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Transport ATPases in Biological Systems and Relationship to Human Disease: A Brief Overview

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Interest in the field of transport ATPases has grown dramatically during the past 20 years and gained considerable visibility for several reasons. First, it was shown that most transport ATPases can be lumped into only a few categories designated simply as P, V, F, and ABC types, the latter consisting of a large superfamily. Second, it has been shown that many transport ATPases have a clear relevance to human disease. Third, the field of transport ATPases has become rather advanced in the study of the reaction mechanisms and structure–function relationships associated with several of these enzymes. Finally, the Nobel committee recently recognized major accomplishments in this field of research. Here, the author provides a brief discussion of transport ATPases that are present in biological systems and their relevance or possible relevance to human disease.

KEY WORDS: ATPase; transport ATPase; ion-motive ATPases; P-type ATPase; V-type ATPase; F-type ATPase; ATP synthase; ABC transporters; human disease.

INTRODUCTION

The field of transport ATPases had its origin in Denmark, where a Danish scientist named Jens Christian Skou, working on crab nerves in the Department of Physiology at the University of Aarhus, reported his finding more than four decades ago (Skou, 1957). In the paper entitled "The Influence of Some Cations on the Activity of an Adenosine Triphosphatase in Peripheral Nerves" the Na^+/K^+ ATPase became known to the scientific world for the first time. Forty years later, Skou was awarded the Nobel prize.

Following the discovery of the Na^+/K^+ ATPase, many other transport ATPases were discovered. To consolidate thinking under one scientific umbrella about how these ATPases work, 30 years later Carafoli and the author published two papers entitled "Ion Motive ATPases I. Ubiquity, Properties, and Significance to Cell Function" (Pedersen and Carafoli, 1987a) and "Ion Motive ATPases: Energy Coupling and Work Output" (Pedersen and Carafoli, 1987b). Wherein, we indicated that all known transport ATPases at the time could be lumped into three categories, which we named P-, V-, and F-type ATPases, for simplicity. P-type ATPases are those that catalyze reactions proceeding through a covalent phosphorylated "P" intermediate; V-type ATPases are those found associated with vacuoles; and F-type ATPase are those referred to as F_0F_1 ATPases, and now also referred to as ATP synthases. Subsequently, a number of scientific meetings were organized, which brought scientists from around the world to one location to discuss under the same roof their recent work on P-, V-, and F-type ATPases, a discussion format that has now been expanded to include the ABC-type ATPases, i.e., members of the ABC transporter superfamily (Higgins, 1992).

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DISCUSSION

A Current List of Well-Known ATPase Types

Shown in Fig. 1 is a hypothetical human cell showing some of the most well-known ATPase types found in biological systems. These include the transport-type

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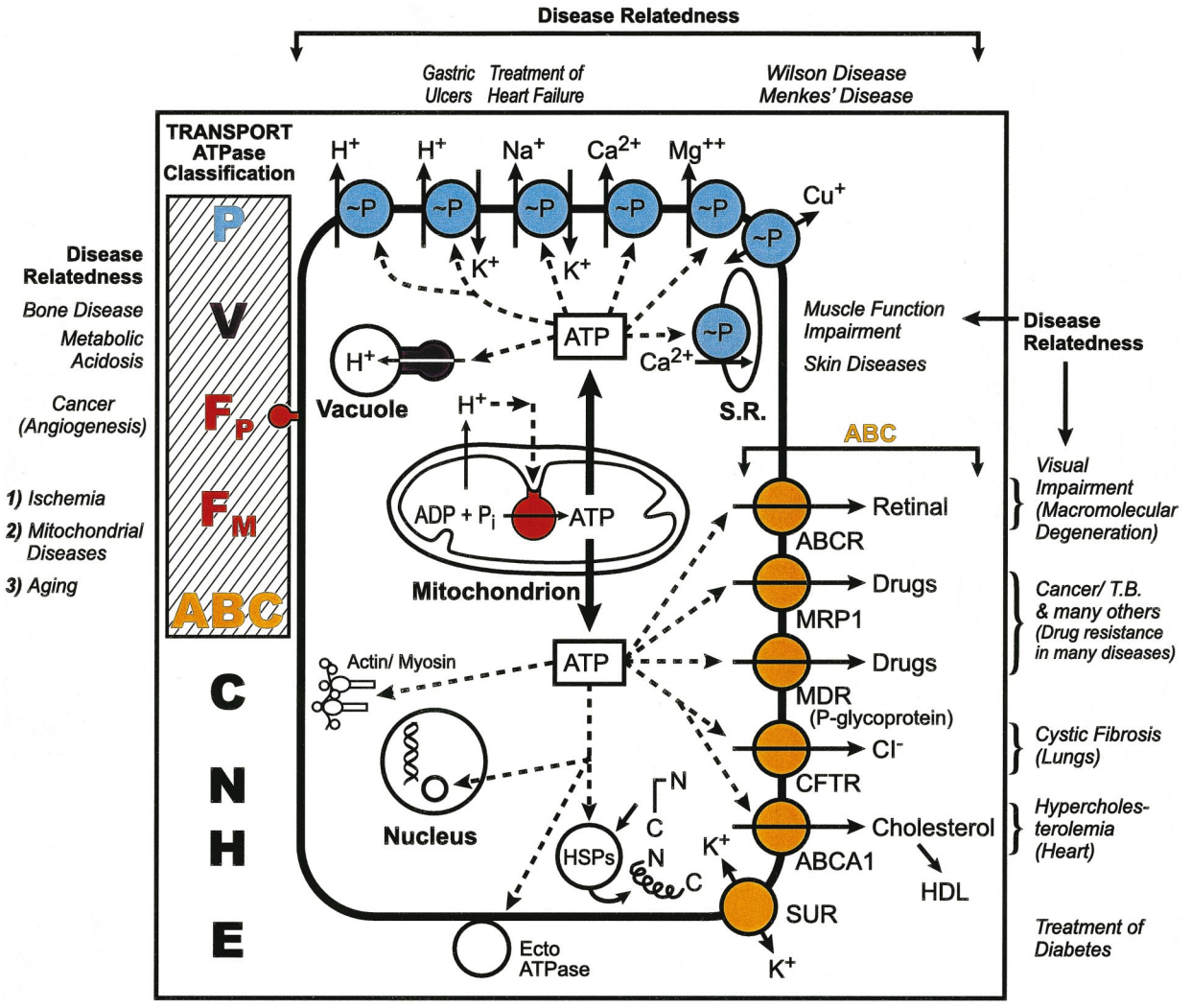

Fig. 1. Listing of The Major ATPases in Biological Systems. ATPases in biological systems can be subdivided into those that are involved in transport of ions or metabolites across biological membranes, i.e., "Transport ATPases" and those that are associated with cellular functions that do not involve a membrane. This minireview series focuses on the "Transport ATPases," the P, V, F, and ABC types, and their relevance to human disease. The central part of the figure shows which ions or metabolites are transported by the "Transport ATPases" while near the edges of the Figure is summarized the disease relevance of each of the ATPase types. Details are provided in the text of the review.

ATPases P, V, F, and ABC noted earlier together with non-transport-type ATPases designated here as C, N, H, and E. Here, C refers to myosin-like ATPases involved in contraction, N refers to ATPases involved in bringing about changes in nucleic acids, H refers to ATPases in the heat shock protein family, and E refers to ecto ATPases or those located on the cell surface. It is likely that this list is not complete and that other ATPase types are yet to be discovered, or have already been discovered but remain unknown to the author.

The F-type ATPase has been considered to be the "master" (Pedersen and Carafoli, 1987a) as it normally works from its mitochondrial location in human cells in the direction of ATP synthesis providing all other ATPases, i.e., the "workhorse ATPases" with the necessary ATP they need to do the cellular work associated with transport. In some cell types, e.g., neuronal cells, a large fraction of the total ATP produced by the mitochondrial F-type ATPase, acting as an ATP synthase, is hydrolyzed to drive ion transport.

The first diffraction quality crystals of a transport ATPase catalytic unit were obtained more than two decades ago with the F_1 moiety of rat liver F_0F_1 ATP synthase (Amzel and Pedersen, 1978). Years later an atomic

resolution structure was obtained for this F_1 preparation (Bianchet *et al*., 1998) and also for bovine heart and thermophilic bacterium F_1 preparations (Abrahams *et al*., 1994; Shirakihara *et al*., 1997). More recently atomic resolution structures have been obtained also for the Ca^{+2} P-type ATPase (Toyoshima *et al*., 2000), and for two bacterial ABC transporters (Chang and Roth, 2001; Locher *et al*., 2002). The first structure, i.e., of bovine heart F_1, was obtained by a team led by John Walker (Abrahams *et al*., 1994), who received the Nobel prize in 1997, the same year in which Jens Skou and Paul Boyer received the prize.

Because of the above landmark structural studies, the field of transport ATPases has been able to propose very detailed mechanisms (e.g., see Ko *et al*., 1999; Pedersen *et al*., 2000a; Senior *et al*., 2002; Scarborough, G.A. (2002)). Major challenges for the future are to obtain an atomic resolution structure for each of the following: V-type ATPase, an ABC transporter from an animal system, in particular CFTR, and a complete F-type ATPase (i.e., F_0F_1 ATP synthase/ATPase complex).

Relevance to Human Disease

Known or possible relevance to human disease is considered briefly below for each of the major transport ATPase types.

P-Type ATPases

The Na^+/K^+ P-type ATPase (Kaplan, 2002) regulates the sodium–potassium balance of animal cells. It has for many years believed to be the major site of action of digitalis glycosides used for the treatment of congestive heart failure (Kjeldsen *et al*., 2002). Moreover, there are some views that interaction of subtoxic digitalis concentrations reduce maximum sodium transport (Reviewed in Smith, 1984). Although this is an area that remains controversial, the role of the Na^+/K^+ ATPase in heart function and in the treatment of heart failure remains a topic of considerable interest and investigation.

The Na^+/K^+ ATPase's closest relative is perhaps the H^+/K^+ ATPase that is involved in acid secretion in the stomach. Some of the most potent gastric acid suppressing agents in clinical use today act at the level of this P-type ATPase. Among the inhibitors that inhibit this ATPase are omeprazole, lansoprazole, and pantoprazole (Sachs, 1997). These agents are believed to be more effective than histamine two-receptor antagonists in controlling gastric secretion and therefore for treating gastric ulcers. These agents are used also in combination with antibacterial agents to treat *Helicobacter pylori*-positive duodenal ulcerations (Richardson *et al*., 1998).

The Ca^{+2} ATPase of the sarcoplasmic reticulum (SERCA), long known to play a central role in the contraction–relaxation cycle of muscle (Hussain and Inesi, 1999; Maclennan and Green, 2000), has been implicated in the human disease known as Brody disease (MacLennan *et al*., 1998; MacLennan, 2000; MacLennan and Green, 2000). In some forms of this disease there is loss of SERCA function causing exercise-induced impairment of the relaxation of skeletal muscle. Significantly, in about half of Brody disease families, mutations create stop codons that delete all or part of the Ca^{+2} binding and translocation domain, resulting in loss of SERCA1 function and muscle disease. Interestingly, two other diseases have been linked to mutations in P-type ATPase(s) related to SERCA. These are Hailey-Hailey disease, an autosomal dominant skin disorder characterized by suprabasal cell separation of the epidermis (Dobson-Stone *et al*., 2002), and Darier's disease (Takahashi *et al*., 2001), a rare dominantly inherited skin disorder with abnormal keratinization, and also suprbasal cell separation of the epidermis. In contrast to SERCA and SERCA-related P-type ATPases, there have been no mutations linked to human diseases in the four genes encoding plasma membrane Ca^{+2} ATPases.

Copper P-type ATPases, one of the subjects of this minireview series, are just beginning to gain recognition for their very important roles in a number of biological processes (Cox, 1999). These P-type ATPases appear to be unique relative to the other ATPase types shown in Fig. 1 as they frequently operate in conjunction with so-called "metallochaperones" that deliver copper to them (O'Halloran and Culotta, 2000). These copper chaperones, also a subject of this minireview series, are evidently essential as copper is in short supply in the cell and, if allowed to pass freely into the cytosol, it may reach toxic levels. Significantly, two different copper P-type ATPases, encoded by the ATP7A and ATP7B genes have been linked to human disease. Thus, mutations in the ATP7A gene result in Menkes disease and mutations in the ATP7B gene result in Wilson's disease. Interestingly, Menkes disease is associated with systemic copper deficiency whereas in Wilson's disease copper accumulates in the tissues. This accumulation can be diagnostic for Wilson's disease, particularly in the eye where a brown ring may form around the cornea.

V-Type ATPases

V-type ATPases are closely related to the F-type ATPases as they exhibit a large number subunits and share sequence similarities (Nishi and Forgac, 2002). Also, they

are believed to operate via a mechanism similar to that of F-type ATPases. However, V-type ATPases work exclusively as proton translocating ATPases, whereas in animal cells F-type ATPases (F_0F_1) work primarily during cell life as proton driven ATP synthases. V-type ATPases have been labeled recently as "nature's most versatile proton pumps" as they participate in a wide variety of cellular processes including endocytosis, intracellular transport, membrane fusion, bone resorption, and renal acid–base balance (Nishi and Forgac, 2002). Mutations in V-type ATPase isoforms involved in the latter two processes have been linked to certain pathologies, one of which is osteopetrosis (Frattini *et al*., 2000; Kornak *et al*., 2000) and the other metabolic acidosis (Smith *et al*., 2000). The former disease results because the normal bone resorption mechanism is impaired while the latter disease results when normal renal acid–base balance can no longer be maintained. Finally, V-type ATPases associated with the plasma membrane of tumor cells have been proposed to have a role in metastasis (Martinez-Zagulian *et al*., 1993).

F-Type ATPases

As indicated above the main role of the mitochondrial F_M-type ATPase (F_0F_1), commonly referred to in the recent literature as "ATP synthase," is to make ATP at the expense of an electrochemical gradient of protons generated by the electron transport chain (Pedersen *et al*., 2000b; Senior *et al*., 2002). The mitochondrial ATP synthase is extremely complicated consisting of 16–17 subunit types (Pedersen *et al*., 2000a), two of which are regulatory proteins, Factor B and IF_1. Factor B appears to be an activator of ATP synthase (Bulogrudov and Hatefi, 2002) whereas IF_1 is an inhibitor of the reverse reaction, i.e., ATP hydrolysis (Schwerzmann and Pedersen, 1986). Recent work in a number of laboratories is consistent with the view that ATP synthases consist of two motors, one of which is within F_0 (Oster and Wang, 1999; Yoshida *et al*., 2001). This motor is driven by the electrochemical gradient of protons generated by the electron transport chain. It drives the rotation of a central helical subunit (γ) that transverses the central core of the second motor, the F_1 ATPase catalytic sector (Abrahams *et al*., 1994; Bianchet *et al*., 1998). Then, the mechanically transduced energy alters the binding affinities sequentially at three equally spaced catalytic sites causing $ADP + P_i$ at each site to form ATP. In effect, the proton driven motor within F_0 drives the ATP hydrolysis driven motor within F_1 in the reverse direction.

A major health problem that affects numerous people is ischemia, lack or deficiency of oxygen that can lead to cardiac arrest or stroke. Such hypoxic conditions that slow or arrest the oxygen-dependent mitochondrial electron transport chain driving ATP synthesis now cause the ATP synthase to work in the opposite direction, i.e., as an ATPase. The resultant ATP hydrolysis depletes the cell's ATP supply sending it into cell death (Pedersen, 1999). There has been great interest in the molecular events that take place during the ischemic process as its prevention by therapeutic intervention could potentially save many lives. Considerable research on the subject has revolved around the inhibitor protein IF_1 that does try to stop the process but falls short of really doing a complete job (Das and Harris, 1990; Rouslin, 1991). Ischemic "preconditioning" of the heart, which provides some protection against heart failure, has also been an area of intense interest as it may induce regulatory mechanisms that slow ATP synthase's reverse reaction, ATP hydrolysis (Belisle and Kowaltowski, 2002; Pucar *et al*., 2001).

In addition to ischemia, there are also some mitochondrial diseases that have been traced to the F_M-type ATPase (ATP synthase). Thus, some cases of Leigh's syndrome and Leber hereditary optic neuropathy have been traced to mutations in the F_0 motor of the ATP synthase (Carelli *et al*., 2002; Lamminen *et al*., 1995; Oritz *et al*., 1993). This motor is composed of three subunit types called *a*, *b*, and *c*, with the *a* and *c* subunits containing the proton translocation paths that funnel energy to the F_1 unit to drive ATP synthesis via the rotorary catalytic process described above. Specifically, mutations in subunit *a* are involved in some forms of the diseases noted above.

It should be noted also that in a number of theories of ageing (Lenaz, 1998) the ATP synthase has been implicated, presumably because there is a propensity to accumulate mutations in mitochondrial DNA during one's lifetime. As mitochondrial DNA encodes several proteins within the major complexes involved in oxidative phosphorylation, including ATP synthase, such mutations are believed to impair energy production. Such theories remain of considerable interest but much more research is required to rigorously test them.

Finally, two recent reports of an ATP synthase-like molecule (F_p-type) located on the plasma membrane of endothelial cells, i.e., cells lining the blood vessel, have attracted some attention because of the possible relationship of the enzyme at this location to angiogenesis and cancer pathogenesis (Moser *et al*., 1999, 2001). During cancer progression, tumors stimulate the growth of nearby blood vessels (angiogenesis) causing the vessels to reach the tumor, vascularize it, and subsequently provide it with the nutrients needed to mature rapidly, metastasize (spread), and ultimately kill the host. The investigators studying this enzyme have shown that it makes ATP and serves

as a receptor for endostatin, an inhibitor of angiogenesis. Certainly, it will be interesting to see how studies on this novel ATP synthase-like enzyme "play out" in the future.

ABC-Type ATPases

Clearly, the ABC-Type ATPases, commonly referred to as ABC transporters, are the most abundant of all the transport ATPases (Higgins, 1992). In humans almost 50 of these transporters are known (Dean and Allikmet, 2001). Here, they exist most commonly as long, single polypeptide chains consisting of four domains, two transmembrane domains that form either a transport path or a channel, and two nucleotide domains (NBF1 and NBF2) that hydrolyze ATP to facilitate substrate efflux through the transport path (Gottesman and Ambudkar, 2001; Higgins, 1992).

Current evidence, particularly with the multidrug resistant protein known either as P-glycoprotein or multidrug resistant protein 1 (MDR1), suggests that ATP is hydrolyzed at both NBFs in an alternating catalytic site manner in order for the transporter to function fully (Sauna *et al*., 2001; Senior *et al*., 1995). Although the hydrolytic event at the catalytic site of one NBF may be facilitated by amino acid residues derived from the other, such interactions do not appear to be necessary for basal ATPase activity. Unique among the human ABC transporters is CFTR (cystic fibrosis transmembrane conductance regulator), a gated chloride channel, as it contains a fifth or "regulatory" domain (R) that is subject to phosphorylation by protein kinases (Ko and Pedersen, 2001; Riordan *et al*., 1989). CFTR is also predicted to operate by an alternating catalytic site mechanism (Senior and Gadsby, 1997).

The disease relevance of ABC Transporters is widespread (Dean and Allikmets, 2001; Gottesman and Ambudkar, 2001). Some like MDR1, MRP1, and related "drug efflux" transporters, may pose a serious problem for the continuous treatment of a given disease with an initially effective drug. The most commonly used example of this is in the treatment of cancer. In contrast, other ABC transporters may facilitate the treatment of a given disease as in the case of SUR an ATP-dependent K^+ channel (Aguilar-Bryan and Bryan, 1999; Gribble and Ashcroft, 2000). Sulfonyl ureas by interacting with SUR are widely used to stimulate insulin secretion from pancreatic β cells in the treatment of Type 2 diabetic patients. Finally, and most commonly, mutations in some other ABC transporters may cause disease. There are at least 10 examples of this (Dean and Allikmets, 2001) including CFTR (ABCC7) involved in Cystic Fibrosis, ABCR (ABCA4) involved in Stargardt disease (aged related macular degeneration), ABCA1 (ABC1) involved in Tangier disease (familial HDL deficiency), and ALD (ABCD1) involved in adrenoleukodystrophy. Curing or ameliorating the symptoms of these diseases, either by intervention with appropriate drugs that correct the structural problems caused by the mutation(s) or by replacing via gene therapy the defective protein, will be major challenges in the twenty-first century.

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REFERENCES

Abrahams, J. B., Leslie, A. G. W., Lutter, R., and Walker, J. E. (1994). *Nature (London)* **370**, 621–628.
Amzel, L. M., and Pedersen, P. L. (1978). *J. Biol. Chem.* **253**, 2067–2069.
Aquilar-Bryan, L., and Bryan, J. (1999). *Edocr. Rev.* **20**, 101–135.
Belisle, E., and Kowaltowski, A. J. (2002). *J. Bioenerg. Biomemb.* **34**, 285–298.
Belogrudov, G. I., and Hatefi, Y. (2002). *J. Biol. Chem.* **277**, 6097–6103.
Bianchet, M., Hullihen, J., Pedersen, P. L., and Amzel, L. M. (1998). *Proc. Natl. Acad. Sci. USA* **95**, 11065–11070.
Carelli, V., Bracca, A., Barogi, S., Pallotti, F., Valentino, M. L., Montagna, P., Zeviani, M., Pini, A., Lenaz, G., Baruazzi, A., and Solaini, G. (2002). *Arch. Neurol.* **59**, 264–270.
Chang, G., and Roth, C. B. (2001). *Science* **293**, 1793–1800.
Cox, D. W. (1999). *Br. Med. Bull.* **55**, 544–555.
Das, A. M., and Harris, D. A. (1990). *Biochem. J.* **266**, 355–361.
Dean, M., and Allikmets, R. (2001). *J. Bioenerg. Biomemb.* **33**, 475–479.
Dobson-Stone, C., Fairclough, R., Dunne, E., Brown, J., Dissanayake, M., Munro, C. S., Strachan, T., Burge, S., Sudbrak, R., Monaco, A. P., and Hovnanian, A. (2002). *J. Invest. Dermatol.* **118**, 338–343.
Frattini, A., Orchard, P. J., Sobacchi, C., Giliani, S., Abinum, M., Mattson, J. P., Keeling, D. J., Andersson, A. K., Wallbrandt, P., Zecca, L., Notarangelo, L. D., Vezzoni, P., and Villa, A. (2000). *Nat. Genet.* **25**, 343–346.
Gottesman, M., and Ambudkar, S. V. (2001). *J. Bioenerg. Biomemb*. **33**, 453–458.
Gribble, F. M., and Aschcroft, F. M. (2000). *Metabolism* **49**, 3–6.
Higgins, C. F. (1992). *Annu. Rev. Cell Biol.* **8**, 67–113.
Hussain, A., and Inesi, G. (1999). *J. Memb. Biol.* **172**, 91–99.
Kaplan, J. H. (2002). *Annu. Rev. Biochem.* **71**, 22–37.
Kjeldsen, K., Norgaard, A., and Gheorghiade, M. (2002). *Cardiovas. Res.* **55**, 710–713.
Ko, Y. H., Hong, S., and Pedersen, P. L. (1999). *J. Biol. Chem.* **274**, 28853–28856.
Ko, Y. H., and Pedersen, P. L. (2001). *J. Bioenerg. Biomemb.* **33**, 513–521.
Kornak, U., Schulz, A., Freidrich, W., Uhlhaas, S., Kremens, B., Voit, T., Hasan, T., Bode, U., Jentsch, T. J., Kubisch, C. (2000). *Hum. Mol. Genet.* **9**, 2059–2063.
Lamminen, T., Majander, A., Juvonen, V., Wikstrom, M., Aula, P., Nikoskelainen, E., and Savontous, M.L. (1995). *Am. J. Hum. Genet.* **56**, 1238–1240.
Lenaz, G. (1998). *Biochem. Biophys. Acta* **1366**, 53–67.
Locher, K. P., Lee, A., and Rees, D. C. (2002). *Science* **296**, 1091–1098.
MacLennan, D. H. (2000). *Eur. J. Biochem.* **267**, 5291–5297.
MacLennan, D. H., and Green, N. M. (2000). *Nature* **405**, 633–634.
MacLennan, D. H., Rice, W. J., Odermatt, A., and Green, N. M. (1998). *Acta Physiol. Scand. Suppl.* **643**, 55–67.

Martinez-Zagulian, R., Lynch, R., Martinez, G., and Gilles, R. (1993). *Am. J. Physiol*. **265**, C1015–C1029.
Moser, T. L., Stack, M. S., Asplri, I., Enqhild, J. J., Hojrup, P., Everitt, L., Hubchak, S., Schapner, H. P., and Pizzo, S. V. (1999). *Proc. Natl. Sci.* U.S.A. **96**, 2811–2816.
Moser, T. L., Kenan, D. J., Ashley, T. A., Roy, J. A., Goodman, M. D., Misra, U. K., Cheek, D. J., and Pizzo, S. V. (2001). *Proc. Natl. Acad. Sci. U.S.A.* **98**, 6656–6661.
Moser, T. L., Stack, M. S., Asplin, I., Enghild, J. J., Hojrup, P., Everitt, L., Hubchak, K. S., Schnaper, H. W., and Pizzo, S. V. (1999). *Proc. Natl. Acad. Sci. U.S.A.* **96**, 2811–2816.
Nishi, T., and Forgac, M. (2002). *Nat. Rev. Mol. Cell. Biol*. **3**, 94–103.
O'Halloran, T. V., and Culotta, V. C. (2000). *J. Biol. Chem*. **275**, 25057–25060.
Ortiz, R. G., Newman, N. J., Shoffner, J. M., Kaufman, A. E., Koontz, D. A., and Wallace, D. C. (1993). *Arch. Ophthamol*. **111**, 1525–1530.
Oster, G., and Wang, H. (1999). *Structure Fold Des.* **7**, R67–R72.
Pedersen, P. L. (1999). *J. Bioenerg. Biomemb.* **31**, 291–304.
Pedersen, P. L., and Carafoli, E. (1987a). *Trends Biochem. Sci.* **12**, 146–150.
Pedersen, P. L., and Carafoli, E. (1987b). *Trends Biochem. Sci.* **12**, 186–189.
Pedersen, P. L., Ko, Y. H., and Hong, S. (2000a). *J. Bioenerg. Biomemb.* **32**, 423–432.
Pedersen, P. L., Ko, Y. H., and Hong, S. (2000b). *J. Bioenerg. Biomemb.* **32**, 325–332.
Pucar, D., Dzeja, P. P., Bast, P., Juranic, N., Macura, S., and Terzic, A. (2001). *J. Biol. Chem.* **276**, 44812–44819.
Richardson, P., Hawkey, C. J., and Stack, W. A. (1998). *Drugs* **56**, 307–335.
Riordan, J. R., Rommens, J. M., Kerem B., Alon, N., Rozmahel, R., Grzelczak, Z., Zielenski, J., Lok, S., Plavsic, N., Chou, J. L., Drumm, M. L., Iannuzzi, M. C., Collins, F. S., and Tsui, L.-C. (1989). *Science* **245**, 1066–1073.
Rouslin, W. (1991). *J. Bioenerg. Biomemb.* **23**, 873–888.
Sachs, G. (1997). *Pharmacotherapy* **17**, 22–37.
Sauna, Z. E., Smith, M. M., Muller, M., Kerr, K. M., and Ambudkar, S. V. (2001). *J. Bioeneg. Biomembran.* **33**, 481–491.
Scarborough. G. A. (2002). *J. Bioenerg. Biomemb.* **34**, 235–250.
Schwerzmann, K. and Pedersen, P. L. (1986). *Arch. Biochem. Biophys.* **250**, 1–18.
Senior, A. E., al-Shawi, M. K., and Urbatsch, I. L. (1995). *FEBS Lett.* **377**, 285–289.
Senior, A. E., and Gadsby, D. C. (1997). *Semin. Cancer Biol.* **8**, 143–150.
Senior, A. E., Nadanaciva, S., and Weber, J. (2002) *Biochem. Biophys. Acta* **1553**, 188–211.
Shirakihara, Y., Leslie, A. G. W., Abrahams, J. P., Walker, J. E., Udea, T., Sekimato, Y., Kambara, M., Sarka, K., Kagawa, Y., and Yoshida, M. (1997). *Structure* **5**, 825–836.
Skou, J. C. (1957). *Biochim. Biophys. Acta* **23**, 394–401.
Smith, T. W. (1984). *J. Pharmacol*. **15** (Suppl. 1) 35–51.
Smith, A. N., Skaug, J., Choate, K. A., Nayir, A., Bakkaloglic, A., Ozen, S., Hulton, S. A., Sanjad, S. A., Al-Sabban, E. A., and Lifton, R. P. (2000). *Nat. Genet.* **26**, 71–75.
Takahashi, H., Atsuta, Y., Sato, K., Ishida-Yamamoto, A., Suzuki, H., and Iizuka, H. (2001). *J. Dermatol. Sci.* **26**, 169–172.
Toyoshima, C., Nakasako, M., Nomura, H., and Ogawa, H. (2000). *Nature* **405**, 647–655.
Yoshida, M., Muneyuhi, E., and Hisabori, T. (2001). *Nat. Rev. Mol. Cell. Biol.* **2**, 669–677.