Channel-forming δ-endotoxin insecticidal crystal protein (ICP) family
1.C.3	α-Hemolysin channel-forming toxin (αHL) family
1.C.4	Aerolysin channel-forming toxin (aerolysin) family
1.C.5	Channel-forming ε-toxin (ε-toxin) family
1.C.6	Yeast killer toxin K1 (YKT-K1) family
1.C.7	Diphtheria toxin (DT) family
1.C.8	Botulinum and tetanus toxin (BTT) family
1.C.9	Vacuolating cytotoxin (VacA) family
1.C.10	Pore-forming hemolysin E (HlyE) family
1.C.11	Pore-forming RTX toxin (RTX-toxin) family
1.C.12	Thiol-activated cytolysin (TAC) family
1.C.13	Channel-forming leukocidin cytotoxin (Ctx) family
1.C.14	Cytohemolysin (CHL) family
1.C.15	Whipworm stichosome porin (WSP) family
1.C.16	Magainin (magainin) family
1.C.17	Cecropin (cecropin) family
1.C.18	Melittin (melittin) family
1.C.19	Defensin (defensin) family
1.C.20	Nisin (nisin) family
1.C.21	Lacticin 481 (lacticin 481) family
1.C.22	Lactococcin A (lactococcin A) family
1.C.23	Lactocin S (lactocin S) family
1.C.24	Pediocin (pediocin) family
1.C.25	Lactococcin G (lactococcin G) family
1.C.26	Lactacin X (lactacin X) family
1.C.27	Divergicin A (divergicin A) family
1.C.28	AS-48 (AS-48) family
1.C.29	Plantaricin EF (plantaricin EF) family
1.C.30	Plantaricin JK (plantaricin JK) family
1.C.31	Channel-forming colicin V (colicin V) family
1.C.32	Amphipathic peptide mastoparan (mastoparan) family
1.C.33	Cathilicidin (cathilicidin) family
1.C.34	Tachyplesin (tachyplesin) family
1.C.35	Amoebapore (amoebapore) family
1.C.36	Bacterial type III-target cell pore (IIITCP) family
1.C.37	Lactococcin 972 (lactococcin 972) family
1.C.38	Pore-forming equinatoxin (equinatoxin) family
1.D. Non-ribosomally synthesized channels	
1.D.1	Gramicidin A (gramicidin A) channel family
1.D.2	Syringomycin channel-forming (syringomycin) family
1.D.3	Syringopeptin channel-forming (syringopeptin) family
1.D.4	Tolaasin channel-forming (tolassin) family
1.D.5	Alamethicin channel-forming (alamethicin) family
1.D.6	Complexed poly 3-hydroxybutyrate Ca^{2+} channel (cPHB-CC) family
2.A. Porters: uniporters, symporters, and antiporters	
2.A.1	Major facilitator superfamily (MFS)
2.A.1.1	Sugar Porter (SP) family
2.A.1.2	Drug:H^+ antiporter-1 (12 spanner) (DHA1) family
2.A.1.3	Drug:H^+ antiporter-2 (14 spanner) (DHA2) family
2.A.1.4	Organophosphate:P_i antiporter (OPA) family
2.A.1.5	Oligosaccharide:H^+ symporter (OHS) family
2.A.1.6	Metabolite:H^+ symporter (MHS) family
2.A.1.7	Fucose:H^+ symporter (FHS) family
2.A.1.8	Nitrate/nitrite porter (NNP) family
2.A.1.9	Phosphate:H^+ symporter (PHS) family
2.A.1.10	Nucleoside:H^+ symporter (NHS) family
2.A.1.11	Oxalate:formate antiporter (OFA) family
2.A.1.12	Sialate:H^+ symporter (SHS) family
2.A.1.13	Monocarboxylate porter (MCP) family

Continued on following page

TABLE 2—*Continued*

TC no. (subclass)	Family
2.A.1.14	Anion:cation symporter (ACS) family
2.A.1.15	Aromatic acid:H^+ symporter (AAHS) family
2.A.1.16	Siderophore-iron transporter (SIT) family
2.A.1.17	Cyanate permease (CP) family
2.A.1.18	Polyol permease (PP) family
2.A.1.19	Organic cation transporter (OCT) family
2.A.1.20	Sugar efflux transporter (SET) family
2.A.1.21	Drug:H^+ antiporter-3 (12 spanner) (DHA3) family
2.A.1.22	Vesicular neurotransmitter transporter (VNT) family
2.A.1.23	Conjugated bile salt transporter (BST) family
2.A.1.24	Unknown major facilitator-1 (UMF1) family
2.A.1.25	Peptide-acetyl-coenzyme A transporter (PAT) family
2.A.1.26	Unknown major facilitator-2 (UMF2) family
2.A.1.27	Phenyl propionate permease (PPP) family
2.A.1.28	Unknown major facilitator-3 (UMF3) family
2.A.1.29	Unknown major facilitator-4 (UMF4) family
2.A.2	Glycoside-pentoside-hexuronide (GPH):cation symporter family
2.A.3	Amino acid-polyamine-organocation (APC) superfamily
2.A.3.1	Amino acid transporter (AAT) family
2.A.3.2	Basic amino acid/polyamine antiporter (APA) family
2.A.3.3	Cationic amino acid transporter (CAT) family
2.A.3.4	Amino acid/choline transporter (ACT) family
2.A.3.5	Ethanolamine transporter (EAT) family
2.A.3.6	Archaeal/bacterial transporter (ABT) family
2.A.3.7	Glutamate:γ-aminobutyrate (GABA) antiporter (GGA) family
2.A.3.8	L-Type amino acid transporter (LAT) family
2.A.3.9	Spore germination protein (SGP) family
2.A.3.10	Yeast amino acid transporter (YAT) family
2.A.4	Cation diffusion facilitator (CDF) family
2.A.5	Zinc (Zn^{2+})-iron (Fe^{2+}) permease (ZIP) family
2.A.6	Resistance-nodulation-cell division (RND) superfamily
2.A.6.1	Heavy metal efflux (HME) family
2.A.6.2	(Largely gram-negative bacterial) hydrophobe/amphiphile efflux-1 (HAE1) family
2.A.6.3	Putative nodulation factor exporter (NFE) family
2.A.6.4	SecDF (SecDF) family
2.A.6.5	(Gram-positive bacterial) hydrophobe/amphiphile efflux-2 (HAE2) family
2.A.6.6	Eukaryotic (putative) sterol transporter (EST) family
2.A.6.7	(Largely archaeal putative) hydrophobe/amphiphile efflux-3 (HAE3) family
2.A.7	Small multidrug resistance (SMR) family
2.A.8	Gluconate:H^+ symporter (GntP) family
2.A.9	L-Rhamnose transporter (RhaT) family
2.A.10	2-Keto-3-deoxygluconate transporter (KDGT) family
2.A.11	Citrate-Mg^{2+}:H^+ (CitM)–citrate:H^+ (CitH) symporter (CitMHS) family
2.A.12	ATP:ADP antiporter (AAA) family
2.A.13	C_4-dicarboxylate uptake (Dcu) family
2.A.14	Lactate permease (LctP) family
2.A.15	Betaine/Carnitine/Choline transporter (BCCT) family
2.A.16	Telurite resistance/dicarboxylate transporter (TDT) family
2.A.17	Proton-dependent oligopeptide transporter (POT) family
2.A.18	Amino acid/auxin permease (AAAP) family
2.A.19	Ca^{2+}:cation antiporter (CaCA) family
2.A.20	Inorganic phosphate transporter (PiT) family
2.A.21	Solute:sodium symporter (SSS) family
2.A.22	Neurotransmitter:sodium symporter (NSS) family
2.A.23	Dicarboxylate/amino acid:cation (Na^+ or H^+) symporter (DAACS) family
2.A.24	Citrate:cation symporter (CCS) family
2.A.25	Alanine or glycine:cation symporter (AGCS) family
2.A.26	Branched-chain amino acid:cation symporter (LIVCS) family
2.A.27	Glutamate:Na^+ symporter (ESS) family
2.A.28	Bile acid:Na^+ symporter (BASS) family
2.A.29	Mitochondrial carrier (MC) family
2.A.30	Cation-chloride cotransporter (CCC) family
2.A.31	Anion Exchanger (AE) family
2.A.32	Silicon transporter (Sit) family
2.A.33	NhaA Na^+:H^+ antiporter (NhaA) family
2.A.34	NhaB Na^+:H^+ antiporter (NhaB) family
2.A.35	NhaC Na^+:H^+ antiporter (NhaC) family
2.A.36	Monovalent cation:proton antiporter-1 (CPA1) family

Continued on following page

TABLE 2—*Continued*

TC no. (subclass)	Family
2.A.37	Monovalent cation:proton antiporter-2 (CPA2) family
2.A.38	K^+ transporter (Trk) family
2.A.39	Nucleobase:cation symporter-1 (NCS1) family
2.A.40	Nucleobase:cation symporter-2 (NCS2) family
2.A.41	Concentrative nucleoside transporter (CNT) family
2.A.42	Hydroxyl/aromatic amino acid permease (HAAAP) family
2.A.43	Lysosomal cystine transporter (LCT) family
2.A.44	Formate-nitrite transporter (FNT) family
2.A.45	Arsenite-antimonite (ArsB) efflux family
2.A.46	Benzoate:H^+ symporter (BenE) family
2.A.47	Divalent anion:Na^+ symporter (DASS) family
2.A.48	Reduced folate carrier (RFC) family
2.A.49	Ammonium transporter (Amt) family
2.A.50	Triose-phosphate/nucleoside-sugar transporter (TP-NST) family
2.A.51	Chromate ion transporter (CHR) family
2.A.52	Ni^{2+}-Co^{2+} transporter (NiCoT) family
2.A.53	Sulfate permease (SulP) family
2.A.54	Mitochondrial tricarboxylate carrier (MTC) family
2.A.55	Metal ion (Mn^{2+}-ion) transporter (Nramp) family
2.A.56	Tripartite ATP-independent periplasmic transporter (TRAP-T) family
2.A.57	Equilibrative nucleoside transporter (ENT) family
2.A.58	Phosphate:Na^+ symporter (PNaS) family
2.A.59	Arsenical resistance-3 (ACR3) family
2.A.60	Organo anion transporter (OAT) family
2.A.61	C_4-dicarboxylate uptake C (DcuC) family
2.A.62	NhaD Na^+:H^+ antiporter (NhaD) family
2.A.63	Monovalent cation (K^+ or Na^+):proton antiporter-3 (CPA3) family
2.A.64	Type V secretory pathway or twin-arginine-targeting (Tat) family
2.A.65	Bilirubin transporter (BRT) family
2.A.66	Multi antimicrobial extrusion (MATE) family
2.A.67	Oligopeptide transporter (OPT) family
2.A.68	*p*-Aminobenzoyl-glutamate transporter (AbgT) family
2.A.69	Auxin efflux carrier (AEC) family
2.A.70	Malonate:Na^+ symporter (MSS) family
2.A.71	Folate-biopterin transporter (FBT) family
2.A.72	K^+ uptake permease (KUP) family
2.A.73	Inorganic carbon (HCO_3^-) transporter (ICT) family
2.A.74	4-TMS multidrug endosomal transporter (MET) family
2.A.75	L-Lysine exporter (LysE) family
2.A.76	Resistance to homoserine/threonine (RhtB) family
2.A.77	Cadmium resistance (CadD) family
2.A.78	Carboxylate/amino acid/amine transporter (CAAT) family
2.B Non-ribosomally synthesized porters	
2.B.1	Valinomycin carrier (valinomycin) family
2.B.2	Monensin (monensin) family
2.B.3	Nigericin (nigericin) family
2.B.4	Macrotetrolide antibiotic (MA) family
2.B.5	Macrocyclic polyether (MP) family
2.C Ion gradient-driven energizers	
2.C.1	TonB-ExbB-ExbD/TolA-TolQ-TolR (TonB) family of auxiliary proteins for energization of outer membrane receptor (OMR)-mediated active transport
3.A Diphosphate bond hydrolysis-driven transporters	
3.A.1	ATP-binding cassette (ABC) superfamily
ABC-type uptake permeases (All from prokaryotes [bacteria and archaea])	
3.A.1.1	Carbohydrate uptake transporter-1 (CUT1) family
3.A.1.2	Carbohydrate uptake transporter-2 (CUT2) family
3.A.1.3	Polar amino acid uptake transporter (PAAT) family
3.A.1.4	Hydrophobic amino acid uptake transporter (HAAT) family
3.A.1.5	Peptide/opine/nickel uptake transporter (PepT) family
3.A.1.6	Sulfate uptake transporter (SulT) family
3.A.1.7	Phosphate uptake transporter (PhoT) family
3.A.1.8	Molybdate uptake transporter (MolT) family
3.A.1.9	Phosphonate uptake transporter (PhnT) family
3.A.1.10	Ferric iron uptake transporter (FeT) family

Continued on following page

TABLE 2—*Continued*

TC no. (subclass)	Family
3.A.1.11	Polyamine/opine/phosphonate uptake transporter (POPT) family
3.A.1.12	Quaternary amine uptake transporter (QAT) family
3.A.1.13	Vitamin B_{12} uptake transporter ($VB_{12}T$) family
3.A.1.14	Iron chelate uptake transporter (FeCT) family
3.A.1.15	Manganese/zinc/iron chelate uptake transporter (MZT) family
3.A.1.16	Nitrate/nitrite/cyanate uptake transporter (NitT) family
3.A.1.17	Taurine uptake transporter (TauT) family
3.A.1.18	Putative cobalt uptake transporter (CoT) family
3.A.1.19	Thiamine uptake transporter (ThiT) family
3.A.1.20	*Brachyspira* iron transporter (BIT) family
	ABC-type efflux permeases (prokaryotic)
3.A.1.101	Capsular polysaccharide exporter (CPSE) family
3.A.1.102	Lipooligosaccharide exporter (LOSE) family
3.A.1.103	Lipopolysaccharide exporter (LPSE) family
3.A.1.104	Teichoic acid exporter (TAE) family
3.A.1.105	Drug exporter (DrugE1) family
3.A.1.106	Putative lipid A exporter (LipidE) family
3.A.1.107	Putative heme exporter (HemeE) family
3.A.1.108	β-Glucan exporter (GlucanE) family
3.A.1.109	Protein-1 exporter (Prot1E) family
3.A.1.110	Protein-2 exporter (Prot2E) family
3.A.1.111	Peptide-1 exporter (Pep1E) family
3.A.1.112	Peptide-2 exporter (Pep2E) family
3.A.1.113	Peptide-3 exporter (Pep3E) family
3.A.1.114	Probable glycolipid exporter (DevE) family
3.A.1.115	Na^+ exporter (NatE) family
3.A.1.116	Microcin B17 exporter (McbE) family
3.A.1.117	Drug exporter-2 (DrugE2) family
3.A.1.118	Microcin J25 exporter (McjD) family
3.A.1.119	Drug/siderophore exporter-3 (DrugE3) family
	ABC-type efflux permeases (mostly eukaryotic)
3.A.1.201	Multidrug resistance exporter (MDR) family
3.A.1.202	Cystic fibrosis transmembrane conductance exporter (CFTR) family
3.A.1.203	Peroxysomal fatty acyl coenzyme A transporter (FAT) family
3.A.1.204	Eye pigment precursor transporter (EPP) family
3.A.1.205	Pleiotropic drug resistance (PDR) family
3.A.1.206	**a**-Factor sex pheromone exporter (STE) family
3.A.1.207	Conjugate transporter-1 (CT1) family
3.A.1.208	Conjugate transporter-2 (CT2) family
3.A.1.209	Major histocompatibility complex peptide transporter (TAP) family
3.A.1.210	Heavy-metal transporter (HMT) family
3.A.2	H^+- or Na^+-translocating F-type, V-type, and A-type ATPase (F-ATPase) superfamily
3.A.3	P-type ATPase (P-ATPase) superfamily
3.A.4	Arsenite-antimonite (ArsAB) efflux family
3.A.5	Type II (general) secretory pathway (IISP) family
3.A.6	Type III (virulence-related) secretory pathway (IIISP) family
3.A.7	Type IV (conjugal DNA-protein transfer or VirB) secretory pathway (IVSP) family
3.A.8	Mitochondrial protein translocase (MPT) family
3.A.9	Chloroplast envelope protein translocase (CEPT or Tic-Toc) family
3.A.10	H^+-translocating pyrophosphatase (H^+-PPase) family
3.A.11	Bacterial competence-related DNA transformation transporter (DNA-T) family
3.B. Decarboxylation-driven active transporters	
3.B.1	Na^+-transporting carboxylic acid decarboxylase (NaT-DC) family
3.C. Methyl transfer-driven transporters	
3.C.1	Na^+-transporting methyltetrahydromethanopterin:coenzyme M methyltransferase (NaT-MMM) family
3.D. Oxidoreduction-driven active transporters	
3.D.1	Proton/sodium-translocating NADH dehydrogenase (NDH) family
3.D.2	Proton-translocating transhydrogenase (PTH) family
3.D.3	Proton-translocating quinol:cytochrome *c* reductase (QCR) superfamily
3.D.4	Proton-translocating cytochrome oxidase (COX) superfamily
3.D.5	Na^+-translocating NADH:quinone dehydrogenase (Na-NDH) family
3.D.6	Putative ion (H^+ or Na^+)-translocating NADH:ferredoxin oxidoreductase (NFO) family
3.D.7	H_2:heterodisulfide/oxidoreductase (HHO) family
3.D.8	Na^+- or H^+-pumping formyl methanofuran dehydrogenase (FMF-DH) family

Continued on following page

TABLE 2—*Continued*

TC no. (subclass)	Family
3.E. Light-driven active transporters	
3.E.1	Ion-translocating fungal/archaeal rhodopsin (FAR) family
3.E.2	Photosynthetic reaction center (PRC) family
4.A. Phosphoryl transfer-driven group translocators	
4.A.1	PTS glucose-glucoside (Glc) family
4.A.2	PTS fructose-mannitol (Fru) family
4.A.3	PTS lactose-*N*,*N*′-diacetylchitobiose-β-glucoside (Lac) family
4.A.4	PTS glucitol (Gut) family
4.A.5	PTS galactitol (Gat) family
4.A.6	PTS Mannose-fructose-sorbose (Man) family
8.A. Auxiliary transport proteins	
8.A.1	Membrane fusion protein (MFP) family
8.A.2	Secretin auxiliary lipoprotein (SAL) family
8.A.3	Cytoplasmic membrane-periplasmic auxiliary-1 (MPA1) protein with cytoplasmic (C) domain (MPA1−C or MPA1+C) family
8.A.4	Cytoplasmic membrane-periplasmic auxiliary-2 (MPA2) family
8.A.5	Voltage-gated K^+ channel β-subunit (VICβ) family
8.A.7	Phosphotransferase system enzyme I (EI) family
8.A.8	Phosphotransferase system HPr (HPr) family
8.A.9	rBAT transport accessory protein (rBAT) family
8.A.10	Slow voltage-gated K^+ channel accessory protein (MinK) family
8.A.11	Phospholamban (Ca^{2+}-ATPase regulator) (PLB) family
8.A.12	ABC bacteriocin exporter accessory protein (BEA) family
8.A.13	Tetratricopeptide repeat (Tpr1) family
9.A. Transporters of Unknown Classification	
9.A.1	Polysaccharide transporter (PST) family
9.A.2	MerTP mercuric ion (Hg^{2+}) permease (MerTP) family
9.A.3	MerC mercuric ion (Hg^{2+}) uptake (MerC) family
9.A.4	Nicotinamide mononucleotide (NMN) uptake permease (PnuC) family
9.A.5	Cytochrome oxidase biogenesis (Oxa1) family
9.A.6	Intracellular nucleoside transporter (INT) family
9.A.8	Ferrous iron uptake (FeoB) family
9.A.9	Low-affinity Fe^{2+} transporter (FeT) family
9.A.10	Oxidase-dependent Fe^{2+} transporter (OFeT) family
9.A.11	Copper transporter-1 (Ctr1) family
9.A.12	Copper transporter-2 (Ctr2) family
9.A.13	Short-chain fatty acid transporter (scFAT) family
9.A.14	Nuclear pore complex (NPC) family
9.A.15	Putative amide transporter (Ami) family
9.A.16	Septal DNA translocator (SDT) family
9.A.17	Metal ion transporter (MIT) family
9.A.18	Peptide uptake permease (PUP) family
9.A.19	Mg^{2+} transporter-E (MgtE) family
9.A.20	Low-affinity cation transporter (LCT) family
9.B. Putative uncharacterized transporters	
9.B.1	Metal homeostasis protein (MHP) family
9.B.2	Ca^{2+} homeostasis protein (CHP) family
9.B.3	Putative bacterial murein precursor exporter (MPE) family
9.B.4	Putative efflux transporter (PET) family
9.B.5	KX blood group antigen (KXA) family
9.B.6	Toxic Hok/Gef protein (Hok/Gef) family
9.B.7	Putative bacteriochlorophyll delivery (BCD) family
9.B.8	Canalicular bile acid transporter (C-BAT) family
9.B.9	Urate transporter (UAT) family
9.B.10	6TMS putative MarC transporter (MarC) family
9.B.11	Mitochondrial mRNA splicing-2 protein (MRS2) family
9.B.12	Stress (salt or low-temperature)-induced hydrophobic peptide (SHP) family
9.B.13	Putative pore-forming entericidin (ECN) family
9.B.14	Putative heme exporter protein (HEP) family
9.B.15	Putative chloroquine resistance Na^+/H^+ exchanger of *Plasmodium falciparum* (CQR) family
9.B.16	Putative ductin channel (ductin) family
9.B.17	Putative fatty acid transporter (FAT) family
9.B.18	SecDF-associated single transmembrane protein (SSTP) family
9.B.19	Mn^{2+} homeostasis protein (MnHP) family
9.B.20	Putative Mg^{2+} transporter-C (MgtC) family

Continued on following page

TABLE 2—*Continued*

TC no. (subclass)	Family
9.B.21	Frataxin (frataxin) family
9.B.22	Putative permease (PerM) family
9.B.23	Verapamil-reversible chloroquine resistance (VCR) family
9.B.24	Testis-enhanced gene transfer (TEGT) family
9.B.25	YbbM (YbbM) family
9.B.26	PF27 (PF27) family
9.B.27	YdjX-Z (YdjX-Z) family
9.B.28	YqaE (YqaE) family
9.B.29	YebN (YebN) family
9.B.30	Hly III (Hly III) family
9.B.31	YqiH (YqiH) family
9.C. Functionally characterized transporters with unidentified sequences	
9.C.1	Endosomal oligosaccharide transporters (EOT)
9.C.2	Volume-sensitive anion channels (VAC)
9.C.3	*Rhodococcus erythropolis* porin (REP) family

[a] The approved abbreviation for each family listed is given in parentheses.

a very few instances. When such an effect is reported, we shall usually classify the permease in accordance with the more complicated energy-coupling mechanism (in this case, as an ATP-driven primary active transporter [class 3] rather than as a secondary carrier [class 2]). However, in this unique case, the TC nomenclature panel of the IUBMB has recommended that a second family describing the PMF-driven ArsB homologs be included in the TC system (TC 2.A.45), as many ArsB homologs function by ATP-independent, ArsA-independent mechanisms.

Examples of secondary carrier families in which promiscuous transport modes have been reported include the mitochondrial carrier family (TC 2.A.29) and the triose phosphate/nucleotide sugar transporter (TP-NST) family (TC 2.A.50). Proteins of both families are apparently restricted to eukaryotic organelles. Members of these families normally catalyze carrier-mediated substrate-substrate antiport and are therefore classified as secondary carriers. However, treatment of mitochondrial carrier family members with chemical reagents, such as *N*-ethylmaleimide or Ca^{2+} (16, 17, 28, 29, 56), or imposition of a large membrane potential ($\Delta\Psi$) across a membrane into which a TP-NST family member has been incorporated (131, 132, 149), has been reported to convert these antiport-catalyzing carriers into anion-selective channels capable of functioning by uniport. Another secondary carrier that may be capable of exhibiting channel-like properties is the KefC protein of *E. coli* (13), which is a member of the CPA2 family (TC 2.A.37). "Tunneling" of ions and other solutes through carriers with little or no conformational change has been discussed (42). Again, the more complicated carrier-type mechanism, which appears to be relevant under most physiological conditions, provides the basis for classifying these proteins (i.e., as class 2 carriers rather than class 1 channels).

CHARACTERISTICS OF THE FAMILIES

Table 3 summarizes some of the key characteristics of most of the transporter families that we have identified. Categories 1.D and 2.B (non-ribosomally synthesized channels and carriers, respectively), 8 (auxiliary transport proteins), and 9.B and 9.C (putative but uncharacterized transporters) have been omitted (compare Table 1 with Table 3). Table 3 provides the family TC numbers, the abbreviations of the families, and the substrates of transporters included within each family. Substrates that are common to one transporter are separated by commas, while substrates of different transporters within the family are separated by semicolons. Thus, in the major intrinsic protein (MIP) family (TC 1.A.8), aquaporins generally transport water but not organic compounds, while glycerol facilitators generally transport short, straight-chain polyols but not water. A few members of the family may transport both (see reference 97 for a review). A recent report has provided evidence that a member of the MIP family can accommodate anions (154), but this observation is of uncertain physiological significance.

Table 3 also includes the size ranges of the individual protein members of the families and the numbers of (putative) transmembrane α-helical segments (TMSs) included within the permease polypeptide chains. All members of a family usually exhibit similar topological features, although several exceptions have been noted. When a homo- or heterooligomeric structure has been established for an intact permease, this fact is also indicated. Finally, the kingdoms in which members of the family have been identified, the approximate number of members that have been identified in each family, and representative examples of well-characterized members are also provided. The table is largely self-explanatory, but detailed information as well as primary and secondary references are provided on our web site and may be available in book form in the near future (Saier et al., unpublished data).

CROSS-REFERENCING PERMEASES BY ACCESSION NUMBER

Protein accession numbers can generally be used to find protein sequences of any sequenced protein referred to in the TC system. An accession number never changes once entered into a database. It therefore provides a quick and easy means of identifying a specific protein sequence. Moreover, it allows access to the database description of the sequenced protein, including structural, topological, and functional information. SwissProt (SP) database entries provide the most detailed information about the proteins, and SwissProt accession numbers are therefore provided when available. When not available, other accession numbers will be provided.

The accession numbers of all representative transport proteins included in the tables of the current TC system can be found on our web site. Accession numbers usually consist of

TABLE 3. Properties of families of transport systems included in the TC system

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
1.A. α-Type channel-forming proteins and peptides							
1.A.1	VIC	Na^+; K^+; Ca^{2+}; multiple cations	Widely varied	$(2)_4$; $(4)_2$; $(6)_4$; $(8)_2$; $(12)_2$; $(24)_1$; often hetero-oligomeric	E, A, B	>500	Voltage-gated Na^+ channels; voltage-gated Ca^{2+} channels; K^+ channels sensitive to voltage, Ca^{2+}, or cyclic nucleotides of *Homo sapiens*
1.A.2	IRK-C	K^+	390–430	$(2)_n$	E (An)	>10	Inward rectifier K^+ channels (ATP activated or G-protein regulated) of *Homo sapiens*
1.A.3	RIR-CaC	Ca^{2+}	5,000 or 2,500	$(6)_n$	E (An)	>10	Ryanodine receptor Ca^{2+} channels; inositol 1,4,5-triphosphate receptor Ca^{2+} channels of *Homo sapiens*
1.A.4	TRP-CC	Ca^{2+}	800; 1,300	$(6)_n$	E (An)	>10	Transient receptor potential Ca^{2+} channel (TRP) of *Drosophila melanogaster*
1.A.5	PCC	Na^+, K^+, Ca^{2+}	4,000	16–18	E (An)	3	Polycystin 1 of *Homo sapiens*
1.A.6	ENaC	Na^+; cations	640–700	$(2)_3$	E (An)	>20	Epithelial Na^+ channels; degenerins; peptide-gated ionotropic receptors of animals
1.A.7	ACC	Cations (monovalent cations; Ca^{2+})	380–600	$(2)_n$	E (An)	10	ATP-gated cation channel ($P2X_1$) of *Homo sapiens*
1.A.8	MIP	H_2O; glycerol, urea, polyols, NH_3, CO_2	220–310	$(6)_{2\text{ or }4}$	B, A, E	>100	Aquaporins (Aqp1) of *Homo sapiens*; glycerol facilitators (GlpF) of *E. coli*
1.A.9	LIC	Cations or chloride	400–500	$(3–5)_5$; often heterooligomeric	E (An)	>100	Acetylcholine or serotonin-activated cation channels; glycine, glutamate or GABA-regulated Cl^- channels of *Homo sapiens*
1.A.10	GIC	Monovalent cations and Ca^{2+}	800–1,000	$(3–5)_5$	E (An)	10	Glutamate-regulated ionotropic channels of *Rattus norvegicus*
1.A.11	ClC	Cl^-, anions	400–1,000	10–12	B, A, E	>30	Voltage-gated Cl^- channel (ClCl) of *Homo sapiens*
1.A.12	O-ClC	Cl^-, anions	240–440	$(2)_n$	E (An)	>10	Organellar voltage-sensitive Cl^- channels of *Bos taurus*
1.A.13	E-ClC	Cl^-, anions	900–1,000	$(4)_n$	E (An), B	>20	Ca^{2+}-activated Cl^- channel-2 of *Homo sapiens*
1.A.14	NSCC1	Monovalent cations: Na^+, Li^+, K^+	423	$(4)_n$	E (An)	1	Nonselective cation channel (NSC1) of *Mus musculus*
1.A.15	NSCC2	Monovalent cations: Na^+, K^+, Cs^+ as well as Ca^{2+} (slow)	283–402	$(2)_n$	E (An, Y, F)	>10	Nonspecific channel translocation protein-1 (NS1) of *Homo sapiens*; Sec62 of *Saccharomyces cerevisiae*
1.A.16	Mid1	Ca^{2+}	540	1 or 2	E (Y)	2	Mid1 of *Saccharomyces cerevisiae*
1.A.17	CSC	Ions; solutes	177	β-Structure?	E (Pl)	1	Chloroplast outer envelope solute channel (CSC) of *Pisum sativum*
1.A.18	Tic110	Anions; proteins	1,000	2	E (Pl)	1	Protein import-related anion-selective channel (Tic110)
1.A.19	IVC	H^+	~100	$(1)_n$ (n = 4?)	E (influenza virus)	1	Matrix protein (M2) of influenza virus
1.A.20	CybB	H^+	740	6	E (Pl, An)	~20	$gp91^{phox}$ human phagocyte NADPH oxidase-associated cytochrome b_{558} H^+ channel

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TABLE 3—*Continued*

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
1.A.22	MscL	Proteins, ions (slightly cation selective)	130–140	$(2)_6$	B	10	Large mechanosensitive ion channels (MscL) of *E. coli*
1.A.23	MscS	Ions (slight anion selectivity)	240–1,120	2–14	B, A	~50	Small conductance mechanosensitive ion channel (KefA) of *E. coli*
1.A.24	Connexin	Small molecules (no discrimination)	200–600	$(4)_n$	E (An)	>50	Vertebrate connexin 43 (gap junction α-1 protein) (CX43) of *Rattus norvegicus*
1.A.25	Innexin	Small molecules (no discrimination)	300–600	$(4)_n$	E (An)	>50	Invertebrate innexin (UNC-7) of *Caenorhabditis elegans*
1.A.26	SAT	NH_4^+	346	$(1)_n$	E (Pl)	1	Symbiotic NH_4^+ transporter-1 (SAT1) of *Glycine max*
1.A.27	PLM	Cl^- (anion selective), taurine, lactate, glutamate, isethionate, gluconate	70–100	$(1)_n$	E (An)	>10	Phospholemman; Cl^- conductance inducer protein Mat-8
1.A.28–1.A.43	Holin functional superfamily (16 families)	Proteins; small molecules	70–150	$(2–4)_n$	B, phage, plasmids	>50	Lysis protein S of phage lambda
1.A.44	UT	Urea, water	380–400	10	E (An)	>10	Kidney vasopressin-regulated urea transporter (UT2)
1.A.45	Mot	H^+; Na^+	500–1,000 (2 subunits)	4 (A) + 1 (B)	B	~10	H^+ uptake-driven flagellar motor (MotAB) of *E. coli*
1.B. Outer membrane porins (β-structure)							
1.B.1–1.B.26[e]	Various outer membrane porins and export proteins		250–1,000	8–24 β-strands	G-B; phage; E (mito, plastids)	>200	OmpF of *E. coli*; VDAC of *Bos taurus*; AidA of *E. coli*
1.C. Toxins							
1.C.1	Colicin	Ions; small molecules	500–700 (150–180 for the channel domain)	$(4?)_n$	B, plasmids	10	Colicin E1 of *E. coli*
1.C.2	IPC	Ions; small molecules	500–1,300 (~220 for the channel domain)	$(6?)_n$	B	>50	Cry3A insecticidal γ-endotoxin of *Bacillus thuringiensis*
1.C.3	αHL	Ions; small molecules	300–400	$(2β)_7$	B	>10	α-Hemolysin of *Staphylococcus aureus*
1.C.4	Aerolysin	Ions; small molecules	440–490	$(2β)_7$	B, E (Pl)	7	Aerolysin of *Aeromonas hydrophila*
1.C.5	ε-Toxin	Ions; small molecules	330	?	B	~10	ε-Toxin of *Clostridium perfringens*
1.C.6	YKT-K1	Cation selective	300	?	E (Y)	1	Yeast killer toxin of *Saccharomyces cerevisiae*
1.C.7	DT	DT, A-chain (protein)	340	?	B	1	Diphtheria toxin (DT) of corynebacteriophage β
1.C.8	BTT	BTT, L-chains	800	$(2?)_n$	B	>10	Botulinum and tetanus toxin channels of *Clostridium* species
1.C.9	VacA	Ions; small molecules	1,290	$(?)_6$	B	1	VacA of *Helicobacter pylori*
1.A.21	Bcl-2	Protein (cytochrome *c*)	200–240	$(1–5)_n$ (n = 2?)	E (An)	>20	Apoptosis regulator [Bcl-X(L)] of *Homo sapiens*
1.C.10	HlyE	Ions/moderately cation selective	305	?	B	1	HlyE of *E. coli*
1.C.11	RTX-toxin	Ions; small molecules	900–1,100	?	B	>10	Hemolysin A (HlyA) of *E. coli*

Continued on following page

TABLE 3—*Continued*

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
1.C.12	TAC	Ions; small molecules	470–580	?	B	>50	Perfringolysin O of *Clostridium perfringens*
1.C.13	Ctx	Ions	286	?	B	1	Leucocidin cytotoxin (Ctx) of *Pseudomonas aeruginosa* phage phiCTX
1.C.14	CHL	Ions and other solutes	500–800	?	B	~10	Cytohemolysin (HlyA) of *Vibrio cholerae*
1.C.15	WSP	Small molecules	400	?	An	2	Whipworm stichosome porin of *Tichuris trichiura*
1.C.16–1.C.30	Channel-forming peptide functional superfamily (15 families)	Ions	25–75 small peptides, often from large precursors	$(1 \text{ or } 2)_n$	B, A, E	>1,000	Maganins; cecropins; melittin, defensins; bacteriocins
1.C.31	Colicin V	Ions; small molecules	100	$(1–2)_n$	B (plasmid)	1	Colicin V of *E. coli*
1.C.32	Mastoparan	Small molecules	13–14	$(1)_n$	E (insects)	>10	Mastoparan of *Vespa xanthoptera*
1.C.33	Cathilicidin	Small molecules	90–220 (precursor), 18 (mature)	$(1)_n$	E (An)	>10	premyeloid cathilicidin 1 of *Equus caballus*
1.C.34	Tachyplesin	Small molecules	18–77	$(1)_n$	E (An)	>10	Tachyplesin I of *Tachyplesus tridentatus*
1.C.35	Amoebapore	Small molecules	90–100	$(1)_n$	E (Pr)	>10	Amoebapore of *Entamoeba histolytica*
1.C.36	IIITCP	Proteins	300–600	$(2)_n$	B	>20	Type III protein secretion target cell pore protein (YopB) of *Yersinia pseudotuberculosis*
1.C.37	Lactococcin 972	Small molecules	60–90	$(1)_n$	B	1	Lactococcin 972 of *Lactococcus lactis*
1.C.38	Equinatoxin	Small molecules	170–220	$(1)_{3 \text{ or } 4}$	An (sea urchins)	>10	Equinatoxin of *Actinia tenebrosa*
2.A. Carrier-type facilitators							
2.A.1	MFS	Various small molecules	400–600	12 or 14	B, A, E	>1,000	Lactose permease (LacY) of *E. coli*; drug efflux permease (EmrD) of *E. coli*
2.A.2	GPH	Sugars	~500	12	B, A, E (An, Pl)	>20	Melibiose permease (MelB) of *E. coli*
2.A.3	APC	Amino acids, polyamines, choline	440–630	12	B, A, E	>100	Lysine permease (LysP) of *E. coli*
2.A.4	CDF	Cd^{2+}, Co^{2+}, Ni^{2+}	300–750	6	B, A, E	>10	Heavy-metal uptake and efflux permeases of bacteria, eukaryotic plasma membranes, and mitochondria (CzcD of *Ralstonia eutropha*)
2.A.5	ZIP	Zn^{2+}, Fe^{2+}	220–430	8	E (Y, Pl, An)	>10	Zinc uptake transporter (Zrt1) of *Saccharomyces cerevisiae*
2.A.6	RND	Heavy metal ions; multiple drugs; oligosaccharides; organic solvents, fatty acids, phospholipids; cholesterol	800–1,200	~12	B, A, E	>100	Drug efflux pump (AcrA) of *E. coli*
2.A.7	SMR	Multiple drugs and dyes (mostly cationic)	110	$(4)_3$	B	>20	Cationic drug efflux pump (Smr) of *Staphylococcus aureus*
2.A.8	GntP	Gluconate, iodonate	450	12–14	B	>10	Gluconate permease (GntP) of *Bacillus subtilis*
2.A.9	RhaT	Sugars	280–340	10	B	5	Rhamnose transporter (RhaT) of *E. coli*

Continued on following page

TABLE 3—*Continued*

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
2.A.10	KDGT	2-Keto-3-deoxygluconate	400	10–12	B	5	2-Keto-3-deoxygluconate transporter (KdgT) of *Erwinia chrysanthemi*
2.A.11	CitMHS	Citrate	400	12	B	5	Citrate:Me^{2+}:H^+ symporter (CitM) of *Bacillus subtilis*
2.A.12	AAA	ATP, ADP	500	12	B, E (Pl)	>10	ATP/ADP exchange translocase of *Rickettsia prowazekii*
2.A.13	Dcu	C_4-dicarboxylates	440	12	G−B	5	Dicarboxylate uptake porter-A (DcuA) of *E. coli*
2.A.14	LctP	Lactate	510–516	12	B, A	10	Lactate permease (LctP) of *E. coli*
2.A.15	BCCT	Glycine betaine; carnitine; choline	480–680	12	B	10	Carnitine transporter (CaiT) of *E. coli*
2.A.16	TDT	Tellurite; dicarboxylates	320–440	10	B, A, E	>10	Tellurite uptake permease (TehA) of *E. coli*
2.A.17	POT	Peptides; nitrates; amino acids	450–600	12	B, E	>30	Dipeptide transporter (DtpT) of *Lactococcus lactis*
2.A.18	AAAP	Amino acids and their derivatives	400–710	11	E (An, Pl, Y, F)	>30	Amino acid/auxin:H^+ symporter (Aux-1) of *Arabidopsis thaliana*
2.A.19	CaCA	Ca^{2+}	460–1,200	10–12	B, A, E	>30	Ca^{2+}:H^+ antiporter (ChaA) of *E. coli*
2.A.20	PiT	Inorganic phosphate	410–680	10–12	B, A, E	>20	Phosphate transporter (PitA) of *E. coli*
2.A.21	SSS	Sugars; amino acids; vitamins; nucleosides; inositols; iodide; urea	400–700	12–15	B, A, E	>30	Pantothenate:Na^+ symporter (PanF) of *E. coli*
2.A.22	NSS	Neurotransmitters; amino acids; osmolytes; taurine; creatine	600–700	12	B, A, E (An)	>50	Serotonin:Na^+ symporter of *Homo sapiens*
2.A.23	DAACS	C_4-dicarboxylates; acidic and neutral amino acids	420–580	10–12	B, A, E	>20	Glutamate/aspartate permease (GltP) of *E. coli*
2.A.24	CCS	Mono-, di-, and tricarboxylates	450	12	B	>10	Citrate:Na^+ symporter (CitS) of *Klebsiella pneumoniae*
2.A.25	AGCS	Alanine, glycine	440–540	8–12	B	10	Alanine/glycine transporter (DagA) of *Alteromonas haloplanktis*
2.A.26	LIVCS	Branched-chain amino acid	~440	12	B	10	Branched-chain amino acid transporter (BraB) of *Pseudomonas aeruginosa*
2.A.27	ESS	Glutamate	~400	12	B	5	Glutamate:Na^+ symporter (GltS) of *E. coli*
2.A.28	BASS	Bile acids	360–480	12	B, E (An)	10	Bile acid uptake system of *Rattus norvegicus*
2.A.29	MC	ATP/ADP; P_i; organic anions; H^+; carnitine/acyl carnitine; basic amino acids; FAD	300	6	E (mito)	>100	ATP/ADP exchanger of *Homo sapiens*
2.A.30	CCC	K^+, Na^+, Cl^-	1,000–1,200	12	B, A, E (An, Pl, Y)	>30	NaCl/KCl cotransporter of *Rattus norvegicus*
2.A.31	AE	Inorganic anions	900–1,250	14	E (An, Y)	>20	Anion exchanger (AE1) of *Homo sapiens*
2.A.32	Sit	Silicate	550	12	E (diatoms)	6	Sit1 of *Cylindrotheca fusiformis*
2.A.33	NhaA	Na^+/H^+	~400	12	B	10	Na^+:H^+ antiporter (NhaA) of *E. coli*
2.A.34	NhaB	Na^+/H^+	~520	12	B	5	Na^+:H^+ antiporter (NhaB) of *E. coli*
2.A.35	NhaC	Na^+/H^+	~460	12	B	5	Na^+:H^+ antiporter (NhaC) of *Bacillus firmus*

Continued on following page

TABLE 3—*Continued*

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
2.A.36	CPA1	Na^+/H^+; Na^+ or K^+/H^+	500–900	10–12	B, A, E	>40	Na^+:H^+ antiporter (Nhe-1) of *Rattus norvegicus*
2.A.37	CPA2	Na^+/H^+ or K^+/H^+	330–630	10–12	B, A, E	>20	K^+ efflux protein-C (KefC) of *E. coli*
2.A.38	Trk	K^+:H^+?	420–560	10–12	B, A, E	>10	K^+ uptake permease (TrkH) of *E. coli*
2.A.39	NCS1	Nucleobases; thiamine; nucleosides	420–640	12	B, A, E	>20	Cytosine permease (CodB) of *E. coli*
2.A.40	NCS2	Nucleobases; urate	420–600	12	B, A, E	>20	Uracil permease (UraA) of *E. coli*
2.A.41	CNT	Nucleosides	390–660	12–14	B, A, E	>20	Nucleoside:H^+ symporter (NupC) of *E. coli*
2.A.42	HAAAP	Hydroxy and aromatic amino acids	400–450	11	B	>20	Tyrosine permease (TyrP) of *E. coli*; serine permease (SdaC) of *E. coli*
2.A.43	LCT	Cystine	300–400	7	E (An, Pl, Y)	>20	Lysosomal cystine transporter (cystinosin) of *Homo sapiens*
2.A.44	FNT	Formate; nitrite; acetate	250–630	6–8	B, A, E (Y)	>10	Formate efflux permease (FocA) of *E. coli*
2.A.45	ArsB	Arsenite, antimonite	400–900	12	B, A, E	>20	Arsenical resistance efflux pump of *Staphylococcus aureus*
2.A.46	BenE	Benzoate	400	12	B	2	Benzoate:H^+ symporter (BenE) of *Acinetobacter calcoaceticus*
2.A.47	DASS	Dicarboxylates; phosphate; sulfate	430–920	11–14	B, A, E	>20	Dicarboxylate translocator (SodiT1) of *Spinacia oleracea*
2.A.48	RFC	Reduced folate	500–600	12	E (An)	>10	Reduced folate carrier (RFC) of *Mus musculus*
2.A.49	Amt	Ammonium	390–620	12	B, A, E	>20	Ammonium transporter (AmtB) of *E. coli*
2.A.50	TP-NST	Triosephosphates; glucose 6-phosphate, P_i; nucleotide sugars; nucleotides	300–450	5–12	E (An, Pl, Y) (chloroplasts; plastids; other organelles)	>20	Triosephosphate translocator (TPT) of *Zea mays* chloroplasts; UDP-galactose:UDP exchange transporter of *Homo sapiens* endoplasmic reticulum and Golgi
2.A.51	CHR	Chromate; sulfate (uptake or efflux)	~400	10	B, A	~10	Chromate transporter (ChrA) of *Alcaligenes eutrophus*
2.A.52	NiCoT	Ni^{2+}, Co^{2+}	300–400	8	B	>10	Ni^{2+} uptake permease (HoxN) of *Ralstonia eutropha*
2.A.53	SulP	Sulfate	430–900	10–13	B, A, E	>50	Sulfate permease (SulP) of *Homo sapiens*
2.A.54	MTC	Di- and tricarboxylates	~290	3–6	E (mito)	>10	Mitochondrial tricarboxylate carrier (MTC) of *Rattus norvegicus*
2.A.55	Nramp	Divalent metal ions (uptake)	540–580	8–12	B, A, E	~20	Divalent metal ion; H^+ symporter (Nramp2) of *Homo sapiens*
2.A.56	TRAP-T	C_4-dicarboxylates; acidic amino acids; sugars?	~1,000 (three components)	12 + 4	B, A	>20	Dicarboxylate transporter (DctPQM) of *Rhodobacter capsulatus*
2.A.57	ENT	Nucleosides	~450	11	E	>10	Equilibrative nucleoside transporter-1 (hENT1) of *Homo sapiens*
2.A.58	PNaS	Inorganic phosphate	300–650	>10	B, E	>20	Renal Na^+-dependent phosphate transporter-2 (NPT2) of *Rattus norvegicus*
2.A.59	ACR3	Arsenite	400	10	B, A, E	1	Arsenical resistance-3 protein (ACR3) of *S. cerevisiae*
2.A.60	OAT	Organic anions; prostaglandins; bile acids; bile conjugates	600–700	10–12	E (An)	10	Organic anion transporter (OATP1) of *Rattus norvegicus*; prostaglandin transporter (PGT) of *Rattus norvegicus*

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TABLE 3—*Continued*

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
2.A.61	DcuC	Dicarboxylates	461	10–12	B	10	C_4-dicarboxylate uptake-C porter (DcuC) of *E. coli*
2.A.62	NhaD	Na^+/H^+	410–430	10–12	B, A	3	Na^+/H^+ antiporter (NhaD) of *Vibrio parahaemolyticus*
2.A.63	CPA3	K^+ or Na^+/H^+	>700; possibly multiple components	17 (+?)	B	2	K^+:H^+ antiporter (PhaA-G) of *Rhizobium meliloti*; Na^+:H^+ antiporter (Nha1) of an alkalophilic *Bacillus* species
2.A.64	Tat	Redox proteins	>800 (5 subunits)	9 (1+1+5 or 6+0+1)	B, A, E	>10	Redox protein targeting and translocation (MttA-E) (TatA-E) system of *E. coli*
2.A.65	BRT	Bilirubin	340	5	E (An)	1	Bilirubin transporter of *Rattus norvegicus*
2.A.66	MATE	Drugs	400–700	12	B, A, E	>40	Norfloxacin and other drug efflux pump (NorM) of *Vibrio parahaemolyticus*
2.A.67	OPT	Peptides	600–900	12–15	B, A, E	>20	Oligopeptide transporter (OPT1) of *Candida albicans*
2.A.68	AbgT	Aminobenzoylglutamate	~500	12–13	B	1	Aminobenzoylglutamate transporter (AbgT) of *E. coli*
2.A.69	AEC	Auxin (efflux)	600–700	8–12	B, A, E	~20	Auxin efflux carrier (PIN1)
2.A.70	MSS	Malonate	255 + 129	7 + 4	B	<10	Malonate:Na^+ symporter (MadLM) of *Malonomonas rubra*
2.A.71	FBT	Folate, biopterin, methotrexate	450–650	12	B, E (Pr, Pl)	<10	Folate-biopterin transporter of *Leishmania donovani*
2.A.72	KUP	K^+ (uptake)	400–800	12	B, E (Y, Pl)	~30	The K^+:H^+ symporter (Hak1) of *Neurospora crassa*
2.A.73	ICT	HCO_3^-	380–480	10	B	3	HCO_3^-:Na^+ symporter of *Synechococcus* PCC7942
2.A.74	MET	Thymidine, drugs, steroids	230–270	4	E (An)	5	Lysosomal hydrophobe/amphiphile transporter (MTP) of *Mus musculus*
2.A.75	LysE	Basic amino acids	190–240	5	B	10	Lysine/arginine exporter (LysE) of *Corynebacterium glutamicum*
2.A.76	RhtB	Neutral amino acids and their derivatives	190–230	5	B	10	Neutral amino acid exporter (RhtB) of *E. coli*
2.A.77	CadD	Cd^{2+}; cations	190–220	5	B	4	Cadmium resistance protein (CadD) of *Staphylococcus aureus*
2.A.78	CAAT	Carboxylates, amino acids, amines (efflux)	280–320	10	B, A	>50	MadN of *Malonomonas rubra*
2.C. Ion gradient-driven energizers							
2.C.1	TonB	H^+?; drives solute uptake across outer bacterial membranes	~1,000	1 + 3 + 1	B	>10	TonB-ExbBD outer membrane energizer of *E. coli*
3.A. Pyrophosphate bond hydrolysis-driven transporters							
3.A.1	ABC	All sorts of inorganic and organic molecules of small, intermediate, and large sizes, from simple ions to macromolecules	1,000–2,000 (multidomain; usually multisubunit	10 or 12; variable	B, A, E	>1,000	Maltose permease (MalEFGK) of *E. coli*; multidrug resistance protein (MDR) of *Homo sapiens*
3.A.2	F-AT-Pase	H^+; Na^+	>4,000 (multiple subunits)	$(2)_{12}$, + $(1)_2$, + $(6)_1$	B, A, E (chloro; mito)	>100	F_0F_1-ATPase of *E. coli*

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TABLE 3—*Continued*

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
3.A.3	P-ATPase	Na^+; H^+; K^+; Ca^{2+}; Mg^{2+}; Cd^{2+}; Cu^{2+}; Zn^{2+}, Cd^{2+}, Co^{2+}, Ni^{2+}; Ag^+; phospholipids (flipping)	600–1,200 (sometimes multisubunit)	6–12	B, A, E	>100	KdpABC (K^+ uptake) of *E. coli*
3.A.4	ArsAB	Arsenite, antimonite, tellurite	~1,100 (multidomain; two subunits)	12	B, A, E	>10	Arsenite efflux pump (ArsAB) of *E. coli*
3.A.5	IISP	Proteins	>2,000 (multiple subunits)	SecY (10)	B, A, E	>50	Type II secretory pathway system (SecAYEDFG) of *E. coli*
3.A.6	IIISP	Proteins	>2,000 (multiple subunits)	6 integral membrane constituents	B	>20	Type III secretory pathway system (YscNDRSTUC; LcrD) of *Yersinia* species
3.A.7	IVSP	Proteins, protein-DNA complexes	>2,000 (multiple subunits)	3 integral membrane constituents	B	>20	Type IV secretory pathway system (VirB4, B6, B7, B9, B10, B11) of *Agrobacterium tumefaciens*
3.A.8	MPT	Mitochondrial proteins	>2,000 (multiple subunits)	9 integral membrane constituents	E (mito)	2	Mitochondrial protein translocase (Tom and Tim) proteins of *Saccharomyces cerevisiae*
3.A.9	CEPT	Chloroplast proteins	>2,000 (multiple subunits)	Several integral membrane constituents	E (chloro)	2	Chloroplast envelope protein translocase (IAP) proteins of *Pisum sativum*
3.A.10	H^+-PPase	H^+	660–780	15	E (plant vacuoles), B, A	>10	Vacuolar H^+-pyrophosphatase (V-PPase) of *Arabidopsis thaliana*
3.A.11	DNA-T	Single-stranded DNA	>1,000 (multiple subunits)	3 subunits	B	5	Competence-related DNA transformation transporter (ComEA-EC-FA) of *Bacillus subtilis*
3.B. Decarboxylation-driven active transporters							
3.B.1	NaT-DC	Na^+	~1,000 (3 subunits)	11 (β-subunit)	B, A	10	Oxaloacetate decarboxylase of *Salmonella typhimurium*
3.C. Methyl transfer-driven active transporters							
3.C.1	NaT-MMM	Na^+	~8 subunits; most integral membrane constituents	?	A	2	Na^+-transporting methyltetrahydromethanopterin: coenzyme M methyltransferase of *Methanobacterium thermoautotrophicum*
3.D. Oxidoreduction-driven active transporters							
3.D.1	NDH	H^+ or NA^+(efflux)	14–40 subunits	Multiple integral membrane subunits	B, E (mito, chloro)	>10	NDH of *E. coli*
3.D.2	PTH	H^+ (efflux)	~2,000 (1–3 proteins; 3 domains; dimeric)	$(12–14)_2$	B, E (mito)	>10	PTH of *E. coli*
3.D.3	QCR	H^+ (efflux)	2,000–6,000 multiple (3–11) subunits; dimeric	$(13)_2$	B, E (mito; chloro)	>20	Cytochrome bc_1 complex of *Paracoccus denitrificans*
3.D.4	COX	H^+ (efflux)	2,000–6,000 multiple (3–11) subunits; dimeric	$(12–20)_2$	B, A, E (mito)	>20	Quinol oxidase (Cyo) of *E. coli*
3.D.5	Na-NDH	Na^+ (efflux)	Multiple subunits	?	B	1	Na^+-translocating NADH-quinol reductase of *Vibrio alginolyticus*

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TABLE 3—*Continued*

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
3.D.6	NFO	H^+ or Na^+ (efflux)	Multiple subunits	?	B	~10	H^+- or Na^+-translocating NADH:ferredoxin oxidoreductase (RnfA–H) of *Rhodobacter capsulatus*
3.D.7	HHO	H^+ (efflux)	Multiple subunits	?	A	~5	H_2:heterodisulfide oxidoreductase of *Methanosarcina mazei* Göl
3.D.8	FMF-DH	H^+ or Na^+ (efflux)	Multiple subunits	?	A	~5	Formyl methanofuran dehydrogenase (FwdA-G) of *Methanobacterium thermoautotrophicum*
3.E. Light-driven active transporters							
3.E.1	FAR	H^+ efflux; Cl^- uptake	~250	7	A, E (Y, F)	>20	Bacteriorhodopsin of *Halobacterium salinarum*
3.E.2	PRC	H^+ (efflux)	Multiple subunits	-	B, E (plant chloro)	>20	Reaction center and cytochrome b_6f complex of *Rhodobacter sphaeroides*
4.A. Phosphotransferase systems							
4.A.1	Glc	Glucose; *N*-acetylglucosamine; α- and β-glucosides	~2,000 (3 domains; dimeric)	$(8)_2$	B	~30	Glucose IICB-IIA of *E. coli*
4.A.2	Fru	Fructose; mannitol	~2,000 (3 domains; dimeric)	$(6)_2$	B	~30	Fructose IIB′BC-IIAMH of *E. coli*
4.A.3	Lac	Lactose; cellobiose; *N*,*N*′-diacetylchitobiose	~2,000 (3 domains; dimeric)	$\sim(8)_2$	B	~20	Lactose IICB-IIA of *Staphylococcus aureus*
4.A.4	Gut	Glucitol	~2,000 (3 domains; dimeric)	$(8)_2$	B	2	Glucitol IICB-IIA of *E. coli*
4.A.5	Gat	Galactitol	~2,000 (3 domains; dimeric)	$\sim(8)_2$	B	1	Galactitol IIC-IIB-IIA of *E. coli*
4.A.6	Man	Glucose, mannose, fructose, sorbose, etc.	~2,000 (4 domains; probably dimeric)	$(6(IIC) + 1(IID))_2$	B	5	Mannose IIAB-IIC-IID of *E. coli*
9.A. Transporters of unknown classification							
9.A.1	PST	Polysaccharides (export)	400–500	12	B	>10	Lipopolysaccharide exporter (RfbX1) of *E. coli*
9.A.2	MerTP	Hg^{2+} (uptake)	~200	2	B	~10	Mercuric ion transporter (MerTP), encoded on the IncJ plasmid pMERPH of *Shewanella putrefaciens*
9.A.3	MerC	Hg^{2+} (uptake)	137	1	B	~10	Mercuric ion uptake transporter (MerC), encoded on the IncJ plasmid pMERPH of *Shewanella putrefaciens*
9.A.4	PnuC	Nicotinamide mononucleotide (uptake)	~320	7	B	~10	Nicotinamide mononucleotide uptake permease (PnuC) of *Salmonella typhimurium*
9.A.5	Oxal	Proteins	400–600	3–5	B, E (mito; chloro)	~10	Cytochrome oxidase biogenesis protein Oxal of *Saccharomyces cerevisiae*
9.A.6	INT	Nucleosides	230–270	4	E (An)	~10	Intracellular nucleoside transporter (MTP) of *Mus musculus*
9.A.8	FeoB	Fe^{2+} (uptake)	~800	8–13	B, A	~10	Fe^{2+} uptake transporter (FeoB) of *E. coli*
9.A.9	FeT	Fe^{2+} (Co^{2+}, Cd^{2+}) (uptake)	552	6	E (Y)	1	Fe^{2+} transporter (Fet4p) of *Saccharomyces cerevisiae*
9.A.10	OFeT	Fe^{2+} (uptake)	404	6	B, A, E	~10	Oxidase-dependent Fe^{2+} transporter (Ftr1p) of *Saccharomyces cerevisiae*

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TABLE 3—*Continued*

TC no.	Family	Substrate(s)[a]	Size range[b] (no. of residues)	*n*TMSs[c]	Organisms[d]	No. of members	Examples
9.A.11	Ctr1	Cu^{2+} (uptake)	406	2–3	E (Y)	1	Copper transporter (Ctr1p) of *Saccharomyces cerevisiae*
9.A.12	Ctr2	Cu^{2+} (uptake)	160–200	3	E	~10	Copper transporter (Ctr2p) of *Saccharomyces cerevisiae*
9.A.13	scFAT	Short-chain fatty acids (uptake)	~470	12	B, A	~5	AtoE of *E. coli*
9.A.14	NPC	RNA; proteins, small molecules, etc.	30–50 proteins	—	E	—	Nuclear pore complex of *Saccharomyces cerevisiae*
9.A.15	Ami	Short-chain aliphatic amides; urea (uptake)	170–210	6	B	~5	Amide transporter (AmiS) of *Pseudomonas aeruginosa*
9.A.16	SDT	DNA	~800	4	B	>10	Septum DNA translocation pore (SpoIIIE) of *Bacillus subtilis*
9.A.17	MIT	Heavy-metal ions, Mg^{2+}, Mn^{2+}, Co^{2+}, Ni^{2+}, Fe^{2+}; Al^{3+}; Mn^{2+}	200–360	2–3	B, A, E	>50	Metal uptake and efflux permease (CorA) of *E. coli*
9.A.18	PUP	Peptides, antibiotics (uptake)	406	7	B	2	Microbin uptake permease (SbmA) of *E. coli*
9.A.19	MgtE	Mg^{2+}, Co^{2+} (uptake)	310–470	4–5	B, A	10	Mg^{2+} transporter E (MgtE) of *Bacillus firmus*
9.A.20	LCT	Monovalent cations	570	8–10	E (Pl)	1	Low-affinity cation transporter (LCT1) of *Triticum aestivum*

[a] Substrates of single transporters within a family are separated by commas; substrates transported by different protein members of the family are separated by semicolons.

[b] Size range (in number of amino acyl residues) when a single type of subunit is present, or for the entire complex when several types of subunits are present.

[c] *n*TMSs, number of transmembrane α-helical segments in a polypeptide chain. Subscripts refer to the number of polypeptide chains in the complex when known. A subscript *n* indicates an oligomeric structure of unknown or poorly defined number of subunits. If alternative structures are found for different transporters within a single family, these are separated by semicolons. In a few ambiguous cases, the subunit is indicated in parentheses.

[d] Abbreviations: B, bacteria; A, archaea; E, eukaryotes; G− B, gram-negative bacteria; G+ B, gram-positive bacteria; Y, yeasts; F, fungi; Pr, protozoans; Pl, plants; An, animals; mito, mitochondria; chloro, chloroplasts; plastids, nonphotosynthetic plastids of plants; phage, bacteriophages. These abbreviations are used in the tables. Other abbreviations are defined in Table 2 or the text.

[e] See Tables 8 and 9.

one or two letters followed by four, five, or six digits. A given letter is used by only one database: GenBank (GB), SP, or Protein Information Resource (PIR). Thus, for example, O, P, and Q are used exclusively by SP; D, J, K, L, M, U, X, Y, and Z are used exclusively by GB; and A, B, C, H, I, and S are used exclusively by PIR. However, when AB, AE, or AF is followed by a six-digit number, this is an alternative GB accession number, and when JC, JH, or JN is followed by a four-digit number, this is a PIR accession number. It should be noted that a single sequenced protein may have multiple accession numbers, but no SP or PIR accession number refers to more than one protein. Because a GB accession number refers to a nucleotide sequence that may encode multiple proteins, a GB accession number may provide the sequences of several proteins.

A table entitled Cross-Referencing Permeases by Accession Number is included in our web site. In this table, accession numbers for most of the proteins included in the TC system as of June 1999 are tabulated in alphabetical and numerical order. These may be of general utility to the student of transport, as their availability allows one to easily search all databases using the various BLAST search tools (3). Knowledge of a TC number allows one to quickly identify (i) the protein referred to, (ii) the transport system of which that protein is a constituent, (iii) the substrate specificity of that system, (iv) the family to which that permease belongs, (v) the mode of transport used, (vi) the energy-coupling mechanism used, and (vii) many of the characteristics of that permease family. Thus, cross-referencing by accession number is useful when trying to identify the family to which a newly sequenced protein belongs.

As noted above, one needs only to conduct a BLAST search, and all sufficiently similar homologs will be displayed. When the accession number of any one of these retrieved homologs is shown to correspond to one of the established members of a family, the family to which the newly sequenced protein belongs is immediately known. Furthermore, by identifying the proteins with the highest BLAST scores (smallest *P* values), one can immediately recognize the closest homologs. This information provides an indication of the most likely substrate specificity, energy-coupling mechanism, and physiological function of the newly sequenced permease protein. Cross-referencing of accession numbers and TC numbers therefore provides a simple and rapid approach to the initial characterization of a newly sequenced porter. I and my colleagues, working with Andrei Lupas (SmithKline-Beecham), are currently developing a search tool based on the TC system that will allow anyone to search the complete TC system using sequence, sequence motif, accession number, gene name, protein name, family name, etc. (A. Lupas et al., unpublished data).

GROUPING TRANSPORT SUBSTRATES BASED ON BIOLOGICAL SIGNIFICANCE

In 1993 and again in 1996, Monica Riley presented an extensive tabulation of *E. coli* gene products (114, 115). To fa-

TABLE 4. Classification of transport system substrates based on biological significance

Category and substrate type	Subcategories
I. Inorganic molecules	A. Nonselective B. Water C. Cations D. Anions E. Others
II. Carbon compounds	A. Sugars, polyols, and their derivatives B. Monocarboxylates C. Di- and tricarboxylates D. Organic anions (noncarboxylates) E. Others
III. Amino acids and their derivatives	A. Amino acids and conjugates B. Amines, amides, and polyamines C. Peptides D. Other related organocations E. Others
IV. Bases and their derivatives	A. (Nucleo)bases B. Nucleosides C. Nucleotides D. Other nucleobase derivatives E. Others
V. Vitamins, cofactors, and their precursors	A. Vitamins and vitamin or cofactor precursors B. Enzyme and redox cofactors C. Siderophores; siderophore-Fe complexes D. Signaling molecules E. Others
VI. Drugs, dyes, sterols, and toxics	A. Multiple drugs and dyes B. Specific drugs C. Bile salts and conjugates D. Sterols and conjugates E. Others
VII. Macromolecules	A. Carbohydrates B. Proteins C. Nucleic acids D. Lipids E. Others
VIII. Miscellaneous compounds	

cilitate this endeavor, enzymes were classified based on the nature of the molecule(s) (substrates) acted on. In order to cross-reference transport systems based on substrate specificities, a basis for classifying potential substrates had to be devised. We have done so, creating a system that includes virtually all currently recognized transport substrates. This system of cross-referencing transporters is described here.

All known transport substrates have been classified into eight categories (Table 4). These categories are I, inorganic molecules; II, carbon compounds, III, amino acids and derivatives; IV, bases and derivatives; V, vitamins, cofactors, signaling molecules, and their precursors; VI, drugs, dyes, sterols, and toxic substances; VII, macromolecules; and VIII, miscellaneous compounds.

Most inorganic molecules (category I) are cationic or anionic. However, some channel proteins are nonselective, and aquaporins of the MIP family (TC 1.1) transport water selectively. The four defined subcategories for category I therefore include A, nonselective; B, water; C, cations; and D, anions. Inorganic compounds not falling into one of these subcategories are classified as others (subcategory E), and this subcategory can be subdivided in the future if desired.

Carbon compounds (category II) have similarly been grouped into four defined subcategories: A, sugars, polyols, and their derivatives; B, monocarboxylates; C, di- and tricarboxylates; and D, noncarboxylic organic anions (organophosphates, phosphonates, sulfonates, and sulfates). Subcategory E (others) encompasses all other carbon compounds.

Amino acids and their derivatives (category III) have been subdivided into A, amino acids and conjugates; B, amines, amides, and polyamines; C, peptides; D, other related organocations; and E, others. Bases and their derivatives (category IV) have been subcategorized into A, nucleobases; B, nucleosides; C, nucleotides; D, other related derivatives; and E, others. Vitamins, cofactors, and cofactor precursors (category V) have been subcategorized into A, vitamins and vitamin or cofactor precursors; B, enzyme and redox cofactors; C, siderophores and siderophore-iron complexes; D, signaling molecules; and E, others. Drugs, dyes, sterols, and toxics (category VI) have been subcategorized into A, multiple drugs and dyes; B, specific drugs; C, bile salts and conjugates; D, sterols and conjugates; and E, others. Category VII is devoted to macromolecules: A, complex carbohydrates; B, proteins; C, nucleic acids; D, lipids; and E, others. Finally, category VIII (miscellaneous) encompasses any transport substrate that does not fall into categories I to VII. So far no transport substrate has been relegated to category VIII, and very few of those in categories I to VII have fallen into the "other" category.

A few compounds belong to more than one category. For example, bile acids fall into both category II.B and category VI.C. Theoretically, oligosaccharides (e.g., lactose, raffinose, and maltooligosaccharides) could be classified either in II.A or in VII.A. We have elected to put oligosaccharides into category II.A and reserve category VII.A for larger molecules such as polysaccharides, teichoic acids, and lipooligosaccharides. Thus, category II.A generally refers to smaller carbohydrates normally taken up by cells for purposes of carbon catabolism, while category VII.A refers to structural carbohydrates that are synthesized by cells and exported intact.

Some permease systems transport a range of compounds that fall into more than one category. For example, a single ABC export system may catalyze efflux of multiple drugs (VI.A) and peptides (III.C), and it may also facilitate phospholipid flipping between the two bilayers of a membrane (VII.D). Such permeases are rare, but when they do occur, they will be included in all applicable categories.

DISTRIBUTION OF TRANSPORTER TYPES BASED ON SUBSTRATE SPECIFICITY

As described above, Table 4 groups potential transport substrates according to structure and biological significance. This system of substrate classification has been used to cross-reference transport systems according to the types of substrates and the specific substrate(s) transported.

Table 5 presents the distribution of transporter types based on substrate specificity. In this table, permeases are categorized into four groups: α-type channels, β-type porins, primary carriers (regardless of the primary source of energy utilized and including PTS-type group translocators), and secondary carriers (including uniporters, symporters, and antiporters).

TABLE 5. Distribution of transporter families based on substrate specificity

Substrate	No. of families of indicated type using substrate[a]				
	Channels	Porins	Primary carriers	Secondary carriers	Total
I. Inorganic molecules	**32**	**13**	**16**	**44**	**105**
A. Nonselective	13	9	0	0	22
B. Water-selective	2	0	0	0	2
C. Cations	14	2	15	30	61
D. Anions	4	4	3	17	28
II. Carbon sources	**2**	**3**	**2**	**26**	**33**
A. Sugars and polyols	1	2	2	4	9
B. Monocarboxylates	1	1	0	13	15
C. Di- and tricarboxylates	0	0	1	12	13
D. Organoanions (noncarboxylic)	0	1	1	2	4
III. Amino acids and their derivatives	**3**	**1**	**2**	**20**	**26**
A. Amino acids and conjugates	0	0	1	18	20
B. Amines, amides, and polyamines	3	1	2	8	14
C. Peptides	0	0	1	4	5
IV. Bases and their derivatives	**0**	**2**	**0**	**10**	**12**
A. Nucleobases	0	0	0	2	2
B. Nucleosides	0	1	0	6	7
C. Nucleotides	0	1	0	2	3
V. Vitamins, cofactors, and cofactor precursors	**0**	**2**	**1**	**11**	**14**
A. Vitamins and vitamin or cofactor precursors	0	1	1	7	9
B. Enzyme and redox cofactors	0	1	1	4	6
C. Siderophores; siderophore-Fe complexes	0	2	1	1	4
D. Signaling molecules	0	0	1	3	4
VI. Drugs, dyes, sterols, and toxics	**0**	**1**	**1**	**6**	**8**
A. Multiple drugs	0	1	1	5	7
B. Specific drugs	0	0	1	2	3
C. Bile salts and conjugates	0	0	1	3	4
D. Sterols and conjugates	0	0	0	3	3
VII. Macromolecules	**5**	**9**	**8**	**4**	**26**
A. Carbohydrates	0	3	1	2	6
B. Proteins	5	6	6	1	18
C. Nucleic acids	0	1	2	0	3
D. Lipids	0	0	2	1	3

[a] Totals are boldfaced. Because a family can include members that transport different substrates, it may be found in more than one subcategory. Hence, the number of families in each category is less than or equal to the sum of the numbers of families in the corresponding subcategories.

Transporter types of unknown mode of transport or energy-coupling mechanism (categories 9.A and 9.B) were not included in Table 5.

α-Type channel proteins (TC 1.A) generally either are nonselective (13 types) or function in the transport of inorganic ions (I.C and I.D; 18 families) or proteins (VII.B; 5 families). One type (aquaporins in the MIP family; TC 1.A.8) transports water, while another type of the same family (glycerol facilitators of the MIP family) transports straight-chain polyols and small organic molecules such as urea. Some MIP family proteins may transport both water and small, neutral organic molecules, but with the possible exception of a single MIP family member (154), none of the MIP family channel proteins have been shown to be selective for ions or larger molecules. Besides MIP family members, only two other recognized channel families include members that are specific for organic compounds. These families are the urea transporter family (TC 1.A.44) and the phospholemman (PLM) family (TC 1.A.27). The PLM family includes members that transport organic anions. Channel-forming toxins (TC 1.C) are generally nonspecific, or they exhibit weak charge selectivity (i.e., anion selective or cation selective).

Porins (TC 1.B) are pore-forming proteins that exhibit β-barrel structures or variations on the β-barrel structural theme. They are localized to the outer membranes of gram-negative bacteria, mitochondria, and chloroplasts. They exhibit a wider range of substrate selectivities than do the α-type channel proteins cited above (Table 5). However, like channel protein types, most porins either are nonselective, exhibit some degree of anionic or cationic selectivity, or function in the export of macromolecules across the outer membranes of gram-negative bacteria. Many porins allow passage of any molecule smaller than a certain cutoff size (usually about 500 to 1,000 Da). Of the macromolecular export porin types, more than half export proteins, but several transport complex carbohydrates, and at least one functions in DNA transport.

Examination of the specificities of porins for organic substrates reveals a wide variety of specificities. The maltoporin of *E. coli* (TC 1.B.3.1.1) and the raffinose porin of *E. coli* (TC 1.B.15.1.1) are both inducible by their sugar substrates, but

TABLE 6. Substrate selectivities of cytoplasmic membrane channel proteins (excluding porins)

Channel type	No. of families identified	Families
Nonselective	14	MscL, CAPs, holins, colicin, ICP, αHL, aerolysin, colicin V, ε-toxin, YKT-K1, connexin, innexin, CHL, MscS
Cation selective	9	ACC, VIC, GIC, RIR-CaC, TRP-CC, ENaC, LIC, NSCC1, NSCC2
Anions	6 or 7	LIC, ClC, O-ClC, E-ClC, PLM, Tic110, (ABC)
H^+	2	IVC, CybB
Na^+	2	ENaC, VIC
K^+	2	VIC, IRK-C
Ca^{2+}	5	ACC, VIC, GIC, RIR-CaC, TRP-CC
NH_4^+	1	SAT
Small neutral organic molecules	1	MIP, UT
Organic anions	1	PLM
Urea	2	MIP, UT
Water	2	MIP, UT
Proteins	5	Holins, Bcl-2, DT, MscL, BTT

while maltoporin is quite specific for maltooligosaccharides, the raffinose porin transports a variety of oligosaccharides. Other porins have been reported to preferentially transport organophosphates (TC 1.B.1.1.2), fatty acids (TC 1.B.9.1.1), nucleosides (TC 1.B.10.1.1), or organic solvents such as toluene (TC 1.B.9.2.1). Still another type apparently exhibits specificity for short-chain amides and urea (TC 1.B.16.1.1). Members of the outer membrane receptor family (TC 1.B.14) import vitamin B_{12} and a variety of iron-siderophore complexes in a process that is coupled to the PMF via the TonB-dependent energy-coupling system (TC 2.C.1). To what degree these proteins are biochemically selective is not always clear, although they are often encoded within operons that exhibit specific induction properties, and these systems are constituents of transenvelope transport complexes.

Primary carriers are in general highly specific for one or a few related substrates, and like channels, they are almost always selective for inorganic ions or macromolecules. Those specific for organic molecules of small or intermediate sizes belong to either of two superfamilies, the ABC superfamily (TC 3.A.1) or the PTS functional superfamily (TC 4.A.1-6). The PTS is actually a group translocating system, since it phosphorylates its substrates using phosphoenolpyruvate as the phosphoryl donor. For the purpose of tabulating substrate specificities as presented in Table 5, we have grouped this functional superfamily together with the active transporters. The energy-coupling mechanisms used for the transport of ions are diverse, involving pyrophosphate bond hydrolysis, decarboxylation, methyltransfer, oxidoreduction (both hydride shift and electron flow), and light absorption (Fig. 1). By contrast, all macromolecular primary active transporters use ATP or GTP hydrolysis to drive export, although a few macromolecular secondary active exporters use the PMF as the energy source for transport.

Secondary carrier types (TC class 2, subclass 2.A) exhibit a very different spectrum of substrate specificities. None is nonselective or water selective, but many are selective for specific inorganic cations or anions, and a few appear to function in the export of lipids, proteins, or complex carbohydrates, as is characteristic of primary active transporters. Others function in the transport of the many different types of small organic molecules found in biological systems. Thus, every class of molecules included in Table 5 is transported by one or more currently identified secondary carrier(s). For example, members of four transporter families are known to transport sugars and polyols; 13 types transport monocarboxylates, and 12 types transport di- and tricarboxylates. Eighteen types function in the transport of amino acids and their conjugates. It is clear that secondary carriers are primarily responsible for the transport of small organic molecules in virtually all living organisms.

The last column in Table 5 reveals the total numbers of families involved in the transport of the various types of biologically important compounds. About equal numbers of families are concerned with transport of inorganic and organic compounds, with most of the 105 families for inorganic molecules transporting ionic species. Thirty-three families are concerned with carbon source uptake, while 34 are concerned with the uptake of nitrogen-containing compounds (amino acids, bases, and their derivatives). Only 14 families include members that take up compounds in category V (i.e., vitamins, cofactors, signaling molecules, and related compounds), while only 8 families are concerned with transport of hydrophobic substances (category VI). Macromolecules are exported via the transporters of 26 families. While inorganic molecules and macromolecules are transported by all four types of systems, small organic substances are transported almost exclusively by secondary carriers (Table 5).

SUBSTRATE SELECTIVITIES

Cytoplasmic Membrane Channel Proteins (Excluding Porins)

α-Type channel proteins (TC category 1.A) and pore-forming toxins (TC category 1.C) are largely responsible for the diffusion-limiting flux of inorganic ions between the cell cytoplasm and the external milieu or between intracellular compartments of eukaryotic cells. α-Type channel proteins are ubiquitous, but they are particularly prevalent in animals that use electrical signaling for purposes of neuronal signaling and the control of muscle contraction. Thus, in contrast to all other types of transporters, α-type ion channel protein families are primarily restricted to animals. Pore-forming toxins are most frequently produced by bacteria, but they can target the membranes of either prokaryotic or eukaryotic cells.

Table 6 provides a more detailed breakdown of the substrate selectivities of channel-type proteins. The majority of these channel types either are nonselective or merely exhibit a charge preference, preferring inorganic cations over anions or anions over cations. Many of these nonselective channel types are toxic proteins or peptides (CAPs) that are secreted by one cell in order to kill another. Most other channel types exhibit

a striking degree of specificity. Seven of these are selective for chloride and other anions, two are specific for Na^+, and two are selective for K^+. Five channel types are specific for Ca^{2+}. Only MIP and urea transporter family members transport small neutral molecules, as noted above. Five families include members that preferentially transport proteins, but one of these, the holin functional superfamily, consists of 16 families of functionally (but not phylogenetically) similar proteins. The holins (156) and Bcl-2 (1) are bacterial and animal proteins, respectively, that promote cell suicide or apoptosis. Members of the diphtheria toxin and botulinum and tetanus toxin families transport bacterial toxins into target animal cells (69, 86). The proteins of the MscL family may provide protection against osmotic downshift, but they have been shown to be capable of catalyzing protein export as well (2, 8, 12).

Table 7 provides a detailed breakdown of channel-type proteins according to their substrate specificities. The individual families represented in each category are tabulated according to TC number. The name or abbreviation, source, and mode of regulation (when known) are also provided. Among the nonselective channels is the MscL channel of *E. coli*, which has been reported to exhibit a slight preference for cations over anions and to also transport proteins, while the MscS channel of *E. coli* has been reported to exhibit a slight anionic preference. Holins function primarily in protein export, while connexins and innexins function to form tight junctions between adjacent animal cells in vertebrates and invertebrates, respectively. Otherwise, all nonselective channel proteins or peptides are designed for export from the cell of synthesis for the purpose of biological warfare. These proteins and peptides are derived from phages, bacteria, and eukaryotes. Archaeal protein and peptide toxins that function by pore formation have not yet been characterized functionally.

Low-specificity cation-selective channels include both acetylcholine- and serotonin-activated ligand-gated ion channel family members, ATP-sensitive ATP-gated cation channel family members, glutamate-gated ion channel family members (all of animal origin), and the MscL family proteins noted above (of bacterial origin). Other cation-selective channels include the members of the nonselective cation channel 1 (NSCC1) and NSCC2 families. Two families, the influenza virus matrix-2 channel and CybB families, selectively transport protons. Still others exhibit selectivity for a particular cation, Na^+, K^+, NH_4^+, or Ca^{2+}.

Three of the anion-selective channel types listed appear to transport Cl^- selectively, although at least three additional families include members that transport other anions as well. Thus, chloride channel (ClC) family proteins transport a variety of inorganic anions, while members of the PLM family transport a wide range of monovalent inorganic and organic anions (Table 7). Three types of ligand-gated channel-forming members of the ligand-gated ion channel family specifically transport chloride, and a high degree of selectivity may be a characteristic of the voltage-regulated organellar ClC (O-ClC) proteins. The organellar ClC family is not related to the ClC family. Proteins of the large and ubiquitous ClC family are found in all three domains of life (bacteria, archaea, and eukarya), but only eukaryotic members have been functionally characterized. The epithelial ClC family includes members that can transport a range of anions.

Bacterial Outer Membrane Porins

All characterized integral membrane proteins in the outer (lipopolysaccharide-containing) membranes of gram-negative bacteria are believed to consist largely of β-structure rather than α-structure, and structural features may provide a targeting signal for the outer membrane (18, 47). Among these proteins are the oligomeric (mostly trimeric) porins, several of which have been structurally characterized (48, 55, 81) (see below). These proteins can transport small molecules nonselectively, or they can be highly selective for a single class of molecules (18, 90, 150). They are found in the outer membranes of mitochondria and plant plastids (11, 40) and may be present in the outer mycolic acid-containing membranes of acid-fast gram-positive bacteria, such as species of mycobacteria and *Nocardia* (61, 109, 133). Because of their unique subcellular locations and structures, outer membrane β-barrel porins are classified separately from the α-type channel proteins.

Lipopolysaccharide-containing outer membranes of gram-negative bacteria provide an unusually effective barrier against hydrophobic dyes, detergents, and hydrophobic and amphipathic drugs. However, by virtue of the presence of β-barrel-type porins in these structures, the membranes are generally permeable to hydrophilic molecules smaller than 650 Da (91). While some of these porins are essentially nonspecific, others appear to exhibit a high degree of selectivity. Tables 8 and 9 summarize the substrate specificities of various recognized outer membrane porins. All of these proteins are derived from gram-negative bacteria except for members of the mitochondrial and plastid porin family (TC 1.B.8). Table 8 presents the breakdown of substrate selectivities. The conclusions reached are in some cases based on detailed biochemical analyses, but in other cases, physiological data were used to derive the conclusions presented. Thus, not all of the porins represented may prove to be as selective as indicated. A significant percentage of the porins are either nonselective or selective only with respect to the charge of the transported species. Among the anion-selective porins, some function primarily in the transport of phosphate, pyrophosphate, nucleotides, and/or fatty acids. Other small-substrate-selective porins have been reported to exhibit specificity for nucleosides, oligosaccharides, short-chain amides, or toluene. Still other porin-like proteins are designed for the export of drugs and heavy metals, while others function in the import of iron complexes and the vitamin precursor cobalamine. Several porin types apparently function in the export of complex carbohydrates and proteins. In most of these cases, the degree of specificity exhibited by these porins has not been studied extensively.

Table 9 provides a detailed breakdown of porin types according to their physiologically relevant substrate specificities. The individual TC numbers and families are presented, allowing the reader to trace the proteins cited and to identify the primary references so as to be able to examine the experimental evidence concerning their specificities.

Secondary Carriers

Secondary carriers catalyze (i) the transmembrane transport of a single molecular species (uniport), (ii) the cotransport of a solute with a cation (symport), (iii) the countertransport of a solute against a cation (antiport), or (iv) the exchange of one solute for another (solute-solute antiport). Several can catalyze more than one such process (i.e., uniport or symport as well as solute-solute antiport), and single mutations can interconvert uniporters and symporters (143, 151). Some can cotransport several cations while countertransporting other cations. These proteins exhibit a wide variety of topological types and substrate specificities. They are responsible for the transport of most organic solutes across biological membranes, particularly

TABLE 7. Classification of channel proteins (excluding porins) according to substrate specificity

Substrate	TC no.	Family	Source	Regulation
I.A. Nonselective				
Nonselective	1.A.17	CSC	Pl	
Nonselective (slight cation-over-anion selectivity)	1.A.22	MscL	B	Mechanosensitive (osmotic pressure)
Nonselective (slight anion-over-cation selectivity)	1.A.23	MscS	B	Mechanosensitive (osmotic pressure)
Nonselective	1.A.24	Connexin	An	
Nonselective	1.A.25	Innexin	An	
Nonselective	1.A.28–1.A.43	Holin	B, phage	
Nonselective	1.C	All toxin-type pore-forming proteins/peptides	B, A, E	
I.B. Water				
Water	1.A.8	MIP aquaporins	B, E	Hormonal regulation in animals
Water	1.A.44	UT	An	
I.C. Cations				
Cation selective	1.A.1.11	VIC	An	Voltage sensitive
Cation selective	1.A.3.1	RIR-CaC	An	Ryanodine sensitive
Cation selective	1.A.3.2	RIR-CaC	An	Inositol triphosphate sensitive
Cation selective	1.A.4	TRP-CC	An	Responsive to pain, heat, etc.
Cation selective	1.A.5	PCC	An	Ca^{2+} activated
Cation selective	1.A.6.1.2	ENaC	An	Proton gated
Cation selective	1.A.6.2.1	ENaC	An	
Cation selective	1.A.7	ACC	An	Exogenous-ATP sensitive
Cation selective	1.A.9.1	LIC	An	Acetylcholine activated
Cation selective	1.A.9.2	LIC	An	Serotonin activated
Cation selective	1.A.10.1.3	GIC	An	Exogenous-glutamate sensitive
Cation selective	1.A.13	Gramicidin	B	
Cation selective	1.A.14	NSCC1	An	
Cation selective	1.A.15	NSCC2	E (Y, F, An)	
Cation selective	1.A.16	Mid1	Y	Stretch activated
Cation selective	1.A.26	SAT	Pl	
H^+	1.A.19	IVC	An (virus)	
H^+	1.A.20	CybB	An, Pl	Arachidonate
H^+	1.A.45	Mot	B	
K^+	1.A.1.1–9	VIC	B, A, Y, An, Pl	Voltage sensitive; Ca^{2+} activated; cyclic nucleotide gated
K^+	1.A.2	IRK-C	An	ATP regulated; G protein regulated; voltage sensitive
Na^+	1.A.1.10	VIC	An	Voltage sensitive
Na^+	1.A.6.1.1	ENaC	An	Ligand regulated
Na^+	1.A.6.3.1	ENaC	An	Peptide ligand regulated
Na^+	1.A.45	Mot	B	
Na^+, K^+, (Ca^{2+}) selective	1.A.10	GIC	An	Glutamate gated
NH_4^+	1.A.26	SAT	Pl	
I.D. Anions				
Cl^-	1.A.9.3	LIC	An	Glycine inhibited
Cl^-	1.A.9.4	LIC	An	Glutamate inhibited
Cl^-	1.A.9.5	LIC	An	γ-Aminobutyrate inhibited
Cl^-	1.A.12	O-ClC	An	Voltage regulated
Cl^-	1.A.13	E-ClC	An; B?	Ca^{2+}-calmodulin kinase regulated
Cl^- (anion selective)	1.A.11	ClC	B, A, E	Voltage regulated
Cl^- (anion selective)	1.A.16	Tic110	Pl	
Cl^- (anion selective)	1.A.27	PLM	An	Hyperpolarization activated
II.A. Sugars and polyols				
Glycerol	1.A.8	MIP-glycerol	B, E	Glycerol inducible in bacteria
Propanediol	1.A.8	MIP-glycerol	B, E	Glycerol inducible in bacteria
II.B. Monocarboxylates				
Organic anions	1.A.27	PLM	An	Hyperpolarization activated
Gluconate	1.A.27	PLM	An	Hyperpolarization activated
Glutamate	1.A.27	PLM	An	Hyperpolarization activated
Isethionate	1.A.27	PLM	An	Hyperpolarization activated
Lactate	1.A.27	PLM	An	Hyperpolarization activated
Taurine	1.A.27	PLM	An	Hyperpolarization activated
III.B. Amines, amides, and polyamines				
Methylammonium	1.A.26	SAT	Pl	
Urea	1.A.8	MIP	B	
Urea	1.A.44	UT	An	
VII.B. Proteins				
Autolysins	1.A.28–1.A.43	Holin	B, phage	
Botulinum toxin, L-chain[a]	1.C.8.1.1	BTT	B	
Cytochrome *c*	1.A.21	Bcl-2	An	
Diphtheria toxin, A-chain[a]	1.C.7.1.1	DT	B	
Nucleases	1.A.34.3	Holin	B	
Proteins	1.A.18	Tic110	Pl	
Tetanus toxin, L-chain[a]	1.C.8.1.2	BTT	B	
Thioredoxin	1.A.22.1.1	MscL	B	Osmotic downshift

[a] These toxins in TC category 1.C function to import proteins into the host cell.

TABLE 8. Substrate selectivities of bacterial outer membrane porins

Channel type	No. identified
Nonselective	9
Anions	3
Phosphate	2
Pyrophosphate	1
Cations	4
Heavy metals	1
Nucleotides	1
Nucleosides	1
Monosaccharides	1
Oligosaccharides	2
Fatty acids	1
Short-chain amides	1
Urea	1
Toluene	1
Drugs	1
Iron complexes	2
Vitamins and cofactors	2
Complex carbohydrates	3
Proteins	6

those of eukaryotes that lack nutrient uptake permeases of the ABC superfamily (130).

Table 10 tabulates secondary carriers according to substrate. Secondary carriers are known as those that transport almost any inorganic ion of biological importance. Virtually all inorganic mono-, di-, and trivalent cations as well as a wide variety of biologically important inorganic anions are substrates of these transporters. In addition, all classes of organic molecules are transported by secondary carriers (Table 10). Only a few secondary carriers are believed to function in the export of macromolecules (complex polysaccharides, lipids, and proteins).

Table 11 provides a detailed breakdown of secondary transporters according to substrate, but in contrast to Table 10, Table 11 provides TC number, family, energy-coupling mechanism, and organismal distribution. Monovalent cations are generally transported either in symport with or by antiport against one or more other cations. Thirteen families are primarily concerned with the catalysis of monovalent cation transport. Di- and trivalent cations are probably taken up by uniport or by H^+ or Na^+ symport, and efflux is probably mediated by H^+ antiport. In many of these cases, the energy-coupling mechanism is not well established. Members of 18 families mediate the transport of these ions, and several of these families include members that can catalyze either uptake or efflux.

A large variety of inorganic anions bearing one, two, or three negative charges can be accommodated by secondary carriers, some functioning with inwardly directed polarity and others with outwardly directed polarity (entry I.D). Sometimes fairly close homologs function with opposite polarity, as noted above for multivalent cation permeases. The mechanisms of energy coupling are known for most of these permeases. Sixteen families are represented under entry I.D.

Sugars and polyols are most frequently transported by MFS permeases, and 6 of the 29 MFS families are concerned with sugar transport. However, three other families (solute-sodium symporter, glycoside-pentoside-hexuronide, and L-rhamnose transporter [RhaT]) are also represented under entry II.A. Of these three families, the glycoside-pentoside-hexuronide family appears to be distantly related to the MFS, based on PSI-BLAST results (125) as well as hydropathy analyses. The same could not be demonstrated for the solute-sodium symporter and RhaT families. The RhaT family may be distantly related to proteins of another superfamily, the drug-metabolite transporter superfamily (D. L. Jack and M. H. Saier, Jr., unpublished results). Monocarboxylates are most often taken up by H^+ symport (76), although other mechanisms are sometimes operative. Protein members of 14 families catalyze monocarboxylate transport. Di- and tricarboxylates are also usually accumulated in the cell cytoplasm by H^+ symport, and 13 families are involved. Surprisingly, members of just 2 of the 14 families that transport monocarboxylates also transport dicarboxylates. Thus, 12 families are monocarboxylate specific, while 11 are di- and tricarboxylate specific. Organophosphates are the only noncarboxylic organic anions represented under entry II.D (Table 11), and only two families, the TP-NST family and the MFS, are involved. Inorganic phosphate antiport is the primary mechanism believed to be operative under most physiological conditions for organophosphate ester transport via members of both of these families.

Amino acids and their conjugates (entry III.A) can be taken up by H^+ or Na^+ symport or by substrate-substrate antiport. Twenty characterized families are involved in amino acid transport. Three of these families (amino acid, polyamine-organocation, amino acid/auxin permease, and hydroxy/aromatic amino acid permease) appear to be distantly related to each other, and they constitute the putative amino acid transporter superfamily (155; D. L. Jack, I. T. Paulsen, and M. H. Saier, Jr., submitted for publication). Three families (L-lysine exporter, resistance to homoserine/threonine, and carboxylate/amino acid/amine transporter) appear to be concerned with amino acid efflux in prokaryotes. Amines, amides, and polyamines (entry III.B) are substrates of permeases from nine distinct families, and the same energy-coupling mechanisms observed for amino acids are operative. All of these families include members that can transport amino acids and are therefore listed under entry III.A as well as entry III.B. Four families of secondary carriers appear to mediate peptide uptake, and the mechanism involved is probably proton symport for members of all four families. Two of these families (MFS and proton-dependent oligopeptide transporter) may be distantly related to each other, as indicated by the results of PSI-BLAST searches with iterations (96, 125). Only the resistance-nodulation-cell division (RND) family of secondary permeases have been shown to catalyze peptide export.

Nucleobases (entry IV.A in Tables 10 and 11) are taken up by the two possibly related families, NCS1 and NCS2. The proteins of these two families are of similar sizes and topologies, transport similar substrates, and exhibit limited sequence similarity. The multidrug endosomal transporter family includes members that may transport nucleobases into endosomes of animals. Nucleosides (entry IV.B) are transported by H^+ or Na^+ symport or by uniport, and seven families are involved. Only two families of obligatory antiporters (entry IV.C) appear to mediate nucleotide transport.

Vitamins and their precursors (entry V.A in Tables 10 and 11) and intact cofactors (entry V.B) are taken up into cells by cation symport or product antiport, and 12 families have been identified that provide these functions. Two of these families include members that take up both vitamins and intact enzyme or redox cofactors, but two additional families that transport the latter compounds do not transport vitamins or cofactor precursors. One family within the MFS has recently been shown to transport iron-siderophore complexes (72), and three families have been shown to include members that may transport bacterial signaling molecules such as homoserine lactone derivatives (Table 11).

TABLE 9. Classification of outer membrane porins (most from gram-negative bacteria) according to substrate specificity

Substrate	TC no.	Family(-ies)
I.A. Nonselective		
Nonselective	1.B.1, -2, -4, -6, -7, -8, -21, -23–26	GBP, CP, BRP, OOP, RPP, MPP, OmpG, CBP, MBP, Opr, CDP
Nonselective (slight cation selectivity)	1.B.15.1.1	RafY
I.C. Cations		
Fe^{3+} or Fe^{3+} chelate selective	1.B.14.3.2	OMR
Metal ions	1.B.17.2.1	OMF
Metal ions	1.B.17.2.2	OMF
I.D. Anions		
Anion selective	1.B.1.6.1	GBP
Anion selective	1.B.8	MPP
Anion selective	1.B.13	AEP
Phosphate selective	1.B.1.1.2	GBP
Phosphate selective	1.B.5.1.1	POP
Pyrophosphate selective	1.B.5.1.2	POP
II.A. Sugars and polyols		
Glucose	1.B.19.1.1	OprB
Maltooligosaccharide selective	1.B.3.1.1	SP
Oligosaccharide selective	1.B.3.1.2	SP
II.B. Monocarboxylates		
Fatty acid selective	1.B.9.1.1	FadL
II.E. Others		
Toluene selective	1.B.9.2.1	FadL
III.A. Amides		
Amides, short chain	1.B.16.1.1	SAP
Urea	1.B.16.1.1	SAP
IV.B. Nucleosides		
Nucleoside selective	1.B.10.1.1	Tsx
IV.C. Nucleotides		
Nucleotide selective	1.B.1.1.4	GBP
V.A. Vitamins		
Cobalamin selective	1.B.14.3.1	OMR
V.B. Enzyme and redox cofactors		
Heme	1.B.14.2.3	OMR
Porphyrin	1.B.14.3.1	OMR
V.C. Iron-siderophores		
Iron complex selective	1.B.14.1.1–4; 1.B.14.2–3	OMR
Fe^{3+}-catecholate	1.B.14.3.2	OMR
Fe^{3+}-citrate	1.B.14.1.2	OMR
Fe^{3+}-coprogen	1.B.14.1.3	OMR
Fe^{3+}-enterobactin	1.B.14.1.1	OMR
Ferrichrome	1.B.14.1.4	OMR
Heme	1.B.14.2.3	OMR
Heme	1.B.20.3.1	TEC
VI.A. Multiple drugs		
Drug (multiple) selective	1.B.17.1.1	OMF
Drug (multiple) selective	1.B.17.3.2	OMF
Drug (multiple) selective	1.B.17.3.3	OMF
VII.A. Carbohydrates		
Alginate (acidic polysaccharide)	1.B.13.1.1	AEP
Capsular or exopolysaccharide selective	1.B.18.1.1–2; 1.B.18.2.1	OMA
Lipooligosaccharide selective	1.B.17.3.1	OMF
Polymannonate	1.B.13.1.1	AEP
Polysaccharide selective	1.B.13.1.1	AEP
VII.B. Proteins		
Albomycin	1.B.14.1.4	OMR
Colicin I selective	1.B.14.3.2	OMR
Colicin M selective	1.B.14.1.4	OMR
Colicin B selective	1.B.14.1.1	OMR
Colicin D selective	1.B.14.1.1	OMR
Fimbrial subunit selective	1.B.11.1–3	FUP
Hemin selective	1.B.14.2.3	OMR
Hemoglobin selective	1.B.14.2.2	OMR
Protein (virulence factor) selective	1.B.12	AT
Protein (hemolysin) selective	1.B.17.1.1	OMF
Protein (hemolysin) selective	1.B.20.1.2	TEC
Protein (hemagglutinin) selective	1.B.20.2.1	TEC
Protein (proteases) selective	1.B.17.1.2	OMF
Protein (cyclolysin) selective	1.B.17.2.3	OMF
Protein (toxin) selective	1.B.20.1.1	TEC
Transferrin selective	1.B.14.2.1	OMR
Protein (competence factor) selective	1.B.22.1.6	Secretin
Protein (secretory protein) selective	1.B.22.1.1	Secretin
Protein (fimbrial subunit) selective	1.B.22.1.3	Secretin
Protein (virulence factor) selective	1.B.22.1.2; 1.B.22.1.4; 1.B.22.1.5	Secretin
Protein (phage) selective	1.B.22.1.7	Secretin

TABLE 10. Substrate selectivities of secondary carriers

Substrate	No. of families	Families	Total no. of families/category[a]
I.C. Inorganic cations			**30**
Di- and trivalent ions	18	APC, CaCA, CDF, RND, CadD, NiCoT, Ctr1, Ctr2, ZIP, FeT, OFeT, MerTP, MerC, CitMHS, MgtE, APC, Nramp, MFS	**18**
Monovalent ions			
H^+	10	NhaA, NhaB, NhaC, CPA1, CPA2, Trk, NhaD, CPA3, KUP, MC	**13**
K^+	7	CaCA, CCC, CPA1, CPA2, Trk, CPA3, KUP	
Na^+	9	CCC, NhaA, NhaB, NhaC, CPA1, CPA2, Trk, NhaD, CPA3	
NH_4^+ or NH_3	1	AMT	
I.D. Inorganic anions			**16**
Nonselective	2	AE, SSS	
Arsenite/antimonite	2	ArsB, ACR3	
Bicarbonate	1	AE	
Chlorate	1	POT	
Chloride	2	CCC, AE	
Chromate	1	CHR	
Cyanate	1	MFS	
Iodide	1	SSS	
Nitrate/nitrite	3	MFS, POT, FNT	
Phosphate	5	MFS, PiT, MC, DASS, PNaS	
Silicate	1	Sit	
Sulfate	4	DASS, SulP, CHR, MC	
Thiosulfate	1	MC	
Tellurite	1 or 2	TDT, ArsB (?)	
II. Carbon compounds			**28**
II.A. Sugars	4	MFS, RhaT, SSS, GPH	
II.B. Monocarboxylates	14	FNT, MFS, AAAP, AEC, BenE, RND, BASS, OAT, GntP, KDGT, LctP, CCS, SSS, CAAT	
II.C. Di- and tricarboxylates	13	MFS, CitMHS, CCS, DASS, Dcu, TDT, DAACS, MC, TRAP-T, DcuC, MTC, AEC, MSS	
II.D. Organo anions	2	MFS, TP-NST	
III. Amino acids and derivatives			**23**
III.A. Amino acids and conjugates	20	APC, AGC, AAAP, NSS, Dcu, DAACS, MC, LIVCS, ESS, POT, AEC, MFS, BCCT, SSS, ArsB, LCT, HAAAP, LysE, RhtB, CAAT	
III.B. Amines, amides, and polyamines	9	MFS, MC, APC, AAAP, AEC, SSS, BCCT, NSS, CAAT	
III.C. Peptides	5	MFS, POT, OPT, PUP, RND	
IV. Bases and derivatives			**10**
IV.A. Nucleobases	3	NCS1, NCS2, MET	
IV.B. Nucleosides	7	MFS, SSS, CNT, ENT, INT, NCS1, MET	
IV.C. Nucleotides	2	AAA, MC	
V. Vitamins, cofactors, and their precursors			**14**
V.A. Vitamins; vitamin and cofactor precursors	9	AbgT, SSS, FBT, MFS, MC, RFC, NCS1, NCS2, BRT	
V.B. Enzyme and redox cofactors	5	MFS, APC, TP-NST, MC, PnuC	
V.C. Siderophores	1	MFS	
V.D. Signaling molecules	3	RND, OAT, MFS	
VI. Drugs, dyes, sterols, and toxins			**9**
VI.A. Multiple drugs	6	MFS, RND, SMR, MATE, POT, MET	
VI.B. Specific drugs	5	MFS, RND, POT, RFC, OAT	
VI.C. Bile salts and conjugates	3	MFS, RND, BASS	
VI.D. Sterols and conjugates	3	MFS, RND, OAT	
VII. Macromolecules			**4**
VII.A. Complex carbohydrates	2	PST, RND	
VII.B. Proteins	1 or 2	Tat, RND(?)	
VII.C. Lipids	2	RND, AE	

[a] See footnote *a* to Table 5.

Drugs and other toxic substances (entries VI.A to VI.C in Tables 10 and 11) appear to be expelled from cells exclusively by proton antiport, and nine families of secondary carriers appear to mediate these processes. Surprisingly, Na^+ antiport has not been demonstrated for any such system. Three of these families include members that can exhibit a high degree of specificity for a single compound. One additional family, the bile acid:Na^+ symporter family, appears to include members

TABLE 11. Classification of secondary carriers according to substrate specificity[a]

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
I.C. Inorganic cations: monovalent				
Ag^+ (efflux)	2.A.6.1.3	RND	Ag^+:H^+ antiport	B
H^+	2.A.33.1.1	NhaA	H^+:Na^+ antiport	B
H^+	2.A.34.1.1	NhaB	H^+:Na^+ antiport	B
H^+	2.A.35.1.1	NhaC	H^+:Na^+ antiport	B
H^+	2.A.36.1.3–5	CPA1	H^+:Na^+ antiport	B, Y, An, Pl
H^+	2.A.37.1–5	CPA2	H^+:Na^+ antiport	B, Y
H^+	2.A.38.1–4	Trk	H^+:K^+ symport	B, Y, Pl
H^+	2.A.62.1.1	NhaD	H^+:Na^+ antiport	B
H^+	2.A.63.1.1–3	CPA3	H^+:Na^+ or K^+ antiport	B
H^+ or OH^-	2.A.29.3.1	MC	Uniport (?)	Mito
K^+	2.A.19.3.2	CaCA	K^+:Ca^{2+} symport; Na^+ or H^+ antiport	An
K^+	2.A.30.1–3, 7	CCC	NaCl:KCl symport	An, Pl
K^+	2.A.30.5–6	CCC	KCl symport	An
K^+	2.A.36.4.1	CPA1	K^+:H^+ antiport	Y
K^+	2.A.37.1–5	CPA2	K^+:H^+ antiport	B, Y
K^+	2.A.38.1–4	Trk	K^+ uptake (PMF?); K^+:H^+ or Na^+ symport	B, Y, Pl
K^+	2.A.63.1.1	CPA3	K^+:H^+ antiport	B
K^+	2.A.72.1–3	KUP	K^+ uptake (K^+:H^+ or K^+:Na^+ symport)	B, Y, Pl
Na^+	2.A.30.1–3	CCC	$NaKCl_2$ symport	An
Na^+	2.A.30.4.1	CCC	NaCl symport	An
Na^+	2.A.30.7.1	CCC	$NaKCl_2$ symport	Pl
Na^+	2.A.33.1.1	NhaA	Na^+:H^+ antiport	B
Na^+	2.A.34.1.1	NhaB	Na^+:H^+ antiport	B
Na^+	2.A.35.1.1	NhaC	Na^+:H^+ antiport	B
Na^+	2.A.36.1–4	CPA1	Na^+:H^+ antiport	B, Y, An, Pl
Na^+	2.A.37.2.1	CPA2	Na^+:H^+ antiport	B
Na^+	2.A.38.3.1	Trk	Na^+:K^+ symport	Pl
Na^+	2.A.62.1.1	NhaD	Na^+:H^+ antiport	B
Na^+	2.A.63.1.2–3	CPA3	Na^+:H^+ antiport	B
NH_3 or NH_4^+	2.A.49	Amt	Uniport?	B, A, Y, Pl, An
I.C. Inorganic cations: di- and trivalent				
Al^{3+}	9.A.17.2.1	MIT	Uniport?	Y
Ca^{2+}	2.A.19.1–4	CaCA	H^+ or Na^+ antiport	B, Y (vacuolar), Pl, An
Cations, divalent; nonspecific	9.A.17.1–2	MIT	Uniport?	B, Y
Cations, divalent; nonspecific	2.A.3.10.1	APC	?	Y, F
Cations, divalent; nonspecific	2.A.5.1.2	ZIP	PMF (uptake)	Pl
Cations, divalent; nonspecific	2.A.55.1–2	Nramp	H^+ symport	Y, An
Cd^{2+}	2.A.4.1.1	CDF	PMF? (efflux)	B
Cd^{2+}	2.A.4.2.2	CDF	PMF? (uptake)	Mito
Cd^{2+}	2.A.6.1.2	RND	PMF (H^+ antiport)	B
Cd^{2+}	2.A.77.1.1	CadD	PMF (efflux)	B
Co^{2+}	2.A.4.1.1–2	CDF	PMF? (efflux)	B
Co^{2+}	2.A.4.2.1	CDF	PMF? (uptake)	Mito
Co^{2+}	2.A.6.1.1–2	RND	H^+ antiport	B
Co^{2+}	9.A.17.1.1	MIT	PMF? (uptake) (uniport?)	B
Co^{2+}	2.A.52.1.2	NiCoT	PMF? (uptake)	B
Cu^{2+}	9.A.11.1.1	Ctr1	PMF? (uptake)	Y
Cu^{2+}	9.A.12.1.1	Ctr2	PMF? (uptake)	Y, Pr, Pl, An
Fe^{2+}	2.A.5.1.2	ZIP	PMF?	Y, Pl, An
Fe^{2+}	9.A.17.1.1	MIT	PMF? (uptake)	B, A, Y
Fe^{2+}	9.A.9.1.1	FeT	PMF? (uptake)	Y
Fe^{3+}	9.A.10.1.1	OFeT	PMF? (uptake)	B, Y
Hg^{2+}	9.A.2.1.1	MerTP	PMF? (uptake)	B
Hg^{2+}	9.A.3.1.1	MerC	PMF? (uptake)	B
Mg^{2+}	2.A.11.1.1	CitMHS	H^+ symport	B
Mg^{2+}	9.A.17.1.1	MIT	PMF? (uptake)	B
Mg^{2+}	9.A.19.1.1	MgtE	?	B, A
Mn^{2+}	2.A.3.1.15	APC	PMF? (efflux)	Y
Mn^{2+}	2.A.5.2.1	ZIP	PMF (uptake)	Y
Mn^{2+}	2.A.19.2.2	CaCA	PMF (H^+ antiport)	Y
Mn^{2+}	9.A.17.2.2	MIT	PMF? (efflux)	Y
Mn^{2+}	2.A.55.1.1–2	Nramp	H^+ symport (uptake)	B, A, Y, Pr, Pl, An
Ni^{2+}	2.A.1.21.5	MFS	PMF? (efflux)	B
Ni^{2+}	2.A.6.1.1	RND	H^+ antiport	B
Ni^{2+}	9.A.17.1.1	MIT	PMF? (uptake)	B
Ni^{2+}	2.A.52.1.1	NiCoT	Uniport? (uptake)	B
Zn^{2+}	2.A.4.1.1–2	CDF	PMF? (efflux)	B

Continued on following page

TABLE 11—*Continued*

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
Zn^{2+}	2.A.4.2.1–2	CDF	PMF? (uptake)	Mito
Zn^{2+}	2.A.4.2.3	CDF	PMF? (efflux)	An
Zn^{2+}	2.A.4.3.1	CDF	PMF? (uptake)	An vesicles
Zn^{2+}	2.A.5.1.1	ZIP	PMF? (uptake)	Y, Pl, An
Zn^{2+}	2.A.6.1.2	RND	H^+ antiport	B
I.D. Inorganic anions				
Antimonite	2.A.45.1.1	ArsB	PMF (efflux)	B
Arsenite	2.A.45.1.1	ArsB	PMF (efflux)	B
Arsenite	2.A.59.1.1	ACR3	PMF (efflux)	Y
Bicarbonate	2.A.31.1.1	AE	Anion antiport	An, Y
Bicarbonate	2.A.31.2.1	AE	Na^+ symport	An
Bicarbonate	2.A.73.1.1	ICT	H^+ symporter?	B
Chlorate	2.A.17.3.1, -3	POT	H^+ symport	Pl
Chloride	2.A.30.1–3	CCC	Na^+:K^+ symport	An
Chloride	2.A.30.4.1	CCC	Na^+ symport	An
Chloride	2.A.30.5–6	CCC	K^+ symport	An
Chloride	2.A.30.7.1	CCC	Na^+:K^+ symport	Pl
Chloride	2.A.31.1.1	AE	Anion antiport	An, Y
Chromate	2.A.51.1.1	CHR	PMF (efflux)	B
Cyanate	2.A.1.17.1	MFS	H^+ symport	B
Inorganic anions (general)	2.A.31.1.1	AE	Anion antiport	An, Y, F, Pl
Inorganic monovalent anions (nonselective)	2.A.21.5.1	SSS	Na^+ symport	An
Iodide	2.A.21.5.1	SSS	Na^+ symport	An
Nitrate	2.A.1.8.2–6	MFS	H^+ symport	B
Nitrate	2.A.17.3.1, 3	POT	H^+ symport	Pl
Nitrite	2.A.1.8.1	MFS	Uniport; H^+ antiport?	B
Nitrite	2.A.44.3.1	FNT	H^+ symport?	B
Phosphate	2.A.1.4.1–4	MFS	H^+ symport?; anion:anion antiport	B
Phosphate	2.A.1.9.1–3	MFS	H^+ symport	Y, F, Pl
Phosphate	2.A.1.14.6	MFS	Na^+ symport	An
Phosphate	2.A.20.1–3	Pit	H^+ or Na^+ symport	B, A, F
Phosphate	2.A.29.4.1	MC	OH^- antiport	Mito
Phosphate	2.A.47.2.1	DASS	H^+ symport	Y
Phosphate	2.A.58.1.1	PNaS	Na^+ symport	B, An
Silicate	2.A.32.1.1	Sit	Na^+ symport	Diatoms
Sulfate	2.A.29.15.1	MC	SS antiport	Y (mito)
Sulfate	2.A.47.1.2	DASS	Na^+ symport	An
Sulfate	2.A.53.1–4	SulP	H^+ symport or HCO_3^- antiport	B, Y, F, Pl, An
Sulfate	2.A.51.1.2	CHR	H^+ symport?	B
Thiosulfate	2.A.29.15.1	MC	SS antiport	Y (mito)
Tellurite	2.A.16.1.1	TDT	PMF (efflux)	B
II.A. Sugars and polyols				
Arabinitol	2.A.1.18.1	MFS	H^+ symport	B
Arabinose	2.A.1.1.2	MFS	H^+ symport	B
Arabinose	2.A.1.2.14–15	MFS	H^+ antiport	B
Fructose	2.A.1.1.13	MFS	H^+ symport	An
Fructose	2.A.1.1.16	MFS	Uniport	Pr
L-Fucose	2.A.1.7.1	MFS	H^+ symport	B
Galactose	2.A.21.3.1–2	SSS	Na^+ symport	B, An
Galactose	2.A.1.1.1	MFS	H^+ symport	B
Galactose	2.A.1.1.2	MFS	H^+ symport	B
Galactose	2.A.1.1.6	MFS	Uniport	Y
Galactose	2.A.1.1.9	MFS	H^+ symport	Y
Galactose	2.A.1.7.2	MFS	H^+ symport	B
Glucose	2.A.1.1.4	MFS	Uniport	Y
Glucose	2.A.1.1.6	MFS	Uniport	Y
Glucose	2.A.1.1.12	MFS	Uniport	An
Glucose	2.A.1.1.16	MFS	Uniport	Pr
Glucose	2.A.1.1.17	MFS	?	Pr
Glucose	2.A.1.7.2	MFS	H^+ symport	B
Glucose	2.A.1.7.3	MFS	H^+ symport	B
Glucose	2.A.9.2.1	RhaT	H^+ symport	B
Glucose	2.A.21.3.1	SSS	Na^+ symport	An
Glucose	2.A.21.3.1–4	SSS	Na^+ symport	B, An
Glucose (sensor)	2.A.1.1.18–19	MFS	?	Y
α-Glucosides	2.A.1.1.11	MFS	H^+ symport	Y
Glucuronide	2.A.2.3.1	GPH	H^+ symport	B
Hexoses	2.A.1.1.5	MFS	Uniport	Y, An

Continued on following page

TABLE 11—*Continued*

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
Hexoses	2.A.1.1.14	MFS	H^+ symport	Pl
Hexoses	2.A.1.1.16	MFS	Uniport	Pr
Hexuronates	2.A.1.14.2	MFS	H^+ symport	B
Isoprimeverose	2.A.2.3.3	GPH	H^+ symport	B
Isopropyl-β-thiogalactoside (efflux)	2.A.1.20.1, -4	MFS	H^+ antiport	B
Isopropyl-β-thiogalactoside (efflux)	2.A.1.2.15, -18	MFS	H^+ antiport	B
Lactose	2.A.1.1.9	MFS	H^+ symport	Y
Lactose (efflux)	2.A.1.20.1, -2 and 4	MFS	H^+ antiport	B
Lactose	2.A.1.5.1	MFS	H^+ symport	B
Lactose	2.A.2.2.1	GPH	H^+ symport	B
Maltose	2.A.1.1.10	MFS	H^+ symport	Y
Maltose (efflux)	2.A.1.20.1	MFS	H^+ antiport	B
Mannose	2.A.1.1.16	MFS	Uniport	Pr
Mannose	2.A.1.7.3	MFS	H^+ symport	B
Melibiose	2.A.1.5.4	MFS	H^+ symport	B
Melibiose (efflux)	2.A.1.2.18	MFS	H^+ antiport	B
Melibiose	2.A.2.1.1	GPH	H^+ or Na^+ symport	B
Multiple sugars	2.A.1.20	MFS	H^+ antiport	
Myoinositol	2.A.1.1.8	MFS	H^+ symport	Y
Myoinositol	2.A.1.1.20	MFS	H^+ symport	Pr
Myoinositol	2.A.21.4.1	SSS	Na^+ symport	An
Pentosides	2.A.2.3.2–3	GPH	H^+ symport	B
Raffinose	2.A.1.5.2	MFS	H^+ symport	B
Raffinose	2.A.2.2.2	GPH	H^+ symport	B
L-Rhamnose	2.A.9.1.1	RhaT	H^+ symport	B
Ribitol	2.A.1.18.2	MFS	H^+ symport	B
Ribose	2.A.9.3.1	RhaT	H^+ symport	B
Sialic acid	2.A.1.12.1	MFS	H^+ symport	B
Sucrose	2.A.1.5.3	MFS	H^+ symport	B
Sucrose	2.A.2.4.1	GPH	H^+ symport	Pl
Sugars (efflux) (fairly nonspecific)	2.A.1.20.1, -2, -4	MFS	H^+ antiport	B
Xylose	2.A.1.1.2	MFS	H^+ symport	B
Xylose	2.A.1.1.3	MFS	H^+ symport	B
II.B. Monocarboxylates				
Acetate	2.A.44.4.1	FNT	H^+ symport	Y
Allantoate	2.A.1.14.4	MFS	H^+ symport	Y
Auxin (indole 3-acetate)	2.A.18.1.1	AAAP	H^+ symport	Pl
Auxin (out)	2.A.69.1.1	AEC	Uniport? H^+ antiport?	Pl
Benzoate	2.A.1.15.5	MFS	H^+ symport	B
Benzoate	2.A.46.1.1	BenE	H^+ symport	B
Bile salts	2.A.1.3.13	MFS	H^+ symport	B
Bile salts	2.A.6.2.5	RND	H^+ antiport	B
Bile salts	2.A.28.1.1	BASS	Na^+ symport	An
Bile salts (conjugated)	2.A.1.23.1	MFS	H^+ symport	B
Bile salts (conjugated and unconjugated)	2.A.60.1	OAT	?	An
Bilirubin	2.A.65.1.1	BRT	H^+ symport?	An
Cyanate	2.A.1.17.1	MFS	H^+ symport	B
2,4-Dichlorophenoxy acetate	2.A.1.15.3	MFS	H^+ symport	B
Digoxin	2.A.60.1.1	OAT	Uniport; anion antiport	An
Fatty acids	2.A.6.2.5	RND	H^+ antiport	B
Formate	2.A.1.11.1	MFS	SS antiport	B
Formate	2.A.44.1.1	FNT	Efflux?	B
Formate	2.A.44.2.1	FNT	Uptake?	B
Galacturonate	2.A.1.14.2	MFS	H^+ symport	B
Glucarate	2.A.1.14.1	MFS	H^+ symport	B
Gluconate	2.A.8.1.1	GntP	H^+ symport	B
Gluconate	2.A.8.1.2	GntP	H^+ symport	B
Glucuronate	2.A.1.14.2	MFS	H^+ symport	B
Hexuronate	2.A.1.14.2	MFS	H^+ symport	B
4-Hydroxybenzoate	2.A.1.15.1	MFS	H^+ symport	B
3-Hydroxyphenyl acetate	2.A.1.14.9	MFS	H^+ symport	B
3-Hydroxyphenyl propionate	2.A.1.15.2	MFS	H^+ symport	B
L-Idonate	2.A.8.1.2	GntP	H^+ symport	B
2-Keto-3-deoxygluconate	2.A.10.1.1	KDGT	H^+ symport	B
Lactate	2.A.1.12.2	MFS	H^+ symport	Y
Lactate	2.A.1.13.1	MFS	H^+ symport	Y, F, An
Lactate	2.A.14.1.1	LctP	H^+ symport	B
Lactate	2.A.24.3.2	CCS	SS antiport	B

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TABLE 11—*Continued*

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
Leucotrienes	2.A.60.1.7	OAT	?	An
4-Methyl-*o*-phthalate	2.A.1.6.5	MFS	H^+ symport	B
Mevalonate	2.A.1.13.1	MFS	H^+ symport	Y, F, An
Monocarboxylates	2.A.1.13.1	MFS	H^+ symport	Y, F, An
Organic anions (nonselective)	2.A.1.19.4, -7, -8	MFS	H^+ symport?	An
cis,*cis*-Muconate	2.A.1.15.4	MFS	H^+ symport	B
Organic anions (nonselective)	2.A.1.19.4, -7, -8	MFS	H^+ symport?	An
Organic anions (nonselective)	2.A.21.5.2	SSS	Na^+ symport	An
Organic anions (nonselective)	2.A.60.1	OAT	Uniport; anion antiport	An
Organic anions (nonselective)	2.A.65.1.1	BRT	H^+ symport?	An
Phenylpropionate	2.A.1.27.1	MFS	H^+ symport	B
Phenylacetate	2.A.21.7.1	SSS	H^+ symport	B
Phthalate	2.A.1.14.5	MFS	H^+ symport	B
Prostaglandins	2.A.60.1.2, -7	OAT	Uniport; anion antiport	An
Protocatechuate	2.A.1.15.1	MFS	H^+ symport	B
Pyruvate	2.A.1.12.1	MFS	H^+ symport	Y
Pyruvate	2.A.1.13.1	MFS	H^+ symport	Y, F, An
Quinate	2.A.1.1.7	MFS	H^+ symport	F
Shikimate	2.A.1.6.6	MFS	H^+ symport	B
Sialate	2.A.1.12.1	MFS	H^+ symport	B
Tartrate	2.A.1.14.3	MFS	H^+ symport	B
Tartrate	2.A.47.3.3	DASS	SS antiport	B
Taurocholate	2.A.60.1.3, -7	OAT	?	An
Thromboxanes	2.A.60.1.2, -7	OAT	?	An
Thyroid hormones (thyroxin; triiodothyronine)	2.A.60.1.3	OAT	?	An
II.C. Di- and tricarboxylates				
Citrate	2.A.1.6.1	MFS	H^+ symport	B
Citrate	2.A.11.1.1	CitMHS	Mg^{2+}:H^+ symport	B
Citrate	2.A.11.1.2	CitMHS	H^+ symport	B
Citrate	2.A.24.1.1; 3.1, 3.2	CCS	Na^+ symport	B
Citrate	2.A.47.1.3, 3.2	DASS	Na^+ symport; SS antiport	B, An
Dicarboxylates	2.A.1.6.3	MFS	H^+ symport	B
Dicarboxylates	2.A.13.1.1–2	Dcu	SS antiport	B
Dicarboxylates	2.A.16.2.1	TDT	H^+ symport	Y
Dicarboxylates	2.A.23.1.3	DAACS	H^+ symport	B
Dicarboxylates	2.A.29.2.1–2; 13.1	MC	SS antiport	Mito
Dicarboxylates	2.A.47.1.1, -3	DASS	Na^+ symport	An
Dicarboxylates	2.A.47.3.1–3	DASS	SS antiport	B, chloro
Dicarboxylates	2.A.56.1.1	TRAP-T	H^+ symport	B
Dicarboxylates	2.A.61.1.1	DcuC	H^+ symport	B
α-Ketoglutarate	2.A.1.6.2	MFS	H^+ symport	B
Malate	2.A.16.2.1	TDT	H^+ symport?	Y
Malate	2.A.24.2.1	CCS	H^+ symport	B
Malate	2.A.69.3.1	AEC	H^+ symport?	B
Malonate	2.A.29.15.1	MC	SS antiport	Y (mito)
Malonate	2.A.69.2.1	AEC	H^+ symport?	B
Malonate	2.A.70.2.1	MSS	H^+ symport?	B
Malonate	2.A.70.1.1	MSS	Na^+ symport	B
Oxalate	2.A.1.11.1	MFS	SS antiport	B
Oxaloacetate	2.A.29.15.1	MC	SS antiport	Y (mito)
Tricarboxylates	2.A.29.7.1	MC	SS antiport	Mito
Tricarboxylates	2.A.54.1.1	MTC	H^+ symport	Mito
II.D. Organic anions (noncarboxylates)				
Glucose-phosphate	2.A.1.4.1	MFS	P_i antiport	B
Glucose 6-phosphate (sensor)	2.A.1.4.4	MFS	?	B
Glucose 6-phosphate	2.A.1.4.5	MFS	P_i antiport	An
Glucose 6-phosphate	2.A.50.2.1	TP-NST	P_i antiport	Plastids (Pl)
Phosphoglycerates	2.A.1.4.2	MFS	P_i antiport	B
Glycerolphosphate	2.A.1.4.3	MFS	P_i antiport	B
Hexosephosphates	2.A.1.4.1	MFS	P_i antiport	B
Phosphoenolpyruvate	2.A.1.4.2	MFS	P_i antiport	B
Phosphoenolpyruvate	2.A.50.3.1	TP-NST	P_i antiport	Chloro (Pl)
Sugar-phosphates	2.A.1.4.1	MFS	P_i antiport	B
Triosephosphate	2.A.50.1.1	TP-NST	P_i antiport	Chloro (Pl)
Triosephosphate	2.A.50.2.1	TP-NST	P_i antiport	Plastids (Pl)
III.A. Amino acids and conjugates				
Acidic amino acids	2.A.3.10.13	APC	H^+ symport	Y
Alanine	2.A.3.1.7	APC	H^+ symport	B

Continued on following page

TABLE 11—*Continued*

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
Alanine	2.A.25.1.1	AGCS	Na^+ symport	B
Alanine	2.A.25.1.2	AGCS	Na^+ or H^+ symport	B
β-Alanine	2.A.3.1.5	APC	H^+ symport	B
γ-Aminobutyrate	2.A.3.1.4	APC	H^+ symport	B
γ-Aminobutyrate	2.A.3.1.5	APC	H^+ symport	B
γ-Aminobutyrate	2.A.3.7.1	APC	Glutamate antiport	B
γ-Aminobutyrate	2.A.18.5.1	AAAP	H^+ symport?	An
γ-Aminobutyrate	2.A.22.3.1–2	NSS	Na^+ symport	An
Arginine	2.A.3.10.4	APC	H^+ symport	Y
Arginine	2.A.3.2.3	APC	Antiport with ornithine	B
Arginine	2.A.65.1.1	LysE	H^+ antiport?	B
Aromatic amino acids	2.A.3.1.3	APC	H^+ symport	B
Asparagine	2.A.3.1.8	APC	H^+ symport	B
Asparagine	2.A.3.10.7, -13	APC	H^+ symport	Y
Aspartate	2.A.13.1.1	Dcu	SS antiport	B
Aspartate	2.A.23.1.1	DAACS	H^+ symport	B
Aspartate	2.A.23.1.2	DAACS	H^+ or Na^+ symport	B, An
Aspartate	2.A.23.2.1	DAACS	H^+ or Na^+ symport	B, An
Basic amino acids	2.A.3.1.19	APC	H^+ symport	Y
Basic amino acids	2.A.3.3.1, -2	APC	H^+ symport	An
Basic amino acids	2.A.3.8.3	APC	Uniport or H^+ symport	An
Basic amino acids	2.A.29.9.1	MC	SS antiport	Mito
Branched-chain amino acids	2.A.3.10.11	APC	H^+ symport	Y
Branched-chain amino acids	2.A.26.1.1–3	LIVCS	H^+ or Na^+ symport	B
Cystine	2.A.3.8.5	APC	Glutamate antiport	An
Cystine	2.A.43.1.1	LCT	H^+ antiport	An
General amino acids (all L, some D)	2.A.3.3.3	APC	H^+ symport	Pl
General amino acids	2.A.3.10.2, -14	APC	H^+ symport	Y
General amino acids	2.A.18.2.1	AAAP	H^+ symport	Pl
Glutamate	2.A.3.7.1	APC	γ-Aminobutyrate antiport	B
Glutamate	2.A.3.8.5	APC	Cystine antiport	An
Glutamate	2.A.3.10.13	APC	H^+ symport	Y
Glutamate	2.A.23.1.1	DAACS	H^+ symport	B
Glutamate	2.A.23.1.2	DAACS	H^+ or Na^+ symport	B, An
Glutamate	2.A.23.2.1	DAACS	H^+ or Na^+ symport	B, An
Glutamate	2.A.23.2.2	DAACS	Na^+ symport	An
Glutamate	2.A.27.1.1	ESS	Na^+ symport	B
Glutamine	2.A.3.7.1	APC	H^+ symport	B
Glutamine	2.A.3.10.5, -7	APC	H^+ symport	Y
Glycine	2.A.3.1.7	APC	H^+ symport	B
Glycine	2.A.22.2.2	NSS	Na^+ symport	An
Glycine	2.A.25.1.1	AGCS	Na^+ symport	B
Histidine	2.A.3.1.5, -9	APC	H^+ symport	Y, F
Histidine	2.A.3.10.1	APC	H^+ symport	Y
Histidine	2.A.17.3.2–3	POT	H^+ symport	Pl
Homoserine (efflux)	2.A.76.1.1	RhtB	H^+ antiport	B
Homoserine lactone (efflux)	2.A.76.1.1	RhtB	H^+ antiport	B
Isoleucine	2.A.3.10.6	APC	H^+ symport	Y
Isoleucine	2.A.26.1.1–3	LIVCS	H^+ or Na^+ symport	B
Leucine	2.A.3.10.6	APC	H^+ symport	Y
Leucine (sensor)	2.A.3.10.12	APC	?	Y
Leucine	2.A.26.1.1–3	LIVCS	H^+ or Na^+ symport	B
Lysine	2.A.3.1.2	APC	H^+ symport	B
Lysine	2.A.3.2.2	APC	SS antiport	B
Lysine	2.A.3.2.4	APC	?	B
Lysine	2.A.3.10.10	APC	H^+ symport	Y
Lysine (efflux)	2.A.75.1.1	LysE	H^+ antiport?	B
Methionine	2.A.3.8.4	APC	H^+ symport	Y
Neutral amino acids	2.A.3.8.1–3, -6	APC	Uniport or H^+ symport	An
Neutral amino acids	2.A.18.4.1	AAAP	H^+ symport	F
Neutral amino acids	2.A.23.3.1	DAACS	Na^+ symport	An
Neutral and acidic amino acids	2.A.23.3.2	DAACS	Na^+ symport	An
Neutral and basic amino acids	2.A.23.3.3	DAACS	Na^+ symport	An
Ornithine	2.A.3.2.1	APC	Antiport with putrescine	B
Ornithine	2.A.3.2.3	APC	Antiport with arginine	B
Phenylalanine	2.A.3.1.1	APC	H^+ symport	B
Proline	2.A.1.6.4	MFS	H^+ symport	B
Proline	2.A.3.1.6	APC	H^+ symport	Y

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TABLE 11—*Continued*

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
Proline	2.A.3.10.3	APC	H^+ symport	Y
Proline	2.A.15.1.2	BCCT	H^+ symport	B
Proline	2.A.18.3.1	AAAP	H^+ symport	Pl
Proline	2.A.21.2.1	SSS	Na^+ symport	B
Proline	2.A.22.2.1	NSS	Na^+ symport	An
Serine	2.A.3.1.7	APC	H^+ symport	B
Serine	2.A.23.4.1	DAACS	Na^+ symport	B
Serine	2.A.42.2.1	HAAAP	H^+ symport	B
Threonine	2.A.3.1.9	APC	H^+ symport	B
Threonine	2.A.23.4.1	DAACS	Na^+ symport	B
Threonine	2.A.42.2.2	HAAAP	H^+ symport	B
Threonine (efflux)	2.A.76.1.2	RhtB	H^+ antiport?	B
Tryptophan	2.A.3.1.16–17	APC	H^+ symport	Y
Tryptophan	2.A.22.4.1	SSS	Na^+ symport	B
Tryptophan	2.A.42.1.2–3	HAAAP	H^+ symport	B
Tyrosine	2.A.3.10.8, -9	APC	H^+ symport	Y
Tyrosine	2.A.42.1.1	HAAAP	H^+ symport	B
Tyrosine	2.A.45.2.1	ArsB	?	An
Valine	2.A.3.10.6, -9	APC	H^+ symport	Y
Valine	2.A.26.1.1–3	LIVCS	H^+ or Na^+ symport	B
III.B. Amines, amides, and polyamines				
Acetylcholine	2.A.1.2.13	MFS	H^+ antiport	An
Acylcarnitine	2.A.1.19.3	MFS	Uniport; Na^+ symport	An
S-Adenosylmethionine	2.A.3.10.15	APC	H^+ symport	Y
Acylcarnitine	2.A.29.8.1	MC	SS antiport	Mito
Amines, organic (nonspecific)	2.A.1.19.1–5	MFS	Uniport?	An
γ-Aminobutyrate	2.A.3.4.2	APC	H^+ symport	F, Y
γ-Aminobutyrate	2.A.3.7.1	APC	H^+ symport	B
Auxin	2.A.18.1.1	AAAP	H^+ symport	Pl
Auxin	2.A.69.1.1–2	AEC	Uniport; H^+ antiport?	Pl
Betaine	2.A.1.6.4	MFS	H^+/Na^+ symport	B
Betaine	2.A.23.3.1	SSS	Na^+ symport	An
Butyrobetaine	2.A.1.19.3	MFS	Uniport; Na^+ symport	An
Cadaverine	2.A.3.2.2	APC	Antiport with lysine	B
Carnitine	2.A.1.19.3	MFS	Uniport; Na^+ symport	An
Carnitine	2.A.15.2.1	BCCT	H^+ symport	B
Carnitine	2.A.29.8.1	MC	SS antiport	Mito
Choline	2.A.3.4.1	APC	H^+ symport?	Y
Choline	2.A.15.3.1	BCCT	H^+ symport	B
Choline	2.A.22.3.5	NSS	Na^+ symport	An
Creatine	2.A.22.3.4	NSS	Na^+ symport	An
Dopamine	2.A.1.22.1	MFS	?	An
Dopamine	2.A.22.1.3	NSS	Na^+ symport	An
Ectosine	2.A.15.1.2	BCCT	H^+ symport	B
Ethanolamine	2.A.3.5.1	APC	H^+ symport	B
Glycine betaine	2.A.1.6.4	MFS	H^+/Na^+ symport	B
Glycine betaine	2.A.1.19.3	MFS	Uniport; Na^+ symport	An
Glycine betaine	2.A.15.1.1–2	BCCT	H^+ symport	B
Glycine betaine	2.A.22.3.1	NSS	Na^+ symport	An
Guanidinium	2.A.1.19.2	MFS	H^+ antiport	An
Monoamines	2.A.1.2.11	MFS	H^+ antiport	An
Monoamines	2.A.1.2.12	MFS	H^+ antiport	An
Neurotransmitters (cationic)	2.A.1.19.1	MFS	Uniport?	An
Neurotransmitters (cationic)	2.A.1.19.5–7	MFS	Uniport?	An
Neurotransmitters (cationic)	2.A.1.22.1	MFS	Uniport or H^+ or Na^+ symport	An
Noradrenaline	2.A.22.1.2	NSS	Na^+ symport	An
Organocations	2.A.1.19.1–5	MFS	Uniport or H^+ or Na^+ symport	An
Polyamines (efflux)	2.A.1.2.16	MFS	H^+ antiport	An
Putrescine	2.A.3.2.1	APC	H^+ symport; antiport with ornithine	B
Quaternary amines	2.A.77.1.2	CadD	H^+ antiport?	B
Serotonin	2.A.22.1.1	NSS	Na^+ symport	An
Taurine	2.A.22.3.3	NSS	Na^+ symport	An
Tetramethyl ammonium	2.A.1.19.1	MFS	H^+ antiport; uniport?	An
Urea	2.A.21.6.1	SSS	?	Y
III.C. Peptides				
Glycopeptides, cell wall derived	2.A.1.25.2	MFS	H^+ symport?	B
Peptides	2.A.17.1–4	POT	H^+ symport	B, Y, Pl, An
Peptides	2.A.67.1–2	OPT	H^+ symport?	B, A, Y, Pl

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TABLE 11—*Continued*

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
Peptides	9.A.18	PUP	?	B
Peptides, antibacterial (efflux)	2.A.6.2.5	RND	H^+ antiport	B
Peptides, cell wall derived	2.A.1.25.2	MFS	H^+ symport?	B
IV.A. Nucleobases				
Allantoin	2.A.39.3.1	NCS1	H^+ symport	Y
Cytosine	2.A.39.1.1	NCS1	H^+ symport	B
Cytosine	2.A.39.2.1	NCS1	H^+ symport	Y
Nucleobases	2.A.74.1.1	MET	H^+ antiport?	An
Purines	2.A.39.2.1	NCS1	H^+ symport	Y
Purines	2.A.40.2.1	NCS2	H^+ symport	B
Purines	2.A.40.5.1	NCS2	H^+ symport	F
Uracil	2.A.39.3.2	NCS1	H^+ symport	Y
Uracil	2.A.40.1.1	NCS2	H^+ symport	B
Uric acid	2.A.40.4.1	NCS2	H^+ symport	F
Xanthine	2.A.40.3.1	NCS2	H^+ symport	B
Xanthine	2.A.40.4.1	NCS2	H^+ symport	F
IV.B. Nucleosides				
Nucleosides	2.A.1.10.1	MFS	H^+ symport	B
Nucleosides	2.A.21.3.3	SSS	Na^+ symport	An
Nucleosides	2.A.41.1.1	CNT	H^+ symport	B
Nucleosides	2.A.41.2.2–5	CNT	Na^+ symport	An
Nucleosides (pyrimidine specific)	2.A.41.1.2	CNT	Na^+ symport	B
Nucleosides (purine specific)	2.A.41.2.1	CNT	Na^+ symport	An
Nucleosides	2.A.57.1–3	ENT	Uniport	Y, Pr, An
Nucleosides	2.A.74.1.1	MET	H^+ antiport?	An
Nucleosides	9.A.6.1.1	INT	?	An
Uridine	2.A.39.3.2–3	NCS1	H^+ symport	Y
Xanthosine	2.A.1.10.2	MFS	H^+ symport	B
IV.C. Nucleotides				
ADP	2.A.12.1.1, -2	AAA	SS antiport	B
ADP	2.A.12.2.1	AAA	SS antiport	Pl
ADP	2.A.29.1.1	MC	SS antiport	Mito
ATP	2.A.12.1.1	AAA	SS antiport	B
ATP	2.A.12.1.2	AAA	SS antiport	Pl
ATP	2.A.29.1.1	MC	SS antiport	Mito
Nucleoside triphosphates	2.A.12.3.1	AAA	H^+ symport	B
V.A. Vitamins/cofactor precursors				
L-Ascorbate	2.A.40.6.1	NCS2	Na^+ symport	An
p-Aminobenzoylglutamate	2.A.68.1.1	AbgT	H^+ symport	B
Anionic vitamins	2.A.21.5.2	SSS	Na^+ symport	An
Biopterin	2.A.71.1.1–4	FBT	H^+ symport?	Pr
Biotin	2.A.21.5.2	SSS	Na^+ symport	An
Cationic vitamins	2.A.1.19.1	MFS	Uniport	An
Flavin adenine dinucleotide	2.A.29.10.1	MC	?	Mito
Folate (reduced)	2.A.48.1.1	RFC	H^+ symport	An
Folate	2.A.71.1.1–4	FBT	H^+ symport?	Pr
5-Formyltetrahydrofolate	2.A.48.1.1	RFC	Na^+ symport?	An
Lipoate	2.A.21.5.2	SSS	Na^+ symport	An
Nicotinate	2.A.65.1.1	BRT	H^+ symport	An
Pantothenate	2.A.21.1.1	SSS	Na^+ symport	B
Pantothenate	2.A.21.5.2	SSS	Na^+ symport	An
Tetrahydrofolate	2.A.48.1.1	RFC	Na^+ symport?	An
Thiamine	2.A.39.4.1	NCS1	H^+ symport	Y
Thiamine	2.A.48.2.1	RFC	Na^+ symport?	An
V.B. Enzyme and redox cofactors				
Acetyl coenzyme A	2.A.1.25.1	MFS	SS antiport	An (ER)
S-Adenosylmethionine	2.A.3.10.15	APC	H^+ symport?	Y
CMP-sialate	2.A.50.7.1	TP-NST	CMP antiport	An (ER)
Coenzyme A	2.A.1.25.1	MFS	SS antiport	An (ER)
Flavin adenine dinucleotide	2.A.29.10.1	MC	?	Mito
GDP-mannose	2.A.50.8.1	TP-NST	GMP antiport	An, Y (ER)
Nicotinamide mononucleotide	9.A.4.1.1	PnuC	?	B
UDP-*N*-acetylglucosamine	2.A.50.5.1	TP-NST	UMP antiport	Y, An (ER)
UDP-galactose	2.A.50.6.1	TP-NST	UMP antiport	An (ER)
V.C. Siderophores and Fe-siderophore complexes				
Fe-enterobactin	2.A.1.16.2	MFS	H^+ symport?	Y
Fe-ferrioxamine	2.A.1.16.1	MFS	H^+ symport?	Y
Fe-siderophore	2.A.1.16.1	MFS	H^+ symport?	Y

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TABLE 11—*Continued*

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
Fe-triacetylfusarinine	2.A.1.16.3	MFS	H^+ symport?	Y
V.D. Signaling molecules				
Acyl L-homoserine lactone	2.A.6.2.6	RND	H^+ antiport	B
Isoflavenoids	2.A.6.2.3	RND	H^+ antiport	B
Nodulation factor	2.A.6.3.1	RND	H^+ antiport	B
Prostaglandins	2.A.60.1.2	OAT		An
Steroid hormones	2.A.1.19.4	MFS		An
Steroid hormones	2.A.6.6	RND		An, Y
Thyroid hormones (thyroxine, triiodothyronine)	2.A.60.1.3, -7	OAT		An
VI.A. Multiple drugs and dyes				
Multiple drugs	2.A.1.2.5	MFS	H^+ antiport	B
Multiple drugs	2.A.1.2.6	MFS	H^+ antiport	Y
Multiple drugs	2.A.1.2.7	MFS	H^+ antiport	B
Multiple drugs	2.A.1.2.8	MFS	H^+ antiport	B
Multiple drugs	2.A.1.2.9	MFS	H^+ antiport	B
Multiple drugs	2.A.1.2.10	MFS	H^+ antiport	B
Multiple drugs	2.A.1.2.11–12	MFS	H^+ antiport	An
Multiple drugs	2.A.1.2.16, -17	MFS	H^+ antiport	An
Multiple drugs	2.A.1.3.1	MFS	H^+ antiport	Y
Multiple drugs	2.A.1.3.2	MFS	H^+ antiport	B
Multiple drugs	2.A.1.3.3	MFS	H^+ antiport	B
Multiple drugs	2.A.1.3.4	MFS	H^+ antiport	B
Multiple drugs	2.A.1.3.5	MFS	H^+ antiport	B
Multiple drugs (cationic)	2.A.1.19.1	MFS	Uniport	An
Multiple drugs (aminoglycosides)	2.A.1.20.1	MFS	H^+ antiport?	B
Multiple drugs	2.A.1.21.1–4	MFS	H^+ antiport	B
Multiple drugs	2.A.6.2.1–2, -6	RND	H^+ antiport	B
Multiple drugs and dyes (cationic)	2.A.7.1.1–2	SMR	H^+ antiport	B
Multiple drugs (antibiotics) (uptake)	2.A.17.4.1	POT	H^+ symport	An
Multiple drugs (antibiotics) (uptake)	2.A.1.25.2	MFS	H^+ symport	B
Multiple drugs (antibiotics)	2.A.66.1–3	MATE	H^+ antiport	B, Y
Multiple drugs (antibiotics)	2.A.74.1.1–2	MET	H^+ antiport?	An
Xenobiotics	2.A.1.19.1	MFS	Uniport	An
VI.B. Specific drugs				
Acriflavin	2.A.1.2.8	MFS	H^+ antiport	B
Acriflavin	2.A.1.3.3	MFS	H^+ antiport	B
Actinorhordin	2.A.1.3.7	MFS	H^+ antiport	B
Actinorhordin	2.A.6.5.1	RND	H^+ antiport	B
Amiloride	2.A.1.2.1	MFS	H^+ antiport	Y
Aminoglycosides	2.A.1.20.1	MFS	H^+ antiport?	B
Aminotriazole	2.A.1.3.1	MFS	H^+ antiport	Y
Ampicillin (in)	2.A.1.25.2	MFS	H^+ symport?	B
Benomyl	2.A.1.2.6	MFS	H^+ antiport	Y
Bicyclomycin	2.A.1.2.7	MFS	H^+ antiport	B
Bile acids	2.A.1.3.13	MFS	H^+ antiport	B
Bleomycin	2.A.1.21.2	MFS	H^+ antiport	B
Carbonylcyanide *m*-chlorophenylhydrazone (CCCP)	2.A.1.2.9	MFS	H^+ antiport	B
CCCP	2.A.1.3.2	MFS	H^+ antiport	B
Cephamycin	2.A.1.3.8	MFS	H^+ antiport	B
Chloramphenicol	2.A.1.2.3	MFS	H^+ antiport	B
Chloramphenicol	2.A.1.2.8	MFS	H^+ antiport	B
Cycloheximide	2.A.1.2.2	MFS	H^+ antiport	Y
Cycloheximide	2.A.1.2.6	MFS	H^+ antiport	Y
Daunomycin	2.A.1.2.5	MFS	H^+ antiport	B
Dauxorubicin	2.A.1.2.11	MFS	H^+ antiport	An
Erythromycin	2.A.1.21.1	MFS	H^+ antiport	B
Ethidium bromide	2.A.1.2.5	MFS	H^+ antiport	B
Ethidium bromide	2.A.1.2.8	MFS	H^+ antiport	B
Ethidium bromide	2.A.1.2.11	MFS	H^+ antiport	An
Ethidium bromide	2.A.1.3.3	MFS	H^+ antiport	B
Ethidium bromide	2.A.66.1.1	MATE	H^+ antiport	B
Ethionine	2.A.66.3.1	MATE	H^+ antiport	Y
Fluconazole	2.A.1.2.17	MFS	H^+ antiport	Y
Fluoroquinolones	2.A.1.2.8	MFS	H^+ antiport	B
Fluoroquinolones	2.A.1.3.3	MFS	H^+ antiport	B
Hydrophobic uncouplers	2.A.1.2.9	MFS	H^+ antiport	B
Isoflavinoid	2.A.6.2.3	RND	H^+ antiport	B
Kanamycin	2.A.1.20.1	MFS	H^+ antiport?	B

Continued on following page

TABLE 11—*Continued*

Substrate	TC no.	Family	Energy source or mechanism	Organismal group
Kanamycin	2.A.66.1.1	MATE	H^+ antiport	B
β-Lactams (uptake)	2.A.17.4.1	POT	H^+ symport	An
Lincomycin	2.A.1.3.9	MFS	H^+ antiport	B
Macrolides	2.A.1.21.1	MFS	H^+ antiport	B
Methotrexate	2.A.1.2.6	MFS	H^+ antiport	Y
Methotrexate	2.A.48.1.1	RFC	Na^+ symport?	An
Methotrexate	2.A.60.1.4	OAT	?	An
Methylenomycin	2.A.1.3.10	MFS	H^+ antiport	B
Methylviologen	2.A.1.3.14	MFS	H^+ antiport	B
Nalidixic acid	2.A.1.3.2	MFS	H^+ antiport	B
Nitroquinoline-*N*-oxide	2.A.1.3.1	MFS	H^+ antiport	Y
Norfloxacin	2.A.66.1.1	MATE	H^+ antiport	B
Oleandomycin	2.A.1.21.1	MFS	H^+ antiport	B
Organocations	2.A.1.3.4	MFS	H^+ antiport	B
Organomercurials	2.A.1.3.2	MFS	H^+ antiport	B
Paraquat	2.A.1.2.16	MFS	H^+ antiport	Y
Penicillin and derivatives (in)	2.A.1.25.2	MFS	H^+ symport	B
Pristinamycin	2.A.1.3.5	MFS	H^+ antiport	B
Puromycin	2.A.1.3.11	MFS	H^+ antiport	B
Puromycin	2.A.1.21.2	MFS	H^+ antiport	B
Quinoline	2.A.1.2.10	MFS	H^+ antiport	B
Rifamycin	2.A.1.3.5	MFS	H^+ antiport	B
Rifamycin	2.A.1.3.15	MFS	H^+ antiport	B
Streptomycin	2.A.1.20.1	MFS	H^+ antiport?	B
Streptomycin	2.A.66.1.1	MATE	H^+ antiport	B
Sulfathiazole	2.A.1.27	MFS	H^+ antiport	B
Tetracenomycin	2.A.1.3.12	MFS	H^+ antiport	B
Tetracycline	2.A.1.2.4	MFS	H^+ antiport	B
Tetracycline	2.A.1.3.6	MFS	H^+ antiport	B
Tetracycline	2.A.1.21.2–3	MFS	H^+ antiport	B
VI.C. Bile salts and conjugates				
Bile salts	2.A.1.19.4	MFS	Uniport or H^+ symport	An
Bile salts	2.A.6.2.5	RND	H^+ antiport	B
Bile salts	2.A.28.1.1	BASS	Na^+ symport	An
Bile salt conjugates	2.A.1.23.1	MFS	H^+ symport	B
Organic solvents	2.A.6.2.4	RND	H^+ antiport	B
Toluene	2.A.6.2.4	RND	H^+ antiport	B
VI.D. Sterols/steroids				
Steroids	2.A.6.2.5	RND	H^+ antiport	B
Sterols (nonspecific)	2.A.1.19.4	MFS	Uniport; H^+ symport	An
Sterols (nonspecific; probable)	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Sterols (nonspecific)	2.A.74.1.1	MET	H^+ antiport?	An
Aldosterone	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Cardiac glycosides	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Cortisol	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Dexamethasone	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Estrone-3-sulfate	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Estradiol 17-glucuronide	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Ouabain	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Steroid conjugates	2.A.6.6.1, -3	RND	H^+ antiport?	Y, An
Steroid hormones (conjugated and unconjugated)	2.A.60.1	OAT	?	An
VII.A. Carbohydrates				
Lipooligosaccharides	2.A.6.3.1	RND	H^+ antiport?	B
Exo- and capsular polysaccharides (out)	9.A.1.2.1–2	PST	H^+ antiport?	B
Lipopolysaccharide (out)	9.A.1.1	PST	H^+ antiport?	B
VII.B. Proteins				
Proteins (nonspecific)	2.A.6.4.1	RND (SecDF)	H^+ antiport?	B, A
Redox enzymes/proteins	2.A.64.1.1	TAT	PMF driven	B, A, E
VII.D. Lipids				
Lipids	2.A.6.5.2	RND	PMF driven	B
Lipids	2.A.31.1.1	AE	?	An

[a] Abbreviations are defined in Table 3, footnote *d*, and in Table 2. ER, endoplasmic reticulum; SS antiport, solute/solute antiport.

that transport bile salts but not drugs. Two families, the proton-dependent oligopeptide transporter family and the MFS, include members that have been shown to catalyze drug uptake. This fact may reflect the accidental usage of a carrier designed to transport one substrate for transport of another due to low degrees of specificity. Only four families of secondary carriers are involved in the export of macromolecules (category VII). One of these, the polysaccharide transporter family, is specific for complex carbohydrates, while twin-arginine-targeting family members are specific for redox proteins.

TABLE 12. Substrate selectivities of primary carriers for inorganic ions

Substrate	Polarity	Energy Source	Family(-ies)
Monovalent cations			
H^+	Out	ATP or P_2	F (V or A)-ATPases; P-ATPases; H^+-PPases
		Electron flow	NDH; QCR; COX; NFO; HHO; FMF-DH
		H^- transfer	PTH
		Light absorption	FAR; PRC
Na^+	Out	ATP	ABC; F (and V)-ATPases; P-ATPases
		Decarboxylation	NaT-DC
		Methyl transfer	NaT-MMM
		Electron flow	NDH; Na-NDH; NFO; FMF-DH
K^+	In	ATP	P-ATPase
Divalent cations			
Ca^{2+}	Out	ATP	P-ATPase
Cd^{2+}	Out	ATP	ABC; P-ATPase
Co^{2+}	In	ATP	ABC
Cu^{2+}	Out	ATP	P-ATPase
Cu^{2+}	In	ATP	P-ATPase
Fe^{2+}	In	ATP	ABC; FeoB?
Fe-chelate	In	ATP	ABC
Mg^{2+}	In	ATP	P-ATPase
Mn^{2+}	In	ATP	ABC
Ni^{2+}	In	ATP	ABC
Zn^{2+}	In	ATP	ABC
Anions			
Antimonite	Out	ATP	ArsAB
Arsenite	Out	ATP	ArsAB
Bicarbonate	In	ATP	ABC
Chloride	In	Light absorption	FAR
Cyanate	In	ATP	ABC
Molybdate	In	ATP	ABC
Nitrate	In	ATP	ABC
Nitrite	In	ATP	ABC
Phosphate	In	ATP	ABC
Sulfate	In	ATP	ABC
Tellurite	Out	ATP	ArsAB
Thiosulfate	In	ATP	ABC

Primary Carriers for Inorganic Ions

Primary carriers may function by either a carrier-type mechanism or a channel-type mechanism, but by definition, the transmembrane transport process is always energized by a primary source of energy (chemical, electrical, or solar energy). These pumps are exceptionally important in biological systems because they are responsible for establishing the ion gradients and membrane potentials upon which secondary carriers are dependent for energization. Primary active transporters are believed to be mechanistically more complex than channels or secondary carriers because their transport activities depend on superimposed catalytic activities that break chemical bonds, pass electrons from a donor molecule to an acceptor, or result in the absorption of light energy. The vast majority of these transport systems function either for the pumping of inorganic ions or for the secretion of macromolecules.

Data regarding the substrate specificities of primary carriers for inorganic ions are summarized in Table 12. Protons and Na^+ ions are each transported by four distinct energy-coupling mechanisms, and two of these mechanisms (ATP hydrolysis and electron flow) are known to be utilized for the transport of both ions. Both ions are transported by primary pumps exclusively in the outward direction. Protons can additionally be extruded by hydride transfer (an unusual type of redox reaction for the energization of a vectorial process) and by light absorption (mediated by bacteriorhodopsin and its homologs in archaea and by photosynthetic reaction centers in bacteria and chloroplasts). Na^+ extrusion can additionally be driven by decarboxylation of a carboxylic acid in bacteria and perhaps in archaea and by methyl transfer in archaea. Light-driven ion transport via bacterio- or halorhodopsin and methyl transfer-driven Na^+ efflux via a methyl coenzyme M-dependent mechanism are so far restricted to the archaeal domain, and each of these processes is restricted to just one small group of archaea. Decarboxylation-driven Na^+ efflux has to date been characterized exclusively in bacteria, but homologs of the decarboxylase subunits, including the Na^+-transporting integral membrane β-subunits of these decarboxylases, are found in the archaeon *Archaeoglobus fulgidus*. The functions of the archaeal subunits have not yet been ascertained. Plants, protozoans, archaea, and bacteria possess proteins that belong to a unique family of vacuolar H^+-transporting pyrophosphatases. In plants, these enzymes pump protons into the vacuolar lumen, thereby generating a transmembrane PMF. It has been suggested that these enzymes may be relics of ancient systems that existed before the advent of ATP (6).

Permease proteins of three families function in ATP or pyrophosphate hydrolysis-driven proton efflux, and six different families probably mediate electron flow-driven proton extrusion. Three families have been shown to mediate ATP hydrolysis-dependent Na^+ pumping, and four may catalyze electron flow-dependent Na^+ expulsion. The Na^+-transporting NADH dehydrogenase family is not homologous or related to the H^+- or Na^+-transporting NADH dehydrogenase family. Recently published evidence has shown that the proteins of the latter family are capable of replacing Na^+ with H^+ (64). The commonly assumed equivalence of H^+ and Na^+ as substrates of primary carriers often, but perhaps not always, applies.

Only one family of primary carriers apparently mediates K^+ active transport, and members of this family, the P-type ATPase family, occur in various structural forms (Table 12). These pumps function by K^+:Na^+ or K^+:H^+ antiport in animals but possibly by K^+ uniport in bacteria. An Na^+ extrusion P-type ATPase is found in *Saccharomyces cerevisiae*. In spite of major differences in substrate recognition and subunit composition for the various P-type ATPases, the mechanisms of transport and energy coupling are likely to be similar. However, since the bacterial K^+-transporting ATPases and the eukaryotic Na^+-K^+ ATPases cluster on completely different segments of the phylogenetic tree (4, 36), significant mechanistic differences can be expected.

Primary pumps that drive divalent cation efflux or uptake always utilize ATP hydrolysis, and either two or three families may be involved (Table 12). Closely related P-type ATPases specific for Cu^{2+} can function with either inwardly or outwardly directed polarity, depending on the system. Bacterial Cd^{2+}-transporting P-type ATPases have been shown to catalyze efflux of several heavy metal ions (Zn^{2+}, Co^{2+}, Ni^{2+}, and Pb^{2+}) as well as Cd^{2+} (7, 49, 80).

Anion transport can be driven by ATP hydrolysis either via ArsAB systems (TC 3.A.4), which catalyze efflux, or via ABC systems (TC 3.A.1), which catalyze uptake. In the case of chloride, halorhodopsin can utilize light absorption to drive Cl^- uptake into the halobacterial cell (92, 144). A single amino acid substitution can convert the outwardly directed proton pump of bacteriorhodopsin into an inwardly directed chloride pump (129, 145). The aspartate-for-threonine substitution at position 85 in bacteriorhodopsin appears to alter both the ion selectivity and the direction of transport. Bacteriorhodopsin and halorhodopsin thus have a common transport mechanism, as expected from their high degree of sequence similarity (52, 67), and a single residue in these proteins strongly influences the ionic specificity.

Table 13 summarizes the varied substrate specificities of ABC permeases. These primary pumps are surprisingly versatile with respect to both the substrate transported and the polarity of pumping. Phylogenetic analyses have revealed that the uptake permeases cluster separately from the efflux permeases (130). ABC transporters can recognize almost any type of substrate that might be of biological interest, regardless of whether it is organic or inorganic, small, intermediate, or large. The architectural basis for this remarkable degree of versatility is likely to prove extremely interesting.

Table 14 provides a detailed summary of the pumping activities of well-characterized primary active transporters and group translocators. Although the variation in substrate specificity is extensive, much of this versatility is due to the activities of ABC-type permeases, as noted above. Excluding this one superfamily and the group translocating PTS-type sugar permeases, almost all primary pumps are specific either for inorganic ions or for macromolecules. Macromolecular pumps will be discussed in the next section.

TABLE 13. Varied specificities of ABC permeases

Compound transported	Polarity
Inorganic cations	In or out
Inorganic anions	In or out
Sugars, polyols, oligosaccharides	In
Organic anions	In or out
Organic cations	In or out
Amino acids and derivatives	In
Amines, polyamines, and opines	In
Peptides (including bacteriocins and pheromones)	In or out
Vitamins	In
Fe^{3+} chelates	In
Fe^{3+} siderophores	In
Drugs; bile salts	In or out
Glutathione and glutathione conjugates	Out
Heme	In or out
Siderophores	Out
Steroids and steroid conjugates	Out
Fatty acids and derivatives	Out
Pigments	Out
Lipooligosaccharides	Out
Polysaccharides and lipopolysaccharides	Out
Teichoic acids	Out
Proteins	Out
Lipids	Out (flipping)

CELLULAR MACROMOLECULAR EXPORT SYSTEMS

Table 15 tabulates the transport systems that catalyze the export of macromolecules. The majority of these systems utilize ATP hydrolysis to drive transport, but several also appear to exhibit a dependency on the PMF. PMF-dependent exporters for complex carbohydrates may include those of the polysaccharide transporter family, while those for proteins include members of the twin-arginine-targeting family. Bacterial holins and certain channel-forming toxins are probably energy-independent protein exporters and importers, respectively. Bacterial MscL channels and mammalian Bcl-2 channels probably also function by energy-independent mechanisms. While three recognized families participate in polysaccharide export, 13 tabulated families participate in protein transport. The mitochondrial and chloroplast envelope protein transport systems can be thought of either as matrix uptake systems or as cytoplasmic export systems. It should be noted that the protein-specific holins and ABC exporters as well as the diphtheria and the botulinum and tetanus toxin importers are relatively simple in structure. ABC export systems may function with trans-envelope protein complexes (157, 158). The more general systems, which transport many proteins, however, consist of large complexes of multiple protein constituents. Only a single type of export system tabulated, the type IV secretory pathway family (TC 3.A.7), mediates export of nucleoprotein complexes. However, another type of system, the bacterial competence-related DNA transformation transporter family (TC 3.A.11), mediates uptake of naked single-stranded DNA in bacteria competent for natural transformation, and a poorly characterized family of systems, the septal DNA translocator family (TC 9.A.16), may function in the transmembrane transport of double-stranded DNA. Two types of active transport systems (ABC and P-type ATPases) are believed to mediate phospholipid flipping from the inner leaflet to the outer leaflet of a biomembrane, although the anion exchanger and RND families of secondary carriers include members that have been reported to do the same (4, 46, 137) (Table 15). These last-mentioned transport systems represent the only examples in which macromolecular export systems are ubiquitous, being found in eukaryotes as well as prokaryotes.

CLASSIFICATION OF TRANSPORTERS OF UNKNOWN MECHANISM

Several families of proteins are known in which one or more members have been shown to function as transporters, but either the mode of transport (channel versus carrier) or the energy source driving solute accumulation or expulsion has not been determined. Consequently, it is not possible to assign the transporter family to a defined category (1–4). Such families fall into TC category 9.A. Additionally, families of proteins in

TABLE 14. Classification of primary carriers according to substrate specificity (excluding macromolecular transporters)

Substrate (polarity)	TC no.	Family	Energy source	Organismal group
I.C.1. Inorganic cations: monovalent				
H^+ (both)	3.A.2.1.1, 1.3, and 2.A.3	F-ATPase	ATP	B, mito, chloro
H^+ (out)	3.A.2.2.1	F (V)-ATPase	ATP	B, E
H^+ (out?)	3.A.2.3.1	F (A)-ATPase	ATP	A
H^+ (out?)	3.A.3.1.2	P-ATPase	ATP	An
H^+ (out)	3.A.3.3.1	P-ATPase	ATP	A, E
H^+ (into vacuoles)	3.A.10.1.1	H^+-PPase	Pyrophosphate	B, Pl
H^+ (out)	3.D.1.1.1–3	NDH	Electron flow	B, mito
H^+ (both)	3.D.2.1–4	PTH	Hydride transfer	B, mito
H^+ (out)	3.D.3.1–5	QCR	Electron flow	B, mito, chloro
H^+ (out)	3.D.4.1–7	COX	Electron flow	B, A, mito
H^+ (in?) or Na^+ (in?)	3.D.6.1.1	NFO	Electron flow	B
H^+ (out)	3.D.7.1.1	HHO	Electron flow	A
H^+ (out)	3.D.8.1.1	FMF-DH	Electron flow	A
H^+ (out)	3.E.1.1.1 and 3	FAR	Light driven	A
H^+ (out)	3.E.2.2.1–2	PRC	Light driven	B, chloro
K^+ (in)	3.A.3.1.1	P-ATPase	ATP	An
K^+ (in)	3.A.3.1.2	P-ATPase	ATP	An
K^+ (in)	3.A.3.3.1	P-ATPase	ATP	Pr, F, Pl
K^+ (in)	3.A.3.7.1	P-ATPase	ATP	B
Na^+ (out)	3.A.1.115.1	ABC	ATP	B
Na^+ (both)	3.A.2.1.2	F-ATPase	ATP	B
Na^+ (out)	3.A.2.2.2	F (V)-ATPase	ATP	B
Na^+ (out)	3.A.3.1.1	P-ATPase	ATP	An
Na^+ (out)	3.A.3.9.1	P-ATPase	ATP	Y
Na^+ (out)	3.B.1.1.1	NaT-DC	Oxaloacetate decarboxylation	B
Na^+ (out)	3.B.1.1.2	NaT-DC	Methylmalonyl-coenzyme A decarboxylation	B
Na^+ (out)	3.B.1.1.3	NaT-DC	Glutaconyl-coenzyme A decarboxylation	B
Na^+ (out)	3.B.1.1.4	NaT-DC	Malonate decarboxylation	B
Na^+ (out)	3.D.1.1.1	NDH	Electron flow	B, mito
Na^+ (out)	3.D.5.1.1	Na-NDH	Electron flow	B
Na^+ (out)	3.D.8.1.1	FMF-DH	Electron flow	A
Na^+ (out)	3.C.1.1.1	NaT-MMM	Methyl transfer	A
I.C.2. Inorganic cations: di- and trivalent				
Ca^{2+} (out)	3.A.3.2.1–4; 1.B.2	P-ATPase	ATP	B, E
Cd^{2+} (out)	3.A.1.207.1	ABC	ATP	Y
Cd^{2+} (out)	3.A.3.6.1 and 2	P-ATPase	ATP	B, F, Pr, Pl
Co^{2+} (in)	3.A.1.18.1	ABC	ATP	B
Co^{2+} (out)	3.A.3.6.2	P-ATPase	ATP	B
Cu^{2+} (in)	3.A.3.5.1	P-ATPase	ATP	B, E
Cu^{2+} (out)	3.A.3.5.2–3	P-ATPase	ATP	B, E
Fe^{3+} (in)	3.A.1.10.1	ABC	ATP	B
Fe^{2+} (in)	3.A.1.15.4 and 6	ABC	ATP	B
Fe^{2+} (in)	3.A.1.20.1	ABC	ATP	B
Fe^{2+} (in)	9.A.8.1.1	FeoB	ATP or PMF(?)	B
Fe chelates	3.A.1.14.1–5	ABC	ATP	B
Metal conjugates (out)	3.A.1.203 and 210	ABC	ATP	Y, An
Mg^{2+} (in)	3.A.3.4.1	P-ATPase	ATP	B
Mn^{2+} (in)	3.A.1.15.1	ABC	ATP	B
Mn^{2+} (out)	3.A.3.2.3	P-ATPase	ATP	Y
Ni^{2+} (in)	3.A.1.5.3	ABC	ATP	B
Ni^{2+} (out)	3.A.3.6.2	P-ATPase	ATP	B
Pb^{2+} (out)	3.A.3.6.2	P-ATPase	ATP	B
Zn^{2+} (in)	3.A.1.15.2, 3, 5, and 6	ABC	ATP	B
Zn^{2+} (out)	3.A.3.6.1 and 2	P-ATPase	ATP	B
I.D. Inorganic anions				
Antimonite (out)	3.A.4.1.1	ArsAB	ATP	B
Arsenite (out)	3.A.4.1.1	ArsAB	ATP	B
Bicarbonate (in)	3.A.1.16.3	ABC	ATP	B
Chloride (in)	3.E.1.2.1	FAR (halorhodopsin)	Light driven	A
Chloride (none)	3.A.1.62.1	ABC (CFTR)	Channel (ATP activated)	An
Cyanate (in)	3.A.1.16.2	ABC	ATP	B
Molybdate (in)	3.A.1.8.1	ABC	ATP	B
Nitrate (in)	3.A.1.16.1	ABC	ATP	B

Continued on following page

TABLE 14—*Continued*

Substrate (polarity)	TC no.	Family	Energy source	Organismal group
Nitrite (in)	3.A.1.16.1	ABC	ATP	B
Phosphate (in)	3.A.1.7.1	ABC	ATP	B
Sulfate (in)	3.A.1.6.1	ABC	ATP	B
Tellurite (out)	3.A.4.1.1	Ars	ATP	B
Thiosulfate (in)	3.A.1.6.1	ABC	ATP	B
II.A. Sugars and polyols				
N-Acetylgalactosamine	4.A.6.1.4	PTS	PEP	B
N-Acetylglucosamine	4.A.1.1.2	PTS	PEP	B
N-Acetylglucosamine	4.A.6.1.1–2	PTS	PEP	B
Allose	3.A.1.2.6	ABC	ATP	B
Arabinose	3.A.1.2.2	ABC	ATP	B
Arabinose	3.A.1.2.5	ABC	ATP	B
Arbutin	4.A.1.2.2–3	PTS	PEP	B
Cellobiose	4.A.1.2.2–3	PTS	PEP	B
Cellobiose	4.A.3.1.2	PTS	PEP	B
Cyclodextrins	3.A.1.1.6	ABC	ATP	B
Diacetylchitobiose	4.A.3.2.1	PTS	PEP	B
Fructose	4.A.2.1.1	PTS	PEP	B
Fructose	4.A.6.1.1–2	PTS	PEP	B
Fucose	3.A.1.2.5	ABC	ATP	B
Galactitol	4.A.5.1.1	PTS	PEP	B
Galactose	3.A.1.2.3	ABC	ATP	B
Galactose	3.A.1.2.5	ABC	ATP	B
Glucitol	3.A.1.1.5	ABC	ATP	B
Glucitol	4.A.4.1.1	PTS	PEP	B
Glucosamine	4.A.6.1.1–2	PTS	PEP	B
Glucose	3.A.1.2.3	ABC	ATP	B
Glucose	3.A.1.2.5	ABC	ATP	B
Glucose	4.A.1.1.1	PTS	PEP	B
Glucose	4.A.6.1.1–2	PTS	PEP	B
β-Glucosides	4.A.1.2.2–3	PTS	PEP	B
Lactose	3.A.1.1.4	ABC	PEP	B
Lactose	4.A.3.1.1	PTS	PEP	B
Lichenan oligosaccharides	4.A.3.2.2	PTS	PEP	B
Maltooligosaccharides	3.A.1.1.1	ABC	ATP	B
Maltose	3.A.1.1.1, -7, -8	ABC	ATP	B
Maltose	4.A.1.1.3	PTS	PEP	B
Mannitol	3.A.1.1.5	ABC	ATP	B
Mannitol	4.A.2.1.2	PTS	PEP	B
Mannose	4.A.6.1.1–2	PTS	PEP	B
Melibiose	3.A.1.1.2	ABC	ATP	B
Multiple sugars	3.A.1.1.2, -2.5, -3.1	ABC	ATP	B
Raffinose	3.A.1.1.2	ABC	ATP	B
Ribose	3.A.1.2.1	ABC	PEP	B
Salicin	4.A.1.2.2–3	PTS	PEP	B
Sorbose	4.A.6.1.3	PTS	PEP	B
Sucrose	3.A.1.1.8	ABC	ATP	B
Sucrose	4.A.1.2.1	PTS	PEP	B
Trehalose	3.A.1.1.7, -8	ABC	ATP	B
Trehalose	4.A.1.2.4	PTS	PEP	B
Xylose	3.A.1.2.4	ABC	ATP	B
Xylose	3.A.1.2.5	ABC	ATP	B
II.B. Fatty acids				
Fatty acids (long chain)	3.A.1.203.1–2	ABC	ATP	An, Y
II.D. Organoanions (phosphates, phosphonates, sulfonates) (uptake)				
2-Aminoethyl phosphonate	3.A.1.11.5	ABC	ATP	B
2-Aminoethyl sulfonate (taurine)	3.A.1.17.1	ABC	ATP	B
Glucuronate	3.A.1.1.9	ABC	ATP	B
Glycerol phosphate	3.A.1.1.3	ABC	ATP	B
Phosphates (organic)	3.A.1.9.1	ABC	ATP	B
Phosphonates (organic)	3.A.1.9.1	ABC	ATP	B
Taurine	3.A.1.17.1	ABC	ATP	B

Continued on following page

TABLE 14—*Continued*

Substrate (polarity)	TC no.	Family	Energy source	Organismal group
III.A. Amino acids and derivatives (uptake)				
Agrocinopine	3.A.1.5.4	ABC	ATP	B
Arginine	3.A.1.3.1, -3, -11	ABC	ATP	B
Asparagine	3.A.1.3.7	ABC	ATP	B
Aspartate	3.A.1.3.4	ABC	ATP	B
Aspartate	3.A.1.3.7	ABC	ATP	B
Chrysopine	3.A.1.11.4	ABC	ATP	B
Cystine	3.A.1.3.10	ABC	ATP	B
Diaminopimelate	3.A.1.3.10	ABC	ATP	B
General L-amino acids	3.A.1.3.8	ABC	ATP	B
Glutamate	3.A.1.3.4	ABC	ATP	B
Glutamate	3.A.1.3.7	ABC	ATP	B
Glutamate	3.A.1.3.9	ABC	ATP	B
Glutamine	3.A.1.3.2	ABC	ATP	B
Glutamine	3.A.1.3.7	ABC	ATP	B
Glycine betaine	3.A.1.12.1–2	ABC	ATP	B
Glycine betaine	3.A.1.12.4	ABC	ATP	B
Histidine	3.A.1.3.1	ABC	ATP	B
Isoleucine	3.A.1.4.1	ABC	ATP	B
Leucine	3.A.1.4.1	ABC	ATP	B
Lysine	3.A.1.3.1	ABC	ATP	B
Mannopine	3.A.1.11.3	ABC	ATP	B
Nopaline	3.A.1.3.6	ABC	ATP	B
Octopine	3.A.1.3.5	ABC	ATP	B
Ornithine	3.A.1.3.1, -11	ABC	ATP	B
Proline	3.A.1.12.1	ABC	ATP	B
Valine	3.A.1.4.1	ABC	ATP	B
III.B. Amines, amides, and polyamines (uptake)				
Amides, short chain	?	AmiS (B)	ATP?	B
γ-Butyrobetaine	3.A.1.12.1 and 4	ABC	ATP	B
D- and L-carnitine	3.A.1.12.1 and 4	ABC	ATP	B
Choline	3.A.1.12.1 and 3–4	ABC	ATP	B
Choline-*O*-sulfate	3.A.1.12.4	ABC	ATP	B
Crotonobetaine	3.A.1.12.4	ABC	ATP	B
Dimethylproline	3.A.1.12.1	ABC	ATP	B
Ectoine	3.A.1.12.4	ABC	ATP	B
Glycine betaine	3.A.1.12.1, 2 and 4	ABC	ATP	B
Homobetaine	3.A.1.12.1	ABC	ATP	B
Polyamines	3.A.1.11.1	ABC	ATP	B
Proline betaine	3.A.1.12.1	ABC	ATP	B
Putrescine	3.A.1.11.1, -2	ABC	ATP	B
Quaternary amines	3.A.1.12.1–5	ABC	ATP	B
Spermidine	3.A.1.11.1	ABC	ATP	B
III.C. Peptides				
a-Factor (sex pheromone) (out)	3.A.1.206.1	ABC	ATP	Y
Cationic peptides (in)	3.A.1.5.5	ABC	ATP	B
Cyclic peptides (out)	3.A.1.113.1	ABC	ATP	B
Dipeptides (in)	3.A.1.5.2	ABC	ATP	B
Glutathione (oxidized) (out)	3.A.1.207.1 and 208.2	ABC	ATP	An
Glutathione conjugates (out)	3.A.1.207.1	ABC	ATP	An
MHC peptides (out)	3.A.1.209.1	ABC	ATP	An, Y
Oligopeptides (in)	3.A.1.5.1	ABC	ATP	B
Peptide antibiotics (in)	3.A.1.5.5	ABC	ATP	B
Peptides (out)	3.A.1.111–113, 116, 118	ABC	ATP	B
Peptides (out)	3.A.1.201.1	ABC	ATP	An
Protamines	3.A.1.5.5	ABC	ATP	B
V.A. Vitamins				
Thiamine	3.A.1.19.1	ABC	ATP	B
Vitamin B_{12} (uptake)	3.A.1.13.1	ABC	ATP	B
V.B. Enzyme and redox cofactors				
Eye pigment (export)	3.A.1.204.1	ABC	ATP	An
Fatty acyl coenzyme A	3.A.1.203.1–2	ABC	ATP	Y, An (peroxysomes)
Heme (hemin) (uptake)	3.A.1.14.5	ABC	ATP	B
Heme (hemin) (export)	3.A.1.107.1	ABC	ATP	B
Thiamine pyrophosphate	3.A.1.19.1	ABC	ATP	B

Continued on following page

TABLE 14—*Continued*

Substrate (polarity)	TC no.	Family	Energy source	Organismal group
V.C. Siderophores and Fe-siderophore complexes				
Fe-aerobactin	3.A.1.14.3	ABC	ATP	B
Fe chelates (many)	3.A.1.15.4	ABC	ATP	B
Fe-chrysobactine	3.A.1.14.4	ABC	ATP	B
Fe-coprogen	3.A.1.14.3	ABC	ATP	B
Fe-dicitrate	3.A.1.14.1	ABC	ATP	B
Fe-enterobactin	3.A.1.14.2	ABC	ATP	B
Fe-ferrichrome	3.A.1.14.3	ABC	ATP	B
Fe-ferrioxamine	3.A.1.14.3	ABC	ATP	B
Fe-hydroxamate	3.A.1.14.3	ABC	ATP	B
Siderophores (export)	3.A.1.113, 119	ABC	ATP	B
VI.A. Multiple drugs (mostly efflux)				
Agrocin 84 (uptake)	3.A.1.5.4	ABC	ATP	B
Aminoglycoside antibiotics (uptake)	3.A.1.5.1	ABC	ATP	B
Multiple drugs	3.A.1.108, 117, 119	ABC	ATP	G+ B
Multiple drugs	3.A.1.201, 205, 208, 210	ABC	ATP	B, F, Y, An, B
VI.B. Specific drugs (mostly efflux)				
Agrocin 84 (uptake)	3.A.1.5.4	ABC	ATP	G− B
Albomycin (uptake)	3.A.1.14.3	ABC	ATP	G− B
Daunorubicin	3.A.1.105.1	ABC	ATP	G+ B
Doxorubicin	3.A.1.105.1	ABC	ATP	G+ B
Erythromycin	3.A.1.105.4	ABC	ATP	G+ B
Lantibiotics	3.A.1.114.1	ABC	ATP	G− B
Macrolides	3.A.1.105.3	ABC	ATP	G+ B
Microcin B17	3.A.1.116.1	ABC	ATP	G− B
Oleandomycin	3.A.1.105.2	ABC	ATP	G+ B
Oligomycin	3.A.1.208.3	ABC	ATP	Y
Syringomycin	3.A.1.113.1	ABC	ATP	G− B
Tylosin	3.A.1.105.5	ABC	ATP	G+ B
VI.C. Bile salts and conjugates				
Bile salts	3.A.1.207.2 and 208.2	ABC	ATP	An
Bilirubin	3.A.1.208.2	ABC	ATP	An
Bilirubin glucuronides	3.A.1.208.2	ABC	ATP	An
Glucuronides	3.A.1.208.2	ABC	ATP	An
Glutathione conjugates	3.A.1.207.1	ABC	ATP	Y
Glutathione conjugates	3.A.1.208.1–2	ABC	ATP	An
Leukotrienes	3.A.1.208.1–2	ABC	ATP	An
VI.D. Sterol and conjugates				
Steroids (export)	3.A.1.205.1	ABC	ATP	Y

which no member of the family has been shown to be a transporter are known, although some indirect experimental evidence, or inferences based on topological analyses and/or operon gene product analyses, supports such a possibility. Such families fall into TC category 9.B. Finally, functionally characterized transporters lacking an identified sequence fall into TC category 9.C. The families listed in these categories will either be transferred to one of the established categories when their transport mechanism becomes defined or be eliminated from the TC system if it is shown that these proteins are not actual transporters. In this section, the families that constitute TC class 9.A will be discussed. Those of classes 9.B and 9.C will not be considered further here.

Table 16 tabulates families of known transporters for which no member has yet been clearly defined in terms of either its mode of transport (channel or carrier) or its energy-coupling mechanism. Many of these permeases belong to families that include members which are specific for inorganic ions. Eleven families are inorganic ion specific, and 10 of these are cation specific. Most of these families include members that are specific for a single ion or a few closely related ions. However, one family (low-affinity cation transporter, TC 9.A.20) transports a variety of cations, exhibiting unexpectedly broad specificity.

Some of the category 9.A permeases (belonging to six distinct families) exhibit specificity for small organic compounds. These compounds vary from amides and amines, including urea and uric acid, to peptides and vitamin precursors. Thus, a variety of organocations, organoanions, and neutral molecules are transported. One family (polysaccharide transporter) transports complex polysaccharides, probably by a PMF-dependent mechanism, but the energy-coupling mechanism is still poorly defined. Considerations to be discussed in the next section allow prediction of the modes of action of several of these systems. Putative transporters (category 9.B) are not discussed here but can be evaluated by consideration of the information provided in our web site.

PREDICTIONS OF TRANSPORT MODE BASED ON PROTEIN TOPOLOGY

Examination of the topologies of families of recognized α-type channels (TC 1.A) and secondary carriers (TC 2.A)

TABLE 15. Classification of cellular macromolecular export systems (excluding porins)

Substrate	TC no.	Family(ies)	Energy source	Organismal group
VII.A. Carbohydrates				
Capsular polysaccharide	3.A.1.101.1	ABC	ATP	B
Exopolysaccharide	9.A.1.2.1–2	PST	ATP/PMF?	B
β-Glucan	3.A.1.108.1	ABC	ATP	G− B
Lipooligosaccharide	2.A.6.3.1	RND	PMF	G− B
Lipooligosaccharide	3.A.1.102.1	ABC	ATP	G− B
Lipopolysaccharide	3.A.1.103.1	ABC	ATP	G− B
Lipopolysaccharide	9.A.1.1.1	PST	PMF?	G− B
Teichoic acid	3.A.1.104.1	ABC	ATP	G+ B
VII.B. Proteins				
Autolysins, nucleases	1.A.28–1.A.43	Holins	None	B, phage
Proteins (bacterial)	1.A.22.1.1	MscL	None	B
Proteins (mitochondrial)	9.A.5.1.1	Oxa1	?	Y (mito)
Proteins (mitochondrial)	1.A.21.1.1	Bcl-2	None	An (mito)
Protein toxins	1.C.7; 1.C.8	DT; BTT	None	B
Proteins (redox)	2.A.64	Tat	PMF	B
Proteins and peptides (bacteriocidin, bacteriocins, colicin V, competence factors, cyclolysin, α-hemolysin, nisin, pediosin PA-1, proteases, S-layer proteins, subtilin, etc.)	3.A.1.109–112	ABC	ATP	B
Proteins	3.A.5.1–8	IISP	ATP + PMF	B, A, Y, An, chloroplasts
Proteins, flagellar constituents	3.A.6.1.1–2	IIISP	ATP	B
Proteins	3.A.7.1.1	IVSP	ATP	B
Proteins	3.A.8	MPT	ATP/PMF	Mitochondria
Proteins	3.A.9	CEPT	ATP/PMF	Chloroplast envelope
VII.C. Nucleic acids				
DNA (single stranded)	3.A.11	DNA-T	ATP	B
DNA-protein complexes	3.A.7.1.1	IVSP	ATP	B
VII.D. Lipids				
Aminophospholipids	3.A.3.8.1–2	P-ATPase	ATP	An, Y, F
Glycolipids	3.A.1.114.1	ABC	ATP	B
Lipids (general)	3.A.1.201.1	ABC	ATP	An, Y, F, B
Lipids	3.A.6.4.2	RND	PMF	B
Lipid A	3.A.1.106.1	ABC	ATP	B
Phospholipids	2.A.31.1.1	AE	None	An
Phospholipids	3.A.1.201.1	ABC	ATP	An, Y, F, B

reveals that these two functional types of transporters differ fundamentally both in polypeptide structure and in oligomeric composition. This fact suggests that there are fundamental differences between these two functional types of transporters and that channels and carriers truly represent distinct types of proteins. This structural distinction between the two principal functional types of transporters is evaluated in this section.

As illustrated in Fig. 2, most families of cellular integral membrane α-type channel proteins include members that possess three or fewer TMSs per polypeptide chain (Fig. 2A), while almost all families of secondary carriers include members that possess eight or more TMSs (Fig. 2B). When permease families of unknown transport mode are examined (Fig. 2C), some are found to fall into the 1 to 3 TMS range observed for most channel families, while others fall into the 8 to 14 TMS range observed for most carrier families. It can be anticipated that most of the former proteins will prove to be channels, while the latter will mostly prove to be carriers. The disproportionate number of families of unknown mechanism of action with about 6 TMSs leads to the possibility that new types of transporters, not yet characterized, may be found among these families.

Interestingly, very few carriers have been shown to be capable of functioning as channels under any experimental set of conditions. Two of those that do exhibit this unusual property prove to consist of polypeptide chains that have 6 TMSs. Families of such transporters include the mitochondrial carrier family (TC 2.A.29) and the TP-NST family (TC 2.A.50) (17, 28, 29, 132). The *E. coli* KefB and KefC proteins of the CPA2 family (TC 2.A.37) also seem to have the capacity to function either by a carrier-type K^+:H^+ antiport mechanism or by a K^+-specific channel-type mechanism (38, 39). Proteins of this last-mentioned family exhibit 10 to 14 putative TMSs and therefore have the topology of a typical carrier. They exhibit channel-type activities following treatment with certain chemicals. Ambivalent modes of transport for members of a few other secondary carrier families have also been noted (see reference 121 for further consideration of this point).

On the basis of all of the observations summarized in this section, we propose that, with only a few exceptions, channel proteins are structurally and functionally different from carriers. Channels are proposed to generally consist of oligomeric structures in which the monomeric protein subunits exhibit ≤3 TMSs. Some exceptions to this rule have resulted from the fusion of non-transport-regulatory domains to the channel-forming constituents of transporters (88). Regardless of topology, however, the channel generally results from the proper association of multiple channel-forming subunits or domains. Carriers, on the other hand, are proposed to generally consist of functional monomers that exhibit 8 to 14 TMSs or, less frequently, of functional dimers that have 4 to 7 TMSs. In these situations, the transport pathway requires the participation of just one or, at most, two polypeptide chains. The numbers of known exceptions to this topological rule are small (Fig. 2).

TABLE 16. Classification of transport systems functioning by an unknown mechanism (class 9.A) according to substrate specificity

Substrate	TC no.	Family	Organismal group
I.A. Nonselective			
Nonselective	9.A.14	NPC	E
I.C. Inorganic cations			
Cation (nonselective)	9.A.17	MIT	B, A, E
Cation (nonselective)	9.A.20	LCT	Pl
Cu^{2+} (in)	9.A.11	Ctr1	Y
Cu^{2+} (in)	9.A.12	Ctr2	An, Pl, Pr, Y
Fe^{2+} (in)	9.A.8	FeoB	B, A
Fe^{2+} (in)	9.A.9	FeT	Y
Fe^{2+} (in)	9.A.10	OFeT	Y
Hg^{2+} (out)	9.A.2	MerTP	B
Hg^{2+} (out)	9.A.3	MerC	B
Mg^{2+} (in)	9.A.19	MgtE	B, A
II.B. Monocarboxylates			
Fatty acids (short chain)	9.A.13	scFAT	B
III.B. Amides and amines			
Amides (in)	9.A.15.1.1–2	Ami	B
Urea (in)	9.A.15.1.3	Ami	B
III.C. Peptides			
Peptides (in)	9.A.18	PUP	B
IV.A. Bases			
Uric acid (out)	9.A.17	UAT	An
IV.B. Nucleoside			
Nucleosides (general)	9.A.6	INT	An
IV.C. Nucleotides			
Nicotinamide mononucleotide (in)	9.A.4	PnuC	G− B
V. Vitamins			
Nicotinamide mononucleotide (in)	9.A.4	PnuC	G− B
VII.A. Complex carbohydrates			
Capsular polysaccharides (out)	9.A.1.2.2	PST	G+ B
Exopolysaccharides (out)	9.A.1.2.1	PST	G− B
Lipopolysaccharides (out)	9.A.1.1.1	PST	G− B
VII.B. Proteins			
Proteins	9.A.5	Oxa1	E (mito), B, A
Proteins	9.A.14	NPC	E
Microcins (uptake)	9.A.18.1.2	PUP	G− B
VII.C. Nucleic acids			
DNA	9.A.16.1.1–2	SDT	G+ B
RNA	9.A.14	NPC	E

RECOGNIZED DISTRIBUTION OF TRANSPORTER FAMILIES IN THE THREE DOMAINS OF LIFE

Our studies of the distribution of proteins within the various families of transporters have revealed that most families are restricted to just one of the major domains of life, bacteria, archaea, or eukarya. Other families are ubiquitous, being found in all three domains. If lateral transfer and fixation of genetic material occurred appreciably between these three domains during the past two billion years, one would expect many families to be ubiquitous. Our observations have therefore led to the suggestion that the ubiquitous families are among the oldest families and that they existed before divergence of the three major domains of organisms, some three billion years ago. The domain-specific families are therefore those that arose late, after the "great split." Alternatively, some of these families may have diverged in sequence from their ancestral system at rates that exceed those observed for the recognized ubiquitous families. Even if this occurred, however, the lack of recognizable homologs in the other domains suggests the absence of appreciable lateral transfer.

Table 17 summarizes the distribution of the identified families of various channel types, secondary carriers, primary carriers, group translocators, and transporters of unknown mechanism in the three domains of life: bacteria, archaea, and eukaryotes. Regardless of transporter category, the distribution is simple. Thus, many families of transport systems are found exclusively in either bacteria or eukaryotes, and four have been identified only in archaea. Many of these families may prove to exist in only one of the three major domains of life, and most such families probably arose within that kingdom after the three domains of living organisms separated from each other. It is also possible that some ancient families that existed prior to the divergence of archaea and eukaryotes from bacteria will prove to be restricted to just one or two domains because a particular transport mode or energy-coupling mechanism is incompatible with (or disadvantageous to) the organisms within a particular domain. It is particularly noteworthy in this regard that although hundreds of genes of the PTS (TC 4.A) have been sequenced from bacteria and many of the genes encoding the cytoplasmic constituents of the PTS function in regulation rather than in transport, not a single such gene has yet been found within an archaeal or eukaryotic genome (J. Reizer and M. H. Saier, Jr., unpublished results). Similarly, although ABC-type efflux pumps are universal, extracytoplasmic receptor-dependent ABC-type uptake permeases (TC 3.A.1) as well as receptor-dependent tripartite ATP-independent periplasmic transporter-type uptake permeases (TC 2.A.56) are found only in prokaryotes (107). These

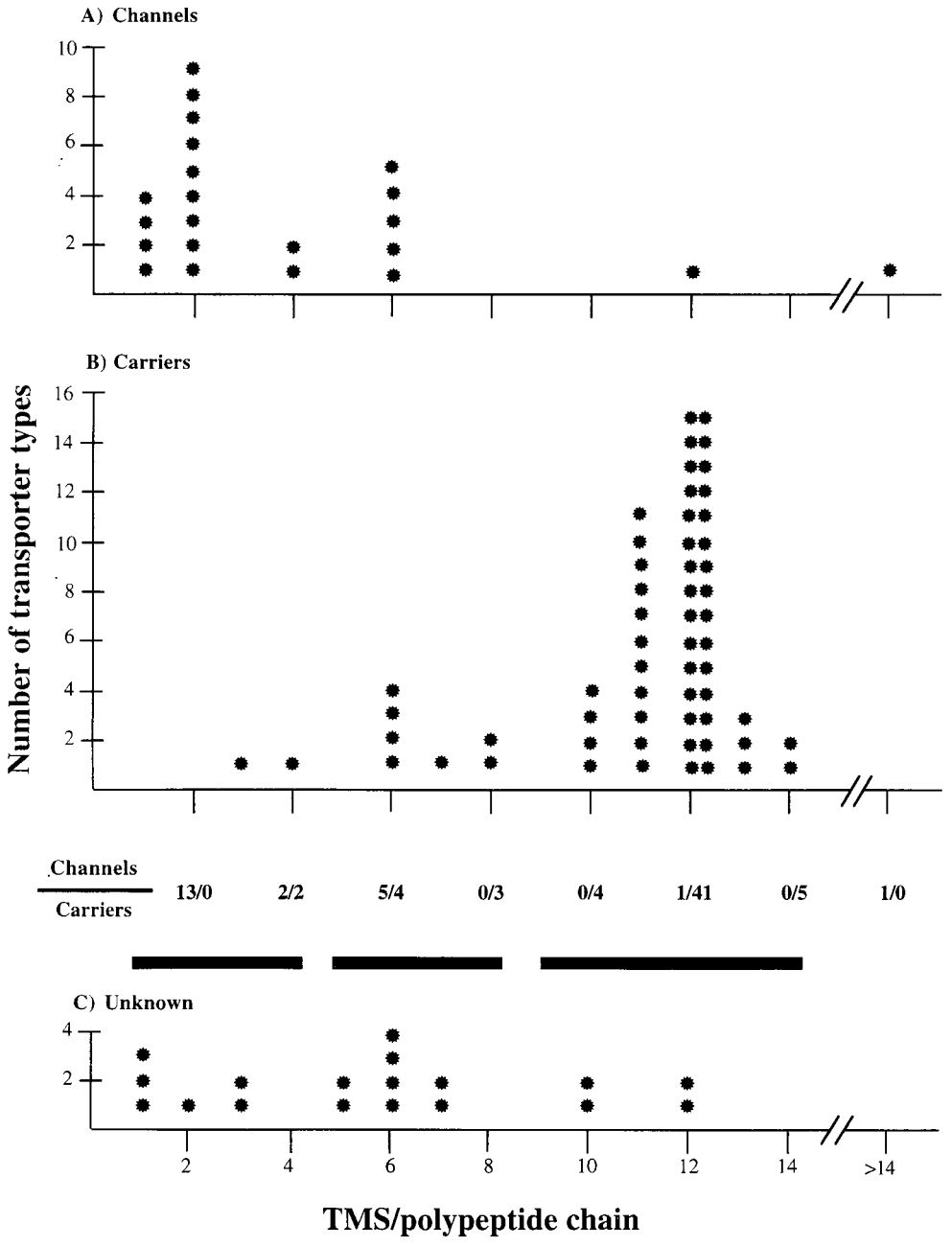

FIG. 2. Established or predicted topologies for channel proteins (A), carrier proteins (B), and proteins of unknown transport mode (C). The proteins included in A are the channel proteins of TC category 1.A, while the carriers represented in B are the families of TC category 2.1A. Because most primary carriers of categories 3 consist of heterooligomers, many of very complex structure, these were not included in the analyses depicted.

observations have led us to conclude that horizontal transmission and fixation of genetic material across the three domains of life has occurred rarely, at least in the case of genes encoding many types of transporters, during the past two billion years.

Several families are found ubiquitously in all three domains of living organisms or are found in at least two of these domains (Table 17). We predict that many (but not necessarily all) of the latter families will prove to be represented in all three domains. The lower representation of transporter types in the archaeal domain presumably reflects, at least in part, the paucity of both sequence data and functional analyses reported for this domain. It should be noted that the vast majority of ubiquitous families (about two-thirds) are families of secondary carriers. The distribution of transporter types and the identification of the relevant families are presented in Table 18 for the various channel types, in Table 19 for the various carrier families, and in Table 20 for the various types of primarily active transporters.

Several interesting conclusions derived from the data in Tables 17 to 20 can be tentatively made. First, of the families of channels, only three families (MIP, VIC, and ClC) are ubiquitous. A fourth such family may prove to be the metal ion transporter family (TC 9.A.17). Second, except for these fam-

TABLE 17. Distribution of transporter families in the three domains of life[a]

Transporter type	No. of families per group						
	B	A	E	BA	BE	AE	BAE
Channels							
Protein channels	2	0	**17**	0	0	0	**3**
Protein toxins	**8**	0	1	0	1	0	0
Peptide toxins	**11**	0	4	0	0	0	0
Holins	**16**	0	0	0	0	0	0
Porins	**22**	0	1	0	0	0	0
Total	**59**	0	**23**	0	1	0	**3**
Secondary carriers	**23**	0	**12**	3	5	0	**27**
Primary carriers	(**6**–10)	4	2	1	(1–**5**)	1	**6**
Group translocators	**6**	0	0	0	0	0	0
Unknown	**8**	0	**6**	3	1	0	**2**
Total	**102**	4	**43**	7	12	1	**38**

[a] Numbers that are boldface and underlined represent the major organismal grouping(s) (B, bacteria; A, archaea; E, eukaryotes; BE, bacteria and eukaryotes; BAE, bacteria, archaea, and eukaryotes) in which each transporter type is found. For the primary carriers, the bold number in parentheses refers to the organismal distribution; the nonbold numbers are the values that would result if mitochondria and chloroplasts are considered to be of bacterial origin. Data are as of 1 January 1999.

TABLE 19. Kingdom distribution of secondary carrier families (TC 2.A)

Kingdom	No. of families	TC nos.[a]
B	25	8–11, 13, 15, 24–27, 33–35, 42, 46, 52, 61, 63, 68, 70, 73, 75–78
E	14	5, 18, 29, 31, 32, 43, 45, 48, 50, 54, 57, 60, 65, 74
A	0	
BAE	31	1–4, 6, 7, 16, 17, 19–23, 28, 30, 36–41, 44, 47, 49, 53, 55, 59, 64, 66, 67, 69
BA	4	14, 51, 56, 62
BE	4	12, 58, 71, 72
AE	0	
Total	78	

[a] Only the family number is provided, as all families are within TC category 2.A.

ilies and two families (MscL and MscS) specific to bacteria, all families of α-type protein channels are found exclusively in eukaryotes. Third, the vast majority of these eukaryotic families are restricted to animals. Finally, the vast majority of protein and peptide toxin families, holin families, and β-strand-type porin families are restricted to bacteria. In the case of the toxin families, this unequal distribution may reflect, at least to some extent, my greater focus (and that of research scientists in general) on bacterial toxins rather than those of eukaryotes.

The distribution of secondary carriers is not so polarized. Thus, 31 carrier families (40%) are ubiquitous, compared to 25 (32%) and 14 (19%) that are specific to bacteria and eukaryotes, respectively. Only eight (10%) are found in two of the three kingdoms. If lateral transfer of genetic material coding for transporters has been minimal, as we have proposed (119, 120), then it would appear that a large proportion of the secondary carrier families came into existence early, before the split between the three domains, compared to channel or primary carrier families.

Primary carrier families are found solely in bacteria, ubiquitously, and in bacteria plus eukaryotes, in decreasing numbers in that order. The relatively large percentage of systems in the last category is due to the presence of three families of H^+-pumping electron or hydride-transferring carriers that are found only in mitochondria and/or chloroplasts of eukaryotes

TABLE 18. Kingdom distribution of channel families: α-type (1.A), porins (1.B), and toxins (1.C)

Channel type	Distribution	No. of families	TC nos.
α-Type channels	Ubiquitous	3 (or 4)	1.A.1; 1.A.8; 1.A.11; (9.A.17)
	B	3	1.A.22; 1.A.23; 1.A.45
	B, A	1	1.A.23
	An	16	1.A.2–1.A.7; 1.A.9; 1.A.10; 1.A.12; 1.A.14; 1.A.21; 1.A.24; 1.A.25; 1.A.27; 1.A.44
	Animal viruses	1	1.A.19
	An, Y	1	1.A.15
	An, Pl	1	1.A.20
	Pl	3	1.A.17; 1.A.18; 1.A.26
	Y	1	1.A.16
α-Autolysins	B	6	1.A.28; 1.A.29; 1.A.31; 1.A.34; 1.A.41; 1.A.42
	Bacteriophages	13	1.A.28–1.A.30; 1.A.32–1.A.40; 1.A.43
β-Porins	B	25	1.B.1–7; 1.B.9–26
	E (organelles)	1	1.B.8
Protein toxins	B	12	1.C.1–3; 1.C.5; 1.C.7–1.C.14; 1.C.36
	B, Pl	1	1.C.4
	Y	1	1.C.6
	An	1	1.C.15
Peptide toxins	B	12	1.C.20–1.C.31
	An	8	1.C.16–1.C.19; 1.C.32–1.C.35
Total		86	

TABLE 20. Kingdom distribution of primary active transporter families (including group translocators)

Kingdom	No. of families	TC nos.					
		ATP hydrolysis	Decarboxylation	Methyl transfer	Redox	Light	PTS
B	11	3.A.6; 3.A.7; 3.A.11			3.D.5; 3.D.6		4.A.1–4.A.6
A	3			3.C.1	3.D.7; 3.D.8		
E	2	3.A.8; 3.A.9					
BAE	7	3.A.1–3.A.5; 3.A.10			3.D.4		
BA	1		3.B.1				
BE	4				3.D.1–3.D.3	3.E.2	
AE	1					3.E.1	
Total	29	11	1	1	8	2	6

in addition to bacteria. Since both of these eukaryotic organelles are believed to have arisen from bacteria long after the split between bacteria and eukaryotes (93), the actual proportion of primary active transporter families specific to bacteria may be considered substantially greater, while that of carriers shared by bacteria and eukaryotes may be smaller (see lightface values in parentheses in Table 17). Finally, one family, the fungal-archaeal rhodopsin family (TC 3.E.1), is unusual in that although these light-driven ion transporters are restricted to one small group of archaea, homologs that may not function in transport are found in yeasts and other fungi. This may represent one of those rare examples where distant homologs of a transporter family have evolved to serve very different functions (see below). The results summarized in Tables 18 to 20 provide a detailed breakdown of channels, secondary carriers, and primary carriers, respectively, and the TC numbers of the families in each category are provided so that the reader can easily identify the relevant families.

It has been noted that archaeal metabolic enzymes and transporters frequently resemble the homologous bacterial sequences more than those of the corresponding eukaryotic proteins, although archaeal proteins of DNA replication, transcription, and translation are more similar to those of eukaryotes (20, 34). This observation has been interpreted to suggest that archaea are mosaic organisms, with nucleic acid and protein-biosynthetic enzymes derived primarily from an early eukaryotic precursor cell, while transport and metabolic functions are derived primarily from a primordial bacterium. If such a "fusion" event was responsible for the generation of the archaeal lineage, a significant number of transporter families should prove to be restricted to bacteria and archaea but lacking in eukaryotes. The availability of four complete archaeal genome sequences has allowed resolution of this question. Of the 200 families represented in Table 17, only 7 (3.5%) are shared by bacteria and archaea but not by eukaryotes. Similarly, very few families are represented in bacteria and eukaryotes but not archaea. Moreover, some of these last-mentioned families are represented only in eukaryotic organelles, suggesting a more recent bacterial origin. Thus, very few families may prove to be restricted to just two of the three domains of living organisms. An alternative view concerning the origin of archaea, such as that proposed recently by Poole et al. (103), may be worth considering.

TRANSPORT PROTEINS FOR WHICH THREE-DIMENSIONAL STRUCTURAL DATA ARE AVAILABLE

An ultimate understanding of transport will depend upon detailed structural data for each of the major classes of transport systems. Until recently, few or no such data were available. The approach of X-ray crystallography has yielded very significant advances in understanding the three-dimensional structures of certain classes of integral membrane proteins. Most of these proteins are of prokaryotic origin, and they do not yet include the major classes represented by secondary carriers, group translocators, and ATP-driven primary pumps. However, channel-type proteins and both light- and electron flow-driven proton pumps are now structurally understood at high resolution.

Table 21 lists the transport proteins for which high-resolution three-dimensional structural data are available. Four types of channel proteins (α-helix-forming channels, β-barrel porins, peptide channels, and protein toxin channels) are represented, as are electron flow-driven and light absorption-driven proton pumps. The structures of these and other membrane proteins have been discussed by Sakai and Tsukihara (128). The fact that no chemically driven primary carriers, no facilitators or secondary carriers, and no group translocators are represented means that we are currently far from a structural understanding of transport. Although the structures of several water-soluble domains (receptors or energy-coupling proteins) of some of these systems have been determined (i.e., ABC-type receptors, the transhydrogenase hydride transfer domains and pumps, and the energy-coupling proteins of the PTS) (105, 106, 126), the structures of the integral membrane constituents of these systems are still unsolved. In fact, high-resolution structures are not available for a single transport system within one of these categories, even though these types of transporters represent the major types found in nature. Much work will be required before molecular transport can be put on a firm structural basis.

TRANSPORTER FAMILIES INCLUDING NONTRANSPORTING HOMOLOGS

Of the currently recognized 250-plus families of established transporters, we have noted that only 7 include transmembrane proteins that have been shown to function in a capacity other than transport. Of these seven families, four include homologs that are believed to serve as receptors (Table 22). In the case of the ammonium transporter family of NH_3 (or NH_4^+) transporters, a yeast homolog, Mep2p, acts as both a sensor and a transporter. In the MFS and amino acid-polyamine-organocation superfamilies, the putative transcriptional regulatory sensors have not been shown to be incapable of transporting their ligands, although the available evidence is against it (57, 65). In the case of the MFS receptors, protein domains that interfere with transport function may be required to convert a transporter into a signaling receptor (57, 65). This scenario is reminiscent of the sensory rhodopsins, for which interaction with transducer proteins blocks proton transport (159). In the case of the RND superfamily, a homologous integral membrane

TABLE 21. Transporters for which three-dimensional structural data have been reported[a]

Transporter type and TC no.	Protein	Family	Source	PDB code[b]
1.A. α-Type channels				
1.A.1.1.1	K^+ channel, KcsA	VIC	*Streptomyces lividans*	1BL8
1.A.9.1.1	Acetylcholine receptor	LIC	*Torpedo* electric organ	3MRA
1.A.21.1.1	Apoptosis regulator, Bcl-X(L)	Bcl-2	*Homo sapiens*	1MAZ
1.A.22.1.2	Mechanosensitive channel	MscL	*Mycobacterium tuberculosis*	1MSL
1.B. β-Barrel porins				
1.B.1.1.1	Porin (OmpC)	GBP	*E. coli*	1IIV (Theo.)
1.B.1.1.2	Porin (PhoE)	GBP	*E. coli*	1PHO
1.B.1.1.3	Porin (OmpF)	GBP	*E. coli*	1OPF
1.B.3.1.1	Maltoporin (LamB)	SP	*Salmonella typhimurium*	1MAL
1.B.6.1.1	Porin (OmpA)	OOP	*E. coli*	1BXW
1.B.7.1.1	Porin (PorCa)	RPP	*Rhodobacter capsulatus*	2POR
1.B.14.1.4	FhuA ferrichrome receptor	OMR	*E. coli*	1BY5
1.C. Pore-forming protein and peptide toxins				
1.C.1.1.1	Colicin Ia	Colicin	*E. coli*	1CII
1.C.1.2.2	Colicin E1	Colicin	*E. coli*	1COL
1.C.2.1.1	Cry 1Aa	ICP	*Bacillus thuringiensis*	1CIY
1.C.2.2.1	Cry 3Aa	ICP	*Bacillus thuringiensis*	1DLC
1.C.3.1.1	α-Hemolysin	αHL	*Staphylococcus aureus*	7AHL
1.C.4.1.1	Aerolysin	Aerolysin	*Aeromonas hydrophila*	1PRE
1.C.18.1.1	Melittin	CAP	Bee venom	2MLT
1.C.19.1.1	Defensin 1	CAP	*Homo sapiens*	1DFN
1.D. Non-ribosomally synthesized channels				
1.D.1.1.1	Gramicidin A	Gramicidin	*Bacillus brevis*	1GMK
3.D. Redox-driven proton pumps				
3.D.3.2.1	Quinol:cytochrome *c* reductase	QCR	*Bos taurus*	1RIE
3.D.4.6.1	Cytochrome *c* oxidase	COX	*Paracoccus denitrificans*	1ARI
3.D.4.7.1	Cytochrome *c* oxidase	COX	*Bos taurus*	1OCC
3.E. Light-driven proton pumps				
3.E.1.1.1	Bacteriorhodopsin	BR	*Halobacterium salinarum*	1BRR
3.E.2.1.1	Reaction center	RC	*Rhodobacter sphaeroides*	1PSS

[a] Three-dimensional structural data for transporters included within TC categories 2, 3A, 3B, 3C, 4, 8, and 9 are not yet available. Structures of water-soluble domains of ABC transporters (TC 3.A.1), F-type ATPases (TC 3.A.2), P-type ATPases (TC 3.A.3), several PTS permeases (TC 4.A.1–4.A.3 and 4.A.6), and the MerTP permease (TC 9.A.2) are also available. Literature citations describing the structural data summarized in this table are available by reference to our web site.

[b] PDB code, code for the protein database containing the three-dimensional structure of this protein.

domain serves as a sterol-binding domain, and this domain is found in several receptors, and even an enzyme, 3-hydroxy-3-methylglutaryl (HMG)-coenzyme A reductase (Table 22). This provides one of the best-documented examples of a family of transporters that has truly diverged in function. As noted above, the established bacteriorhodopsin family includes sensory rhodopsins that mediate phototaxis as well as homologs in *S. cerevisiae* that probably do not function in transport (52, 67, 159). Indirect evidence suggests that most of these yeast proteins are integral membrane heat shock or organic solvent shock proteins (53, 108). They lack the conserved lysine to which retinal binds in Schiff's base linkage in the archaeal proteins. However, a homologous retinal-containing photoreceptor has recently been identified in *Neurospora crassa* (9). It is possible that the fungal chaperone proteins contain noncovalent retinal and/or energize protein folding by catalyzing proton transport through themselves.

Finally, water-soluble constituents, and possibly also the integral membrane transporter domains of both the ABC and PTS superfamilies, have been shown to function in various nontransport capacities (50, 126, 136). Thus, the extracytoplasmic receptors of ABC permeases have homologs that are domains within bacterial transcription factors (90) as well as eukaryotic neurotransmitter receptors of the glutamate-gated ion channel family (TC 1.7) (87, 141). Similarly, a few homologs of the ATP-hydrolyzing ABC proteins function in catalysis of bacterial cytoplasmic processes, and some ABC permease homologs function in regulation of other transporters (21). Bacterial PTS II.A proteins and protein domains function in regulatory processes, sometimes in addition to their transport functions and sometimes instead of their transport function (126). A few PTS transporters (II.C constituents) also serve as sensory transducers (24, 71, 78). Nevertheless, it seems surprising that so few transporter homologs function in a nontransport capacity. This fact greatly facilitates the annotation and functional assignment of putative proteins whose sequences are (or will be) revealed by genome sequencing. It also suggests that transporters evolved as a class of proteins independently of other protein types, such as enzymes, structural proteins, and regulatory proteins.

AUXILIARY TRANSPORT PROTEINS

Proteins that in some way facilitate transport across one or more biological membranes but do not themselves participate directly in transport are classified as auxiliary proteins (Table 23). These proteins by definition always function in conjunction with one or more transport proteins. They may provide a function connected with energy coupling to transport, play a structural role in complex formation, or serve a regulatory function (see section 8.A in Table 2). Examples include the membrane fusion proteins (TC 8.A.1), which provide a periplasmic bridge between primary, energy-coupled efflux permeases in the cytoplasmic membranes of gram-negative bacteria, and outer membrane factors (TC 1.B.17), which provide porin-type channel functions across the latter structures

TABLE 22. Transporter families including nontransporter homologs

Family	TC no.	Occurrence	Nontransporter homolog	TC no.	Organism
MFS	2.A.1	BAE	Low-glucose Tx sensor, Snf3p	2.A.1.1.18	*S. cerevisiae*
			High-glucose Tx sensor, Rgt2p	2.A.1.1.19	*S. cerevisiae*
			Glucose 6-phosphate Tx sensor, UhpC	2.A.1.4.4	*E. coli*
APC	2.A.3	BAE	Leucine Tx sensor, Ssy1p	2.A.3.1.20	*S. cerevisiae*
RND	2.A.6	BAE	"Patched" polarity Tx receptor	2.A.6.6.2	*D. melanogaster*
			SREBP cleavage-activating protein, SCAP	2.A.6.6.4	*C. griseus*
			HMG-CoA reductase	2.A.6.6.5	*H. sapiens*
AMT	2.A.49	BAE	NH_4^+ transporter and sensor, Mep2p	2.A.49.3.2	*S. cerevisiae*
BR	3.E.1	AE	Sensory rhodopsin	3.E.1.3.1	*H. salinarum*
			Heat and shock protein, HSP30	3.E.1.4.1	*S. cerevisiae*
ABC	3.A.1	BAE	SUR1 sulfonylurea receptor; regulator of ATP-sensitive K^+ channels	3.A.1.68.4	*H. sapiens*
			Cytoplasmic and periplasmic protein homologs function in other capacities (Tx, receptors, energizers)	—	Bacteria; animals
PTS	4.A.1–4.A.6	B	*sacX* transcriptional regulator	4.A.1.2.1	*Bacillus subtilis*
			Permeases are generally sugar kinases, but cytoplasmic proteins may function in regulation	—	Bacteria

(63, 99, 138, 157, 158). Membrane fusion protein family proteins allow solute export across both membranes of the gram-negative bacterial cell in a single energy-coupled step (10, 30, 73).

Other proteins that span the cytoplasmic membrane with large domains in the extracytoplasmic space of the gram-negative or gram-positive bacterial cell and sometimes function with additional cytoplasmic domains include members of the cytoplasmic membrane-periplasmic auxiliary 1 (MPA1; TC 8.A.3) and MPA2 (TC 8.A.4) families (33, 98, 152). These proteins are believed to function directly in export and possibly also in the regulation of complex carbohydrate export by virtue of the protein tyrosine kinase activities that are associated with their cytoplasmic domains (147).

A most interesting set of auxiliary transport proteins is the TonB family (TC 2.C.1) of heterotrimeric protein complexes that allow transmission of energy in the form of the PMF across the inner membranes of gram-negative bacteria to energize uptake of iron-siderophore complexes and vitamin B_{12} across outer membranes via proteins of the outer membrane receptor (TC 1.B.14) family. The latter proteins exhibit structural features superficially resembling those of outer membrane porins (37, 74, 99). However, they differ from typical porins in being monomeric and exhibiting 22 antiparallel β-strands in the β-barrel structure. The heterooligomeric TonB-ExbBD complex may prove to transport protons, explaining their capacity to respond to the PMF. Limited sequence similarity of these proteins to the MotAB proteins (TC 1.A.45) (A. Lupas, personal communication) further suggests this possibility.

Other auxiliary proteins include the energy-coupling proteins of the bacterial phosphoenolpyruvate-dependent sugar-transporting PTS (categories 4.A.1 to 4.A.6). Enzymes I and HPr proteins (TC 8.A.7 and 8.A.8, respectively) serve as phosphoryl transfer proteins, thereby providing both energy-coupling and enzyme-catalytic functions (104, 111). The enzymes I are homologous to phosphoenolpyruvate synthases and pyruvate:phosphate dikinases that normally function in phosphoenolpyruvate synthesis (117). We have suggested that the PTS evolved relatively late and depended on the conversion of preexisting phosphoenolpyruvate synthases into phosphoenolpyruvate-dependent phosphoryl transfer enzymes of the PTS (112).

Finally, many proteins are clearly implicated in transport, but they appear to play indirect and ill-defined roles in the process. These proteins include the rBAT (TC 8.A.9) and MinK (TC 8.A.10) family members. rBAT and MinK are believed to function in conjunction with amino acid carriers and potassium ion channels, respectively (35, 77, 116, 146). They may play roles in stability and subcellular targeting.

Many additional auxiliary proteins are included in the tables describing porters of TC categories 1 to 4. Because of their tight association with particular transport systems, they are described as constituents of these systems rather than as auxiliary proteins of the 8.A class.

TABLE 23. Families of auxiliary transport proteins

TC no.	Family
8.A.1	Membrane fusion protein (MFP) family
8.A.2	Secretin auxiliary lipoprotein (SAL) family
8.A.3	Cytoplasmic membrane-periplasmic auxiliary-1 (MPA1) protein with cytoplasmic (C) domain (MPA1−C or MPA1+C) family
8.A.4	Cytoplasmic membrane-periplasmic auxiliary-2 (MPA2) family
8.A.5	Voltage-gated K^+ channel β-subunit (VICβ) family
8.A.7	Phosphotransferase system enzyme I (EI) family
8.A.8	Phosphotransferase system HPr (HPr) family
8.A.9	rBAT (rBAT) family of putative transport accessory proteins
8.A.10	Slow voltage-gated K^+ channel accessory protein (MinK) family
8.A.11	Phospholamban (Ca^{2+}-ATPase regulator) (PLB) family
8.A.12	ABC bacteriocin exporter accessory protein (BEA) family
8.A.13	Tetratricopeptide repeat (Tpr1) family

CONCLUSIONS AND PERSPECTIVES

In this article I have described a comprehensive classification system for transport proteins that has the theoretical potential to include all transmembrane transport systems found in all living organisms on Earth. We have attempted to design this system so that it can accommodate new information and incorporate new systems as these become available with minimal alteration in structure. We have designated this system the transporter classification (TC) system of the Transport Commission of the IUBMB. This system is based on a combination of functional and phylogenetic characteristics of transporters and their constituents. The incorporation of phylogenetic data is a departure from the classification system devised by the Enzyme Commission years ago for the classification of enzymes, but the use of phylogenetic information provides many advantages, as discussed in the introductory section. Thus, phylogeny provides the most reliable guide to structure, function, and mechanism, and it provides valuable information concerning the evolutionary history of a family. The TC system should be capable of incorporating any novel type of molecular transporter that may be discovered in the future as well as the ever-increasing numbers of novel transporters that fall into existing families. Rules have been presented that allow the systematic consolidation of families as evolutionary links between them become available. Our goal is to eventually automate the incorporation of novel transporters into the system without (or with minimal) human intervention. Since the classification system is based on both function and phylogeny, achievement of this goal will require automation of tree construction as each new sequence becomes available in public databases, as well as the incorporation of biochemical, genetic, and physiological data as these become part of the scientific literature. As additional genomes are sequenced, the achievement of this goal will also require that screening techniques and annotation of novel families and family members be streamlined. In conjunction with Andrei Lupas and the bioinformatics group at SmithKline-Beecham (5), automation is now being implemented. Our web site will soon serve as a search tool that, for the analysis of transport proteins, will hopefully prove to be as useful as the BLAST search tools of the National Center for Biotechnology Information. Continual revamping of in silico methods for achieving these goals represents a major challenge that will require cooperation on the parts of computational scientists, molecular biologists, and cell physiologists. Exactly how these goals should best be achieved cannot easily be anticipated, as they are likely to be tightly coupled to technological advances through the years.

Currently recognized transporters include simple proteins as well as large multisubunit complexes that either facilitate passive diffusion of molecules across membranes or use one or more types of energy to drive transport. A large number of potential energy-yielding reactions have already been shown to be coupled to transport. These include several distinct chemical reactions, such as bond breakage reactions (e.g., decarboxylation and pyrophosphate bond hydrolysis), chemical group transfer reactions (e.g., hydride and methyl transfer), and electron flow. In addition, light absorption and the flow of ions down electrochemical gradients can be used to drive transport. In the reverse direction, ion transport can function to drive flagellar rotation, ATP synthesis, or active transport across the outer membranes of gram-negative bacteria. Variations on the established themes as well as entirely new themes are likely to be revealed by the efforts of future investigators. Perhaps, as three-dimensional structural data become available for the major classes of primary and secondary active carriers as well as group translocators, we will be able to delineate the mechanistic details of these processes. As totally new transport modes, not yet imagined, may be revealed, the transport biologist has exciting new discoveries to look forward to. The classification system proposed here, based on both function and phylogeny, is designed to accommodate any such discoveries and will hopefully aid in delineating the applicability of the structural, mechanistic, and evolutionary principles established with a few model systems to the hundreds of transporter types currently recognized and yet to be discovered.

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I wish to dedicate this treatise to my mother, Lucelia Bates Saier, in gratitude for her love, encouragement, confidence, and support.

ADDENDUM IN PROOF

Our recent unpublished results have defined a novel superfamily of secondary carriers consisting of 13 families. We have designated this superfamily the drug/metabolite transporter (DMT) superfamily (TC no. 2.A.7) (D. L. Jack and M. H. Saier, Jr., unpublished data). Some of the families included in the DMT superfamily had been included in our previous TC system, but others were previously unrecognized. The 13 currently recognized families of the DMT superfamily are as follows:

2.A.7.1—the 4 TMS small multidrug resistance (SMR) family (previously the SMR family, 2.A.7)
2.A.7.2—the 5 TMS bacterial/archaeal transport (BAT) family (previously unrecognized)
2.A.7.3—the 10 TMS drug/metabolite exporter (DME) family (previously the CAAT family, 2.A.78)
2.A.7.4—the 10 TMS plant carboxylate/amine transporter (P-CAT) family (previously unrecognized)
2.A.7.5—the 10 TMS glucose/ribose uptake (GRU) family (previously part of the RhaT family, 2.A.9)
2.A.7.6—the 10 TMS L-rhamnose transporter (RhaT) family (previously part of the RhaT family, 2.A.9)
2.A.7.7—the 10 TMS RarD (RarD) family (previously unrecognized)
2.A.7.8—the 10 TMS *Caenorhabditis elegans* ORF (CEO) family (previously unrecognized)
2.A.7.9—the 6-8 TMS triose-phosphate transporter (TPT) family (previously part of the TPNST family, 2.A.50)
2.A.7.10—the 10-12 TMS UDP-*N*-acetylglucosamine:UMP antiporter (UAA) family (previously part of the TP-NST family, 2.A.50)
2.A.7.11—the 10-12 TMS UDP-galactose:UMP antiporter (UGA) family (previously part of the TP-NST family, 2.A.50)
2.A.7.12—the 10-12 TMS CMP-sialate:CMP antiporter (CSA)

family (previously part of the TP-NST family, 2.A.50)

2.A.7.13—the 10 TMS GDP mannose:GMP antiporter (GMA) family (previously part of the TP-NST family, 2.A.50)

As homology has been established for all of these members of the DMT superfamily, they will be included under TC entry 2.A.7, and TC entry numbers 2.A.9, 2.A.50, and 2.A.58 will be assigned to other families of secondary carriers (see our website).

Recently, UreI of *Helicobacter pylori* (spQ09068) was functionally characterized (D. L. Weeks, S. Eskandari, D. R. Scott, and G. Sachs, Science **287:**482–485, 2000). UreI (and AmiS of *Pseudomonas aeruginosa* [spQ51417]) are members of the putative amide transporter (Ami) family, previously designated TC no. 9.A.15 (Tables 2 and 3). Members of this family were known to be encoded within operons that also encode amidases and ureases, and consequently these proteins were assumed to transport urea and short-chain aliphatic amides such as acetamide: (S. A. Wilson, R. J. Williams, L. H. Pearl, and R. E. Drew, J. Biol. Chem. **270:**18818–18824, 1995). Weeks et al. have shown that UreI of *H. pylori*, a 6 TMS protein of 195 amino acyl residues, forms an H^+-gated urea channel. A histidyl residue (His 123), localized to a periplasmic loop of the protein, is essential for H^+ stimulation of channel activity. UreI-mediated urea transport is urea specific, passive, nonsaturable, relatively temperature independent, and nonelectrogenic. It is the H^+-gated urea channel that regulates cytoplasmic urease, the enzyme that allows survival and colonization of the stomach by *H. pylori*. The Ami family (TC no. 9.A.15 in Tables 2 and 3) has therefore been renamed the urea/amide channel (UAC) family and assigned TC no. 1.A.45. The TC number of the Mot family has been changed from 1.A.45 to 1.A.46.

A.-M. Marini, J.-Y. Springael, W. B. Frommer, and B. André (Mol. Microbiol. **35:**378–385, 2000) have recently provided convincing evidence that the soybean SAT1 protein, which had been characterized as an NH_4^+ channel on the basis of its ability to complement an NH_4^+ transport defect in a mutant strain of *Saccharomyces cerevisiae*, is not in fact an NH_4^+ channel protein but instead is probably a transcription factor. SAT1 apparently restores NH_4^+ uptake in the yeast mutant strain by interfering with inhibition of one of the three NH_4^+ transporters of *S. cerevisiae*, Mep3 (Marini et al.). Mep3 is a member of the ammonium transporter (Amt) family (TC no. 2.A.49). TC no. 1.A.26 is therefore no longer assigned to the SAT family and has been reassigned to the plant plasmodesmata (PPD) family (see our website).

Considerable evidence is accumulating for the presence of multiple porins in the outer mycolate-containing membranes of certain high-G+C gram-positive bacteria. These bacteria include *Mycobacterium tuberculosis* (B. Kartman, S. Stengler, and M. Niederweis, J. Bacteriol. **181:**6543–6546, 1999; R. Senaratne et al., J. Bacteriol. **180:**3541–3547, 1998), *Mycobacterium smegmatis* (M. Niederweis et al., Mol. Microbiol. **33:** 933–945, 1999; C. Raynaud et al., Microbiology **145:**1359–1367, 1999), *Mycobacterium bovis* (T. Lichtinger et al., FEBS Lett. **454:**349–355, 1999), *Nocardia farcinica* (F. G. Riess et al., Mol. Microbiol. **29:**139–150, 1998), *Nocardia asteroides* (F. G. Riess et al., Arch. Microbiol. **171:**173–182, 1999), and *Rhodococcus erythropolis* (T. Lichtinger, G. Reiss, and R. Benz, J. Bacteriol. **182:**764–770, 2000). One of these proteins is the OmpATb protein of *M. tuberculosis*, which has been reported to be a member of the OmpA-OmpF porin (OOP) family (TC no. 1.B.6.1.3; see our website); MspA of *M. smegmatis*, another such protein, is a member of a novel family which we have called the mycobacterial porin (MBP) family (TC no. 9.B.24) (M. Niederweis et al., 1999). A third such protein is a partially sequenced protein from *Rhodococcus erythropolis* which we have provisionally referred to as the *R. erythropolis* porin (REP; TC no. 9.C.3) (Lichtinger et al., 2000). The partial sequence available for the latter protein does not exhibit significant similarity to any sequence in the current databases.

The available sequence data suggest that the outer membrane porins of gram-positive bacteria will prove to belong to several distinct families. Although the few fully sequenced proteins currently available from mycolate-containing membranes have been placed under category 1.B (β-barrel porins), it should be noted that structural data are not yet available for any of these proteins. Consequently, they may prove to be more appropriately assigned to a different category in the future.

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