Anion exchanger proteins facilitate the exchange of bicarbonate for chloride across the plasma membrane. When bicarbonate combines with a proton it undergoes conversion into CO$_2$, either spontaneously, or catalyzed by carbonic anhydrase enzymes. The CO$_2$/HCO$_3^-$ equilibrium is the body's central pH buffering system. Rapid bicarbonate transport across the plasma membrane is essential to maintain cellular and whole body pH, to dispose of metabolic waste CO$_2$, and to control fluid movement in our bodies. Cl$^{-}$/HCO$_3^-$ exchangers are found in two distinct gene families: SLC4A and SLC26A. Differences in the tissue distribution, electrogenicity and regulation of the specific anion exchanger proteins allow for precise regulation of bicarbonate transport throughout the human body. This review provides a look into the structural and functional features that make this family of proteins unique, as well as the physiological significance of the different anion exchangers.

**Introduction**

The by-product of mitochondrial respiration is the acid, CO$_2$, which must be removed from the body to maintain pH homeostasis. Our bodies have developed mechanisms to deal with metabolic acid build-up including: CO$_2$ exhalation through the lungs, H$^+$ secretion and HCO$_3^-$ reabsorption in the kidney, and an extensive buffering system to neutralize acid. CO$_2$ is in equilibrium with bicarbonate through the reaction CO$_2$ + H$_2$O ⇌ HCO$_3^-$ + H$^+$. This reaction has a pKa of 6.2, which is close to the physiological pH of 7.2. Thus, the CO$_2$/HCO$_3^-$ equilibrium forms the main pH buffering system in our bodies, since there is a significant amount of CO$_2$ and HCO$_3^-$. Since CO$_2$ is a gas, it can freely move across the plasma membrane; however, HCO$_3^-$ is a membrane impermeant anion. In order to transport HCO$_3^-$ across the plasma membrane, bicarbonate transporter proteins are required. One of the major classes of bicarbonate transporters is the Cl$^-$/HCO$_3^-$ exchangers, also known as anion exchangers. The other functional class of bicarbonate transporters are the sodium bicarbonate co-transporters,$^1$ which will not be discussed in this review.

Anion exchangers move bicarbonate either into or out of the cell in exchange for chloride, thereby either alkalinizing or acidifying the cell, respectively. Anion exchangers not only maintain physiological pH, but are also involved in volume regulation and acid/base secretion. There are a wide range of differences between the members of this family, including differences in tissue distribution, apical or basolateral epithelial expression, electrogenicity, regulation and physiological roles. Due to the importance of these proteins, it is not surprising that they are involved in many diseases including cardiac hypertrophy, hereditary spherocytosis of the erythrocyte, proximal and distal renal tubular acidosis, goiter, epilepsy, Pendred syndrome and cystic fibrosis.$^1$ Most of the mutations that cause this wide range of diseases are the result of endoplasmic reticulum (ER)-retained phenotypes. This may cause a dominant pattern of inheritance, because heterodimers of wild-type and mutant proteins may become ER-retained.$^1$

Anion exchanger proteins have evolved from two distinct gene families, SLC4A and SLC26A (Fig. 1). The Human Genome Organization (HUGO) has termed these proteins solute carrier transport proteins and given them the nomenclature “SLC”.$^2$ The SLC4A family contains sodium bicarbonate co-transporters (NBCs), sodium dependent chloride bicarbonate transporters (NDCBE and NCBE), and sodium independent chloride bicarbonate transporters (AE1, AE2 and AE3). The SLC26A family supports chloride bicarbonate exchange, in some cases with an electrogenic mechanism. There are five different Cl$^-$/HCO$_3^-$ exchangers in this family (SLC26A3, Pendrin, SLC26A6, SLC26A7 and SLC26A9). The confusing nomenclature of the SLC4A and SLC26A families stems from the discovery of these proteins over a 20 year span.$^3,4$ In many cases the same protein was discovered by independent researchers and thus given different names. In Table 1 we have listed all of the anion exchanger proteins by their accepted names, as well as other names given to these proteins. In this paper we have

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used the generally accepted name for all of the proteins that will be discussed. In addition, Table 1 provides a summary of the tissue distribution, substrate specificity and electrogenicity of the Cl/HCO₃⁻ exchangers. The substrate specificity and electrogenicity of some of the Cl/HCO₃⁻ exchangers is still under debate and Table 1 is based on our interpretation of the literature.

**Electroneutral Sodium Independent Cl⁻/HCO₃⁻ Exchangers of the SLC4A Family**

The AE1-3 genes all have a shared architecture. The first anion exchanger gene to be cloned was mouse AE1 in 1985.⁴ AE2, and AE3 were subsequently cloned using probes based on the murine AE1 sequence.⁵,⁶ In addition to the similarities found in the AE1-3 genes, the encoded proteins all share common features. One of these is the ability to perform reversible electoneutral exchange of Cl⁻ for HCO₃⁻, since AE1-3 are simply driven by substrate gradients. This has been demonstrated in HEK293 cells transfected with AE1-3, where the efflux and influx of bicarbonate was measured during changes in chloride gradients.⁷

AE1, AE2 and AE3 are composed of three domains: the N-terminal domain, the transmembrane domain and the C-terminal domain. Both the N and C-terminal domains are cytosolic, and vary in length depending on the isoform. The membrane domain of AE1-3 alone carries out the Cl⁻/HCO₃⁻ exchange function. The membrane domain is approximately 500 residues long, and is the region where the proteins share the highest sequence similarity at nearly 80%.⁸ The exact number of transmembrane segments is unknown, but through hydropathy analysis of the mouse AE1 amino acid sequence, it is hypothesized to be between 12–14 transmembrane segments.⁹ The ambiguity results from possible re-entrant regions in the C-terminal half of the membrane domain (Fig. 2). AE1-3 are all post-translationally modified. AE1 is glycosylated on its fourth extracellular loop at Asn642, whereas AE2 and AE3 are glycosylated on their third extracellular loop.⁴,¹⁰

The C-terminal cytoplasmic domain of all the SLC4A anion exchangers contains a ~40 residue acidic motif involved in interactions with carbonic anhydrases. This interaction with carbonic anhydrase forms the HCO₃⁻ transport metabolon.¹¹,¹² A metabolon is a complex of weakly associated enzymes which interact physically and functionally to facilitate substrate channeling and maximize enzyme kinetics.¹³,¹⁴ Carbonic anhydrases catalyze the conversion of CO₂ + H₂O ↔ HCO₃⁻ + H⁺, and in doing so, they provide anion exchangers with bicarbonate. This interaction increases the flux of HCO₃⁻ through the transporter by about 40%.¹²

AE1 (SLC4A1, band 3). AE1 is expressed as two isoforms, eAE1 (911 residues) expressed in erythrocytes, and kAE1 (846 residues) expressed on the basolateral surface of α-intercalated cells of the renal collecting duct. The two isoforms differ because eAE1 has an additional 65 N-terminal amino acids that are required to establish protein-protein interactions with the erythrocyte cytoskeleton.¹⁵

The approximately 10⁶ copies of AE1 in an erythrocyte comprise 50% of the integral membrane protein of these cells.¹⁶ With the high abundance of AE1 in red blood cells (RBCs), it is not surprising that AE1 fulfills important roles. Firstly, AE1 is a key player in the CO₂/HCO₃⁻ buffering system of the blood, and is thus involved in CO₂ exhalation. When tissues produce CO₂ during mitochondrial respiration, the CO₂ diffuses out of these cells into the capillaries. Once CO₂ is in the blood stream it diffuses into erythrocytes where it is converted into HCO₃⁻ by carbonic anhydrase II (CAII). To avoid buildup of bicarbonate in the red blood cell, AE1 transports the bicarbonate out of the red blood cell into the plasma. Once the blood reaches the lungs this process is reversed. Bicarbonate is transported back into the red blood cell by AE1, where it is converted back into CO₂ by CAII. Finally, CO₂ is exhaled through the lungs. The exchange activity of AE1 is vitally important because it maximizes the blood’s CO₂/HCO₃⁻ carrying capacity, which may be the rate limiting step in cardiovascular performance.¹⁷ To ensure maximum transport of bicarbonate through AE1, the protein physically interacts with CAII through its “DADD” motif in the cytosolic C-terminus (Fig. 2).¹¹,¹⁸ As discussed earlier this forms the bicarbonate transport metabolon, which significantly increases the rate of bicarbonate transport. Maximizing the rate of bicarbonate transport is of special importance in the bloodstream since erythrocytes pass through a capillary in only 0.3 to 1.0 s.

The second major function of AE1 in RBCs is to act as an anchor for the cytoskeleton. Cytoskeletal interactions with AE1 give the erythrocyte the flexibility needed to pass through the capillaries. AE1 exists as dimers and tetramers in the RBC, and in the tetrameric form the cytosolic N-terminal domain of AE1 interacts with ankyrin.¹⁹,²⁰ Through ankyrin, AE1 is linked to the cytoskeletal proteins α and β-spectrin.¹⁵ Other interactions with glycolytic enzymes, hemoglobin and protein 4.1 and 4.2 are mediated by the N-terminal domain of AE1, and these interactions are dependent on the phosphorylation state of the N-terminal domain.¹⁵,²¹
Table 1  Cl⁻/HCO₃⁻ exchangers and their properties

| Transport protein | Alternative names | Tissue distribution/Epithelial location | Mechanism/Electrogenicity | Citations |
|-------------------|-------------------|----------------------------------------|---------------------------|-----------|
| AE1               | SLC4A1, Band 3, kAE1, eAE1 | RBC, kidney, heart/kAE1 Basolateral | Cl⁻/HCO₃⁻ exchange/electroneutral | 4         |
| AE2               | SLC4A2            | Widespread/Basolateral                 | Cl⁻/HCO₃⁻ exchange/electroneutral | 5         |
| AE3               | SLC4A3 AE3c, AE3r | Brain, heart, retina, pituitary, adrenal gland; Non-epithelial | Cl⁻/HCO₃⁻ exchange/electroneutral | 6         |
| NDCBE             | SLC4A8, NDAE1, kNBC-3 | Prefrontal cortex of brain, testis, cardiac myocytes, oocytes | Na⁺-dependent Cl⁻/HCO₃⁻ exchange and (electroneutral NBC: splice variant)/ electroneutral | 60, 123, 124 |
| NCBE              | SLC4A10 NBCn2     | Cardiac myocytes, neurons, kidney, uterus, adrenal cortex, choroid plexus/Basolateral | Na⁺/HCO₃⁻ co-transport or Na⁺-dependent Cl⁻/HCO₃⁻ exchange/electroneutral | 61, 62, 125 |
| SLC26A3           | DRA, CLD         | Colon, ileum, cardiac myocytes ecrine sweat gland/Apical | Cl⁻/HCO₃⁻ exchange/electroneutral | 83, 126, 127 |
| Pendrin           | SLC26A4, PDS      | Thyroid, inner ear, thyroid, kidney, prostate/Apical | Cl⁻/HCO₃⁻ exchange; also I⁻/electroneutral | 97, 128, 129 |
| SLC26A6           | PAT-1, CFEX      | Kidney, heart, bronchial epithelium, pancreas, prostate, thymus, intestine/Apical | Cl⁻/HCO₃⁻ exchange; also oxalate and formate/electroneutral | 98, 107, 130 |
| SLC26A7           | SLC26A7          | Thyroid, kidney, stomach, retina, olfactory epithelium/Basolateral | Cl⁻/HCO₃⁻ exchange/electrogenic | 3, 113, 116 |
| SLC26A9           | SLC26A9          | Salivary gland, heart, brain, stomach, trachea, kidney, lung bronchiolar and alveolar epithelia/Apical | Cl⁻/HCO₃⁻ exchange/electrogenic | 120–122, 131 |

Expression data compiled from the literature and DNA micro-array data of transcript abundance in human and mouse tissues (http://symatlas.gnf.org/SymAtlas/).

Figure 2. Topology model of human erythrocyte AE1 (eAE1). Numbers indicate residue number in eAE1, and boxed numbers indicate transmembrane segments. Residue E681 involved in the permeability barrier and anion translocation pathway is highlighted by a *, and the DADD carbonic anhydrase II binding motif is highlighted in gray. The extended structure is shown in the 806–835 region is not meant to imply folding of the protein in the model. Since there is a large amount of sequence conservation between the SLC4A members, the model likely reflects the topology of the other SLC4A members. SLC26A proteins, however, do not have enough sequence conservation with the SLC4A family to be sure of their topology, although hydropathy analysis suggests 12–14 TMs.
Hereditary spherocytosis (HS), which is the most common inherited RBC membrane disorder, occurs when there is a mutation in AE1 that creates defective interactions with the cytoskeleton and plasma membrane of the RBC. Erythrocytes from HS patients are spheroid with a reduced cell surface and are osmotically fragile. The common features of HS are hyperhaemolysis and anaemia, icterus (yellow coloration of skin and mucus membranes) and splenomegaly. Another RBC membrane disorder is hereditary stomatocytosis, which results from point mutations within the transmembrane segments of AE1 that confer cation channel activity. Stomatocytosis is defined as the expansion of the inner membrane leading to membrane invagination and cup-shaped erythrocytes. Affected RBCs have an increased temperature-dependent Na⁺ and K⁺ leakage, with no reduction of deformability.

In the kidneys, AE1 functions to maintain acid-base homeostasis, as well as fluid and electrolyte balance. kAE1 functions in the kidneys to acidify the urine by reabsorption of bicarbonate. Most bicarbonate reabsorption in the kidney is, however, accomplished by the sodium bicarbonate co-transporter, NBCe1, in the proximal tubule. Cl⁻/HCO₃⁻ exchangers in the α and β-intercalated cells of the distal tubule and collecting duct are involved in the fine-tuning of bicarbonate reabsorption. The β-intercalated cells of the distal tubule secrete HCO₃⁻ under alkalotic conditions. The apical Cl⁻/HCO₃⁻ exchanger of the α-intercalated cells is SLC26A4, also called Pendrin, which will be discussed later in this review. On the basolateral side of the α-intercalated cells kAE1 operates to absorb bicarbonate. Mutations of AE1, which impair Cl⁻/HCO₃⁻ exchange function, in these cells can lead to distal renal tubular acidosis (dRTA). dRTA can be either dominant or recessive, and is characterized by impaired urinary acid secretion, metabolic acidosis, growth retardation, hypercalciuria and hypokalemia.

AE1 is the most extensively studied Cl⁻/HCO₃⁻ exchanger in terms of structure, due to its abundance and ease of purification from RBCs. As of yet, there are no high resolution structures of AE1, but extensive studies have determined residues involved in the mechanism of Cl⁻/HCO₃⁻ exchange. Several amino acids have been identified as involved in the permeability barrier and anion translocation pathway. Residue E681 located in the eighth transmembrane segment (Fig. 2) is a key residue involved in both of these structural features. The crystal structure of the N-terminal cytoplasmic domain has been solved to a resolution of 2.6 Å, and has been found to form a dimer. So far, only a low resolution (20 Å resolution) structure for the AE1 membrane domain has been solved using cryo-electron microscopy. In this structure AE1 is present as a dimer with dimensions of 80 Å x 120 Å. Progress has been made to obtain a high-resolution structure of AE1. Studies of native AE1 from RBCs have been unsuccessful due to the heterogeneity of AE1 from this source. As a result, overexpression of AE1 in Saccharomyces cerevisiae yeast cells has been developed as a homogeneous source for purification.

AE2 (SLC4A2). AE2 does not have a restricted pattern of expression like the other anion exchangers, and thus it has been termed the “house-keeping” anion exchanger. There are different splice variants of AE2, which differ in their N-terminal regions. AE2a is widely expressed in a variety of tissues, while AE2b1 and AE2b2 are localized to epithelial tissues and AE2c is found mainly in the stomach. Due to the wide tissue distribution of AE2, it is responsible for the regulation of cytosolic pH through an acidifying pathway, by secreting HCO₃⁻ in exchange for extracellular Cl⁻. AE2 transport activity is regulated by pH. The transport activity of AE2 is activated by an increase in either extracellular or intracellular pH, by hypertonicity, and by NH₄⁺ through the cytoplasmic “modifier” domain.

This acidifying pathway becomes very important in acid secreting cells, for example the gastric parietal cells of the stomach, which would rapidly alkalinize if basolateral AE2 did export bicarbonate into the blood stream. AE2 also fulfills this role in osteoclasts; when osteoclasts digest old bone in order to synthesize new bone the apical surface expresses a vacuolar H⁺-ATPase to acidify the periosseal lacunar space and digest the bone matrix. In order to stop cell alkalization, AE2 is expressed on the basolateral side to secrete HCO₃⁻ into the bloodstream and maintain pH in the osteoclasts. In this same way AE2 has been implicated in tooth enamel formation. Enamel is synthesized by ameloblasts in two stages: the secretory amelogenesis and the maturation-stage. The maturation stage involves the formation of hydroxyapatite crystals, which generate protons. In order for the mineralization process to occur this acid build-up must be neutralized. AE2 localizes to the basolateral membrane of ameloblasts during the maturation stage, where it may act in a similar manner to gastric parietal cells or osteoclasts.

AE2 also plays a special role in concert with the sodium proton exchanger (NHE) in the brain’s choroid plexus to facilitate NaCl uptake and regulate cerebrospinal fluid (CSF) production. Transport of bicarbonate out of the cell and chloride into the cell by AE2, along with transport of protons out of the cell and sodium into the cell by NHE results in a sodium chloride load inside the cell without a change in pH. This accumulation of NaCl causes the osmotic movement of water into the cell. Volume regulation by AE2 also occurs in lymphocytes, where it works in concert with NHE.

In the renal tubule, basolateral AE2 has only been found to be involved in basal pH regulation and not in HCO₃⁻ reabsorption. No hereditary human diseases have been associated with AE2, suggesting that loss of AE2 activity may be embryonic lethal. In some mice loss of AE2 expression is embryonically lethal. In addition, AE2 knockout mice have major defects in gastric acid secretion and parietal cell development, as well as being toothless. Male AE2⁻/⁻ mice are infertile, while female littermates are not. In male AE2⁻/⁻ mice there is a 40–60% reduction of testes size and weight, disruption of spermiogenesis and abnormal epididymal epithelia. Still, no mutations of the AE2 gene have been linked to human infertility.

AE3 (SLC4A3). AE3 is considered the anion exchanger of excitable tissues, since it is only expressed in the heart, brain and retina, where there is no evidence for polarized expression of the protein. AE3 is found as two different transcripts: full length AE3 (AE3fl) and cardiac AE3 (AE3c), which are generated by alternative promoter usage. The AE3fl isoform has 270 amino acids in the N-terminal domain that are replaced by a unique sequence of 73 amino acids in AE3c.

AE3 plays an important role in regulating intracellular pH in the myocardium, since it is responsible for approximately 50% of the bicarbonate export in myocardial tissue. It is concentrated in the T-tubule and the sarcoplasmic reticulum of the cardiac myocytes. AE3 has been linked to the development of cardiac hypertrophy, working in concert with NHE1 in the hypertrophic cascade.
When NHE1 loads the cardiomyocyte with Na⁺, it causes an accumulation of intracellular Ca²⁺ due to the reduced driving force of the plasma membrane Na⁺/Ca²⁺ exchanger. While NHE1 is transporting Na⁺ into the cell it is also transporting H⁺ out of the cell, however there is no rise in pH due to the export of protons.⁵⁰⁻⁵² In order for NHE1 to continue transport there must be a sustained balancing acid load, because NHE1 is auto-inhibited at alkaline pH.⁵³ The parallel acidifying pathway that is activated during NHE1 hyperactivity is thought to be Cl⁻/HCO₃⁻ exchange. The involvement of bicarbonate transport in the hypertrophic cascade is supported by studies that have shown that carbonic anhydrase inhibitors are able to prevent and revert hypertrophy in cardiomyocytes.⁵⁴ Involvement of AE3 in cardiac hypertrophy is supported by the observation that it is the only family member that can be activated by protein kinase C (PKC), which is the kinase implicated in hypertrophic pathways.⁵⁵ AE3 is also expressed in the retina, where it may regulate pH and HCO₃⁻ levels since the retina is the most energy-utilizing tissue of the body, with an associated high HCO₃⁻ load. In the retina there is differential expression of AE3c and AE3f in the Müller glial cell or horizontal cell layers, and there is also differential expression at different developmental stages.⁵⁶ AE3 knockout mice had altered electroretinograms (ERG) and structural changes of the retina, which resulted in visual impairments consistent with those found in human hereditary vitreoretinal degeneration disorders.⁵⁷ In the brain, a point mutation in AE3 has been associated with idiopathic generalized epilepsy.⁵⁸ In addition, AE3 knockout mice have had a reduced threshold for chemically induced seizures.⁵⁹

**Sodium Dependent Cl⁻/HCO₃⁻ Exchangers of the SLC4A Family**

Two members of the SLC4A family (NDCBE and NCBE) have been reported to be sodium dependent Cl⁻/HCO₃⁻ exchangers. When human NDCBE cDNA was expressed in *X. laevis* oocytes, the protein exchanged 1Na⁺:2HCO₃⁻:1Cl⁻, acting as an electroneutral sodium dependent Cl⁻/HCO₃⁻ exchanger.⁶⁰ The same transport mechanism was observed in *X. laevis* oocytes expressing human NCBE.⁶¹ NCBE also mediates Cl⁻/Cl⁻ exchange, demonstrated by extracellular Cl⁻ stimulated Na⁺ uptake and Cl⁻ efflux. A recent report, however supports the role of NCBE as a Cl⁻/HCO₃⁻ co-transporter rather than a Cl⁻/HCO₃⁻ exchanger.⁶² Thus, NCBE has been proposed to be renamed as NBCn2, joining NBCn1 as an electroneutral NBC protein. NDCBE mRNA is found mainly in brain and testis, but low levels have also been found in the kidney and ovary.⁶⁰ Since NDCBE localizes to brain tissues, it may be involved in regulation of intracellular pH in neurons. NCBE localizes to the basolateral surface of the choroid plexus, where it is involved in regulation of the cerebrospinal fluid Na⁺, HCO₃⁻ and fluid levels.⁶³,⁶⁴ In NCBE null mice there are reduced levels of CSF production, which suggests that NCBE plays a significant role in the choroid plexus.⁶⁵ In addition, mice with a disruption in their NCBE gene are more sensitive to chemically triggered seizures.⁶⁶

**Electrogenic Cl⁻/HCO₃⁻ Exchangers of the SLC26A Family**

Although there are ten members in the SLC26A family, only SLC26A3, 4, 6, 7 and 9 are established as bicarbonate transporters.⁶⁶ Originally the SLC26A family was identified to be a group of sulfate transporters.⁶⁷,⁶⁸ Most of the structural information available today is based on information gathered from sequence alignments. The SLC26A transporters are predicted to have 10–14 transmembrane segments based on hydropathy plots, with cytoplasmic N and C termini.⁶⁹⁻⁷² In the mammalian SLC26A family members there is a 22-residue “sulfate transport motif”, near the N-terminal end of the transmembrane domain, which is hypothesized to be required for anion transport. All of the SLC26A members, except for SLC26A4 contain a class 1 PDZ motif at the extreme C-terminus of the protein.⁷³⁻⁷⁴ This motif is not required for transport function, but may be involved in the protein-protein interactions of the SLC26A proteins.⁷⁵ The C-terminus of these proteins also contains a domain initially identified in Sulfate Transporters and Anti-Sigma antagonists (STAS), which has an unclear role in the function of SLC26A proteins, but is the site of a number of disease causing mutations.⁷⁶ The STAS domain is involved in protein-protein interactions, and has been found to interact with the regulatory domain of cystic fibrosis transmembrane conductance regulator (CFTR) and form the binding site for carbonic anhydrase II.⁷⁵⁻⁷⁷

SLC26A3 (DRA, CLD). In a screen for genes downregulated in adenoma, SLC26A3 was cloned from a colon subtraction library,²⁸ hence its original name: DRA. This protein is also mutated in an autosomal recessive disorder, congenital chloride diarrhea, giving it another alternative name CLD.⁷⁹,⁸⁰ SLC26A3 is expressed primarily in the duodenum and colon, with lower levels found in the ileum and cecum of the gastrointestinal tract.⁸¹⁻⁸⁵ The protein localizes to the brush border of columnar epithelial cells and in the colon SLC26A3 is found at the apical membrane of surface and crypt cells.⁸⁶ SLC26A3 is also found in seminal vesicles, sweat glands and pancreas.⁸⁶

When SLC26A3 is expressed in oocytes it can support Cl⁻/I⁻ and Cl⁻/HCO₃⁻ exchange.²⁸,⁷⁰ The protein also transports low levels of sulfate and oxalate, even though its sulfate transport capabilities are still controversial.⁸⁵,⁸⁷ SLC26A3 may also be capable of transporting bromide, nitrate and acetate.⁸⁸ Cl⁻/HCO₃⁻ exchange by SLC26A3 is sodium independent.⁸⁵ There have been reports that SLC26A3 facilitates 1 HCO₃⁻:≤2 Cl⁻ exchange, and other reports that it is an electroneutral exchanger.⁸⁹⁻⁹¹ Careful physiological studies with knockout mice suggests that SLC26A3 is indeed an electroneutral Cl⁻/HCO₃⁻ exchanger.⁷⁹,⁹² The controversy surrounding the electronegativity of SLC26A3 may be due to species or cell specific differences.

In the apical membrane of the intestine, SLC26A3 serves the dual function of bicarbonate secretion and chloride reabsorption. A high concentration of bicarbonate (125 mM) is secreted into the lumen of the intestine by the pancreas in order to neutralize stomach acid. In order for water reabsorption in the intestine to occur, Cl⁻/HCO₃⁻ exchangers must work together with the apical sodium protein exchanger, NHE3. By working together SLC26A3 and NHE3 reabsorb NaCl and water osmotically follows.⁹³

**Pendrin (SLC26A4, PDS).** Pendrin, the gene defective in Pendred syndrome, was identified by positional cloning.⁹⁴ The Pendrin gene product is expressed on the apical membranes of the kidney collecting ducts and thyroid follicular cells.⁹⁵⁻⁹⁷ Pendrin is also found in regions of the inner ear where endolymphatic fluid reabsorption occurs.⁹⁴ Pendrin exchanges Cl⁻ for not only HCO₃⁻ but also sulfate and formate. The protein is also able to support Cl⁻/T and Cl⁻/OH⁻ exchange.⁹⁸
In the kidney, Pendrin localizes to the apical surface of type B (base secreting) and non-A, non-B type intercalated cells of distal convoluted tubule, connecting tubule and cortical collecting duct. Perendrin transports $\mathrm{HCO}_3^-$ into the renal tubule under alkalotic conditions. In the thyroid, Pendrin $\mathrm{Cl}^-/\mathrm{I}^-$ exchange activity is required for $\mathrm{I}^-$ efflux at the apical surface of thyrocytes; abnormalities of Pendrin thus induce goiter. In Pendrin-knockout mice there are no obvious kidney or thyroid dysfunction. When wild-type mice were given an aldosterone analog, Pendrin expression was upregulated, the mice gained more weight and developed more hypertension compared to Pendrin null mice. In Pendrin null mice treated with the aldosterone analog there was more severe metabolic alkalosis, more acidic urinary pH, decreased pCO$_2$, and decreased numbers of non-A type intercalated cells without changing the numbers of type-A intercalated cells. This data suggests a role of Pendrin as the apical $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchanger of base secreting (Type B) intercalated cells of the distal renal tubule. Pendrin is also implicated in renal fluid balance, since Pendrin null mice on a NaCl restricted diet are hypotensive.

Hearing loss marks Pendred syndrome, from which the protein’s name Pendrin comes from. The bicarbonate transporter function of Pendrin may be critical in regulating the composition of the inner ear’s endolymph. In Pendrin knockout mice there are alterations in the inner ear fluid reabsorption, which results in excessive endolymph volume during a critical stage of inner ear development. In addition, when the endolymph composition was analyzed it revealed that the Pendrin knockout mice had acidic endolymph fluid. The acidity of the endolymph fluid impairs the TRPV5/6 Ca$^{2+}$ channels, which results in an elevation of endolymph Ca$^{2+}$ due to failed Ca$^{2+}$ resorption. The elevation of endolymph Ca$^{2+}$ levels has been proposed to impair sensory transduction, which is required for hearing and to promote degeneration of sensory hair cells.

SLC26A6 (PAT-1, CFEX). The full-length human cDNA of SLC26A6 has been cloned. There are two major transcripts of SLC26A6 created by alternative splicing, which encode proteins differing by the addition or deletion of the first 23 N-terminal amino acids. Three other splice variants are described in humans. SLC26A6 $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchange is either electronegic with a stoichiometry of 2 $\mathrm{HCO}_3^-$:1 $\mathrm{Cl}^-$ or electroneutral. Recent data collected from slc26a6 knockout mice has suggested that indeed slc26a6 is an electroneutral transporter. The protein also transports $\mathrm{Cl}^-$, $\mathrm{SO}_4^{2-}$, $\mathrm{HCO}_3^-$, $\mathrm{OH}^-$ and oxalate.

SLC26A6 has a wide renal distribution, with expression in the distal parts of the proximal tubule, the ascending loop of Henle, macula densa cells and intercalated cells in the collecting duct, SLC26A6, at the tubular surface of the distal portions of the proximal tubule, mediates $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchange. SLC26A6 knockout mice display defective oxalate handling in the kidney, which highlights the ability of slc26a6 to transport oxalate.

SLC26A6 is the predominant $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchanger in the upper villus membrane of duodenum and in the heart. Similarly to AE3, SLC26A6 in the heart is concentrated at the T-tubule and in the intracellular membrane of the sarcoplasmic reticulum. SLC26A6 also localizes apically at the pancreatic duct, where it works in concert with SLC26A3 to secrete bicarbonate into the pancreatic duct. SLC26A6 has also been implicated in cystic fibrosis, since its STAS domain has been found to functionally and physically interact with CFTR in the pancreatic duct.

SLC26A7. SLC26A7 was cloned from human high endothelial venules and kidney. Two SLC26A7 protein isoforms result from alternative splicing, which differ in the 11 C-terminal amino acids. SLC26A7 has been independently identified as a $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchanger by several groups. In addition, SLC26A7 $\mathrm{Cl}^-$-coupled anion exchange activity of oxalate and sulfate has also been observed. When expressed in $X$. laevis oocytes, SLC26A7 manifested as a pH-regulated $\mathrm{Cl}^-$ channel. Due to the conflicts between the different transport activities observed for SLC26A7, there is still controversy on the exact transport mechanism of the protein.

SLC26A7 is localized at the basolateral surface of $\alpha$-intercalated cells, where kAE1 is also localized. This suggests that SLC26A7 may provide $\mathrm{HCO}_3^-$ reabsorption capacity in the event that kAE1 is disrupted. SLC26A7 also localizes at the basolateral surface of parietal cells, where it has been proposed to act in a $\mathrm{Cl}^-$-loading mechanism and possibly explain unidentified $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchange activity not attributed to AE2.

SLC26A9. SLC26A9 is expressed in the alveolar, bronchial and tracheal epithelium of the lung, where it may play a role in maintaining airway surface liquid composition. In addition, it is also expressed in gastric pits and on the apical membrane of gastric epithelial cells in the stomach. SLC26A9 has been proposed to protect the gastric mucosa by secretion of $\mathrm{HCO}_3^-$ onto the surface epithelium. Lower levels of expression have been detected in the pancreas and prostate.

When expressed in HEK293 cells, SLC26A9 mediates $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchange, as well as $\mathrm{Cl}^-$ independent $\mathrm{HCO}_3^-$ transport by an unknown mechanism. In $X$. laevis oocytes expressing SLC26A9 chloride, sulfate and oxalate transport have been detected. There are few studies on SLC26A9 and controversy remains whether the protein is a $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchanger, or a $\mathrm{Cl}^-$ channel, possibly modulated by $\mathrm{HCO}_3^-$.

Conclusion

$\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchangers facilitate rapid transport of bicarbonate across the plasma membrane and are distributed throughout the entire body. Differences between the $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchangers allow a variety of physiological roles in different tissues to be fulfilled and regulated independently. Due to the importance of these proteins hereditary diseases have been associated with them. Many of the mutations causing disease are either due to impaired transport function or impaired cellular targeting of the transporter.

Many studies have been done with knockout mice of $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchanger genes, and the continued direction of this research will provide more knowledge on the function of these proteins in vivo. While this information is extremely valuable, in order to understand the diseases related to $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchangers and possibly discover new treatments, more research must be done to understand the mechanism of these proteins. While considerable research has been done to elucidate a crystal structure, there is still no high-resolution structure available for any of the $\mathrm{Cl}^-/\mathrm{HCO}_3^-$ exchangers. In addition, there is still debate over the substrate specificity of several of the transporters, especially in the SLC26A family. In the future, answers to these questions will provide valuable insights into diseases caused by mutations in the SLC4A and SLC26A genes.
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