CLS Meets the Aquaporin Family: Clinical Cases Involving Aquaporin Systems

WAYNE GADE, BROOKE ROBINSON

Virtually all human cells incorporate aquaporins, or water channel proteins, into their cell membrane. Indeed, many cells produce several aquaporins, each adapted for a specific physiologic function. Thus, it is not surprising that aquaporin malfunctions are associated with numerous important clinical conditions. This article describes the clinical aspects of malfunctions in aquaporins or their regulation.

Although water can diffuse across biological membranes (osmosis) without the aid of a transport system, researchers had predicted for decades that rapid reabsorption by renal tubule cells must be aided by a channel or pore. Yet, not until the 1990s were the first members of the aquaporin (AQP) family identified. Led by Dr. Peter Agre, recipient of the 2003 Nobel Prize in Chemistry, researchers have since amassed an astounding amount of information about AQPs and their function. For example, the flow rate of water through AQP1 is an extraordinary three billion water molecules per second per aquaporin channel, while a relative trickle of water crosses the hydrophobic lipid bilayer of cell membranes devoid of AQPs.

Our understanding of renal physiology and pathophysiology has advanced greatly as we account for the subtle implications of various AQP systems. For example, nephrogenic diabetes insipidus (NDI), the inability to produce concentrated urine, can result from several different malfunctions in the hormonally controlled AQP2 system. The list of diseases known to involve AQPs now includes: early onset of cataracts, Sjogren's syndrome, cerebral and pulmonary edemas, cirrhotic liver development of ascites, and congestive heart failure (CHF).

ABBREVIATIONS: ADH = anti-diuretic hormone; AQP = aquaporin; cAMP= cyclic adenosine monophosphate; CDI = central diabetes insipidus; CHF = congestive heart failure; MIP = major intrinsic protein; NDI = nephrogenic diabetes insipidus; PCT = proximal convoluted tubule; RT-PCR = reverse transcription - polymerase chain reaction; SS = Sjogren's syndrome; SIADH = syndrome of inappropriate ADH secretion.

INDEX TERMS: aquaporins; diabetes insipidus; nephrogenic diabetes insipidus; renal physiology.

Clin Lab Sci 2006;19(2):80

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The first aquaporin, originally called CHIP28, was accidentally discovered during an investigation of Rh antigens, and the authors suggested it was part of the red cell cytoskeleton.[1,2] Elegant demonstration that the CHIP28 protein was indeed a water channel occurred when its insertion into frog oocyte membranes conferred a rapid response to hypotonic (low solute) conditions. Normally, these frog eggs respond very sluggishly in hypotonic buffer, but oocytes expressing CHIP28 rapidly swelled and exploded when placed in a hypoosmolar buffer.[3]

Since this 1991 demonstration, aquaporin research has similarly "exploded" with reports of 11 different AQPs, several thousand AQP articles in the scientific literature, and the 2003 Nobel Prize for Chemistry for their discoverer, Dr. Peter Agre.[1] Hundreds of AQP researchers have purified, characterized, and successfully cloned aquaporins designated AQP0-AQP10.[4-9] Detailed molecular mechanisms of several

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AQPs are known, enabling researchers to relate structural details to their function.[6,8,9] Subtle differences in AQP activities and different tissue localizations have suggested specialized functional roles.[5-9] A more complete description of aquaporin research and methodologies is included in the accompanying article.

A thorough understanding of renal physiology now explains the very rapid reabsorption of water in the proximal convoluted tubule (PCT) by acknowledging the presence of plethora of AQP1 channels in the membranes.[10-13] The relative water impermeability of cells of the ascending limb of the loop of Henle, which was previously mysterious, is now simply explained by their lack of AQPs. Hormonal control of water reabsorption by cells in the collecting duct is known to depend on insertion of an intracellular pool of AQP2 channels following the binding of anti-diuretic hormone (ADH) to the cell surface receptor.[10-13] A more complete description of this system is given in the accompanying article.

Basic science research directed at cell biology inevitably answers many clinical questions, as well as uncovering many more unanswered questions. The list of associations between diseases or pathophysiologic conditions and AQP system malfunctions is constantly expanding.[4-9] Within the next few years, an understanding of AQPs will become an essential part of laboratory medicine. This article will survey diseases associated with AQP defects and provide several case stories to illustrate the pathophysiology.

Table 1. Clinical applications of aquaporins

Disease or condition	AQP involved	Malfunctions and symptoms
Renal diseases - NDI	AQP2 or receptors	Inability to concentrate urine, polyuria, lack of ADH (vasopressin) response
Subclinical NDI	AQP1 deficiency	Subclinical NDI, symptomatic after osmotic stress
Sjogren's syndrome	AQP5, AQP1	Chronically dry mouth, eyes, and lungs. Deficiency or inappropriate signaling of AQP5 suspected.
Eye diseases (cataracts)	AQP0 (MIP)	Inappropriate removal of water from lens
CHF and liver cirrhosis	AQP2 elevation	Changes in aquaporin patterns
Pulmonary edema	AQP2 elevations	Changes in aquaporin patterns
Cerebral edema	AQP4	Malfunction of osmoregulation involving AQP4 suspected. Changes in aquaporin patterns, astrocytes
Colton blood groups	AQP1	Rare blood group related to mutation, produces no functional AQP1*, HDN-complicated pregnancies

*Surprisingly, patients without functional AQP1 have only subclinical renal symptoms in absence of stress. Under stressed conditions, these patients can exhibit a severe inability to concentrate their urine appropriately.[25]

CLINICAL METHODOLOGIES OF AQUAPORIN RESEARCH

Although AQP assays have not yet entered the routine clinical arena, their arrival is imminent as research assays are commercialized. A brief review of the wide array of research methods is included in the accompanying article.

Traditional protein biochemistry techniques have been combined with techniques of molecular biology, such as reverse transcription - polymerase chain reaction (RT-PCR) to convert cellular mRNAs back into DNA for cloning the AQP genes.[14-16] In addition, quantitation of mRNAs leads to understanding of cellular expression and factors that mediate up- or down-regulation of various AQPs.[17-20] The frog oocyte system described above remains a valuable tool to demonstrate water channel activities of normal and mutant genes which are inserted into transgenic mice and laboratory animal models.[21-24] Others have identified and studied AQP-related mutations from actual patients.[22-26] Tsukaguchi and others have evaluated the function of mutations in AQP and ADH receptors by inserting them into oocytes.[24] These and other studies have demonstrated adverse effects in receptor binding and protein processing or trafficking, in addition to actual AQP dysfunctions.[21-24]

Immunostaining, using antibodies to specific AQPs, has been instrumental in establishing tissue localization and in some cases intracellular localization.[26,27]

Several research assays for urinary AQP2 are in the enzyme immunoassay format and could be added to the arsenal of clinical assays, once the clinical utility of detecting elevated levels of AQP2 has been established. Elevations in urinary AQP2 are found in CHF patients and those with liver cirrhosis, and AQP2 expression is known to be affected by numerous physiologic and pharmacologic factors.[11,47]

Finally, as DNA probes become more commonplace in the clinical laboratory, the probes used to identify AQP mutations and assess levels of gene expression may present exciting diagnostic possibilities.

CLINICAL RELEVANCE OF AQUAPORINS

A summary of a range of aquaporin-related diseases can be found in Table 1. While renal diseases are the best characterized conditions resulting from compromised AQP systems, the following discussions will illustrate the mounting evidence for AQP involvement in many diverse pathologies.

Nephrogenic diabetes insipidus

Diabetes insipidus is defined by polyuria, even under conditions of dehydration, when ADH secretion normally maximizes water reabsorption. The inability to concentrate urine by reabsorbing water causes patients to produce 10-20L of very dilute urine per day. Inadequate secretion of ADH defines central diabetes insipidus (CDI), while the nephrogenic condition (NDI) results from an insufficient renal response to ADH. Recently, understanding of AQPs and the systems that control them has added greatly to our understanding of renal physiology and diseases.[10-13]

The complex physiology of NDI starts with the ADH binding to its receptor and the response by CD cells, which requires several distinct steps.[10-13] First, the cell-surface ADH receptors must specifically bind the ADH molecule. Binding to its receptors activates adenylate cyclase, an enzyme that catalyzes the synthesis of cyclic adenosine monophosphate (cAMP) and initiates the "second

Case 1. Congenital NDI*

A six month old male from a German family was diagnosed with congenital NDI following presentation with severe dehydration, hypernatremia, unexplained fever, vomiting, and failure to thrive. The child produced 1.1 L/day, 2-3x normal range. Other urine and serum chemistry levels are shown below, (normal ranges):

	Serum	Urine
Osmolality (mOsmo/kg)	316 (280-300)	104 (300-1000)
Sodium (mmol/L)	164 (135-145)	ND
Serum ADH (pg/ml)	35 (< 2)	ND
Osmo (+Desmopressin)	no change	98 mOsmo/kg

Follow-up testing defined the cause of the NDI to be a mutation in the ADH receptor. The non-functional ADH receptor caused the patient's renal function to be unresponsive to the very elevated levels of ADH shown above.

*Data extracted from a case originally described in Pasel.[22]

messenger" system. Elevated cAMP levels lead to the phosphorylation of the pre-synthesized AQP2 channels, and signals for their insertion into the cell membrane.[10-13] Elevated cAMP also increases AQP2 gene expression, promoting the synthesis of new AQP2 molecules. Once AQP2 molecules are inserted into the cell membrane, they will facilitate water reabsorption.

While malfunctions have been identified in each of these steps, most NDI cases have involved ADH-receptor mutations or disruption of the intracellular transport, rather than mutations of AQP2.[22-24, 32-37] Case 1 describes a congenital NDI caused by an ADH receptor mutation.

Acquired forms of NDI, including lithium treatment

Acquired NDI can be caused by numerous nephrotoxic chemicals or drug treatments. For example, approximately 20% to 30% of patients treated with lithium become polyuric.[22] Lithium's therapeutic mode of action is unclear, but it has been speculated that lithium interferes with second messenger signals, such as cAMP. If this is true, lithium-induced NDI could result from disruption of the signal that normally results from ADH binding to its receptor. Without elevated cAMP, AQP2 is not inserted into the membrane, and water reabsorption is minimized.[10-13]

Marples found that a 25 day course of lithium caused a 95% reduction in AQP2 in rats and the lithium-treated rats developed extreme polyuria,[38] excreting urine volumes equal to their body weight each day. During lithium treatment, administration of ADH could not reverse the down-regulation of AQP2. However, AQP2 levels slowly returned to

Case 2. Subclinical NDI caused by AQP1 deficiency*

A 37 year old woman was found to have antibodies against the Colton blood group during a routine prenatal screen. Follow-up testing showed the patient to be homozygous for a deletion in exon one of the AQP1 gene, making her incapable of producing any functional AQP1.

She had previously had a miscarriage during her first pregnancy, but gave birth to a healthy baby after 34 weeks of her second pregnancy. However, this infant suffered hemolytic disease of the newborn and required three neonatal transfusions. A subsequent pregnancy required five intrauterine and two neonatal transfusions. Fortunately, both children survived.

This woman had no other major medical problems, but occasionally experienced peripheral edema. She typically drank three to four liters of fluid per day and a subclinical polyuria. However, subsequent studies revealed that she lacked normal urine concentration capability that became obvious during periods of fluid deprivation. For example, following a 21 hour fluid deprivation, her serum and urine chemistry values were:

	Pre-restriction		Post-restriction	
	serum	urine	serum	urine
Osmolality (mOsmo/kg)	280	230	287	431
ADH (pg/mL)	1.5	NA	4.7	NA

The normal response to a 15 hour to 24 hour fluid deprivation would be a more concentrated urine (800-1300 mOsmo/kg), compared to this patient's urine of 431 mOsmo/kg. In addition, urine osmolality remained at 400 mOsmo following infusions of hypertonic saline or desmopressin, an ADH agonist.

Following water loading, the patient produced appropriately dilute urine of ~ 80 mOsmo/kg. Immunoassays demonstrated appropriate levels of AQP2, but no AQP1 present. Thus, this patient's chronic subclinical polyuria appears to be related to a less hypertonic medulla, which limited her ability to concentrate urine even during fluid restriction.

*Case was summarized from King.[25]

normal following withdrawal of lithium.[38] Interestingly, fluid deprivation increased AQP2 synthesis without increasing its insertion into the membrane,[38] implying that a second regulatory mechanism exists for AQP2, in addition to ADH system.[11,39]

Other forms of polyuria also appear to be associated with decreased AQP2 expression. Both hypokalemia (low serum potassium) and hypercalcemia (elevated serum calcium) are electrolyte imbalances known to be associated with ADH-resistant polyuria and polydipsia causing down-regulation of AQP2.[5] The antibiotic Amphotericin B has also been shown to decrease AQP2.[40]

AQP1 deficiency and subclinical NDI

As mentioned previously, AQP1 is present in many cells, including red blood cells, and is responsible for the majority of water reabsorbed in the PCT. Since approximately 75% of the filtrate is reabsorbed in the PCT, it is surprising that patients with a complete AQP1 deficiency have only a subclinical form of NDI.[25] Since most of renal water reabsorption is facilitated by AQP1 in the PCT, one might expect a total deficiency of this aquaporin would lead to a devastating NDI and an anemia due to the lack of AQP1 in red blood cells. However, several known cases of AQP1 deficiency have presented with surprisingly mild renal concentration problems and no overt anemia.[25] Cells in the straight portion of the proximal tubule contain substantial amounts of AQP7, which presumably enables these patients to remain subclinical.[4,12] Case 2 was discovered primarily because the altered AQP1 gene resulted in a rare Colton blood group antigen.

Aquaglyceroporins AQP3, AQP7, and AQP9

Aquaglyceroporins have considerable sequence homology with the AQP family, but are uniquely able to transport both glycerol and water.[17,18,41-43] This strange combined specificity primarily facilitates glycerol's uptake by liver cells as a substrate for gluconeogenesis, and its exit from fat cells following lipolysis.[17,18,41-44] Studies of patients without functional AQP3 have shown decreased transport of glycerol in RBCs.[1] In addition, glycerol-transport functions place AQP7 and AQP9 in important metabolic positions; understanding their function and control may significantly increase our understanding of metabolic syndrome. For example, insulin was shown to down-regulate both AQP7 and AQP9 in a report by Kuriyama and coworkers.[18] Thus, insulin would limit gluconeogenesis by decreasing both the AQP7-mediated release of glycerol from fat cells and its AQP9-mediated uptake in the liver. Added to insulin's well established role in enzyme regulation, insulin also limits gluconeogenesis through down-regulation of AQPs. It is, therefore, not surprising that both AQP7 and AQP9 were increased in patients with insulin-resistant conditions, compounding their tendency to develop hyperglycemia.[18] One patient, who produced a non-functional AQP7, was shown to have a decreased release of glycerol from fat cells during exercise.[17]

Strangely, AQP9 is also capable of transporting $As(OH)_3$. This broadened specificity may be unfortunate, because it renders the liver more sensitive to arsenic poisoning.[9,47] In India, where the World Health Organization estimates more than 100 million people are consuming toxic levels of arsenic in their drinking water, an epidemic of liver cancer may be emerging.[9]

Overexpression of AQP2 in cirrhosis and CHF

Liver cirrhosis frequently leads to ascites in the peritoneal cavity, and CHF similarly involves excessive water retention. Schrier proposes the unifying theory that such excessive fluid retention starts with peripheral arterial vasodilation. At plasma osmolalities that would normally suppress ADH secretion, vasodilation temporarily decreases blood pressure, and in turn, increases ADH secretion and an overexpression of AQP2.[5,47] Decreased blood pressure also stimulates release of renin, which initiates the RAAS system and increased sodium retention. Increased plasma osmolality further stimulates ADH secretion and promotes water retention. Thus, a vasodilation leads to retention of both sodium and water, as seen in conditions such as CHF, liver cirrhosis, or pregnancy.[47] Schrier and coworkers found that AQP2-mediated water retention could be reversed by agonists of the ADH receptor (also called V_2) in both animal models of cirrhosis and human patients with these conditions.[5,9,47]

In animal models, AQP2 was induced 150% to 200% by administration of an ADH analog, dehydration, or in a model of cirrhosis produced by carbon tetrachloride.[48] Data from patients with both CHF and cirrhosis were found to show elevated AQP2 levels even when plasma osmolalities would normally have suppressed ADH secretion and decreased AQP2 levels.[5] Thus, it appears that AQP2 overexpression is directly involved in these conditions of volume overload.

Sjogren's syndrome associated with lack of AQP5 function?

Sjogren's syndrome (SS) is an autoimmune disease characterized by dry eyes and dry mouth, and lymphocytic infiltration of the salivary and lacrimal glands.[20,26,27] The majority of SS patients are women in the 30 year to 50 year age group, and the diagnosis is confirmed by demonstration of antibodies

to SS-A and SS-B antigens (see Case 3). Figure 1 shows that AQP3, AQP4, and AQP5 are all expressed in secretory glands, such as salivary and lacrimal glands. Membrane localization of AQP5 in apical membranes of both glands suggests that a deficiency of functional AQP5 might be the cause of the lack of secretions.[26] Controversy exists over the membrane localization and the role of specific AQPs in the Sjogren's disease process.[26,27] Surprisingly, Tsubota and coworkers found increased AQP5, rather than the decreased levels expected in Sjogren's patients (88 g/mg protein compared to 55 g/mg in control patients). [27] However, histochemical techniques demonstrated that the AQP5 in SS patients was cytoplasmic rather than inserted into the membranes, as was seen in controls. Thus, SS patients may synthesize AQP5, but not insert it into the cell membranes.[27]

Other groups have observed differences in AQP5 localization between SS patients and controls, using histological staining and centrifugal fractionation.[28] In addition, Beroukas and coworkers reported a 38% decrease in AQP1 of SS patients, but found no differences in levels of AQP3 and AQP5.[20] "Knockout mice" lacking genes for AQP1, AQP3, AQP4, and AQP5 demonstrate that some combination of AQPs is essential.[30] This controversy is pending, but it appears likely that AQP5 plays a critical role in this regulated secretion.

An intriguing mechanism for Sjogren's pathology could involve a malfunction of the M3-muscarinic receptors that control secretion of tears and saliva. This seems more plausible with the report that AQP5 contains a phosphorylation site homologous with the site that controls AQP2 insertion in the CD cells of the kidney. This phosphorylation site, together with AQP5's reported cytoplasmic localization, suggests a control mechanism for water secretion in these secretory glands analogous to control of renal water retention.[29,30] Thus, signaling malfunctions analogous to the mutations of ADH receptors and AQP2 that cause NDI might be responsible for Sjogren's syndrome. However, much of the

Case 3. Sjogren's syndrome

A 55 year old woman complained of dry eyes and dry mouth over the last six months. The physician suspected possible Sjogren's syndrome and ordered the following autoimmune tests, including ANA, anti-DNA, anti-SS-A, anti-SS-B, anti-Sm, and anti-RNP. CRP and CH-50 tests were added to assess inflammatory activity. Urinalysis, chemistry, and hematology panels were unremarkable except for data shown below:

WBCs (white blood cells)	3,500/mm^3	L	3.9-11.0 x 10^3 / mm^3
% Monocytes	15.9 %	H	5.0%-15.0 %
BUN	20	H	6-19 mg/dL
hsCRP	< 0.3		0.0-0.4 mg/dL
ANA	positive 1:160 (with homogeneous pattern)		
anti-DNA	negative		negative
anti-SS-A	1663 U/mL	H	< 100 U/L
anti-SS-B	543 U/mL	H	< 100 U/L
anti-Sm	28 U/mL		< 100 U/L
anti-RNP	34 U/mL		< 100 U/L
CH-50 complement	61	H	22 U/mL-60 U/mL

The extremely elevated SS-A and SS-B results confirmed the expected diagnosis of Sjogren's syndrome. The normal level of hsCRP and slightly elevated CH-50 complement suggested that she was not experiencing an episode of excessive inflammatory activity. Although this patient was not assessed for AQP mutations or abnormal gene expression, it is likely that Sjogren's cases involve abnormal AQP function of some type.

pathophysiology of SS ultimately relates to the lymphocytic infiltration and destruction of gland function, which may be induced by abnormal proteins of the AQP family.

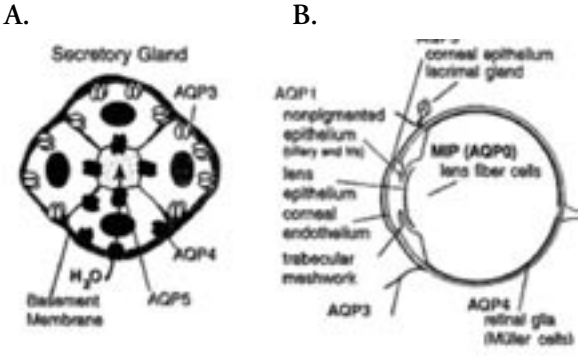
Final resolution of the lacrimal gland controversy is pending, but it appears a general mechanism may occur in other secretory glands and ducts. For example, both AQP1 and AQP5 are observed in pancreatic ducts.[31]

AQUAPORINS AND RESPIRATORY FUNCTION

The respiratory system involves numerous cell types and requires a consistently moist environment for effective function.[49-51] Maintaining moist surfaces can be challenging when conditions range from desert desiccation to humid tropics to subzero arctic temperatures. Researchers are investigating the role that airway humidification plays in conditions such as asthma and the bacterial colonization that leads to pneumonia. Interestingly, corticosteroids have been shown to specifically increase the expression of AQP1.[51] This may help explain part of the therapeutic benefits of corticosteroids in pulmonary infections.

Figure 1. Schematic diagrams illustrate multiple AQPs in secretory glands and eyes

In panel A, AQP3, AQP4, and AQP5 are shown to be involved with the transport of water from the basement membrane (AQP 3 and AQP4) and its secretion into the apex (AQP5). Panel B shows localizations of APQ0, AQP1, AQP3, AQP4, and AQP5 in cells around the eye. As described in the text, AQP0 is associated with cataract development and AQP5 is implicated in the malfunction of lacrimal glands seen in patients with Sjogren's syndrome.



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In the alveolus, AQP1 was identified in the endothelial cells, and AQP5 was identified in the membranes of Type I pneumocytes (as shown in Figure 2). This implies that AQP1 facilitates water movement from the capillary toward the basement membrane, while AQP5 appears to provide humidification to the alveolar surfaces themselves. It has also been suggested that altered expression or function of these AQPs may play a role in such pulmonary conditions as edema and pleural effusions.[1, 49-51]

As described above, AQP5 contains a phosphorylation sequence homologous to the regulatory portion of AQP2. This AQP5 sequence is responsive to hormonal signals and suggests that AQP5 function may also be regulated in the respiratory system. Again, by analogy with the renal-AQP2 scheme, signaling malfunctions and/or direct AQP malfunctions may prove to be involved with pulmonary pathogenesis.[1, 49-51]

Cataracts and AQP0 mutations

Cataracts are defined by abnormal opacities in the lens tissue of the eye. The major membrane protein in this tissue was originally designated MIP (major intrinsic protein) but is known to be an aquaporin (now designated as AQP0).[52] The water content of lenses is lower than most other tissues, and unlike most tissues, the water content increases with age.[53,56] It has been suggested that AQP0 plays a significant role in the maintenance of lens transparency by removing excess water and reducing the amount of light scattering in the lens.[53]

Two families with inherited tendency to develop cataracts were found to have point mutations in AQP0.[54] When the defective genes from both families were inserted into the *Xenopus laevis* oocyte expression system, the results confirmed that the water channel was defective. Shields and others have demonstrated that AQP0-deficient mice developed cataracts and optical dysfunction, and that the severity of the cataracts was dependent on the level of mutant gene expression. In effect, the expression of the mutant AQP0 may have led directly to the development of the proverbial blind mice.[55]

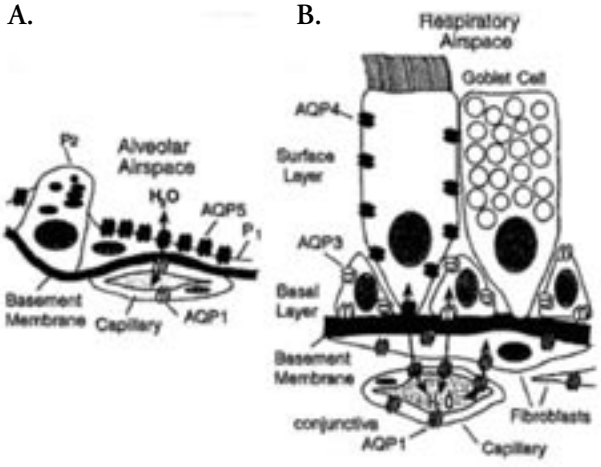
Aquaporins in brain tissues

Brain tissues contain several members of the AQP family, but AQP1 and AQP4 predominate.[1,4,57-60] Research suggests that AQP4 helps reduce excess fluids in the brain when hydrostatic pressure is increased.[57-60] In addition, mice lacking AQP4 had altered tendency to suffer brain edema following water intoxication and ischemic stroke, but no gross abnormality under normal conditions.[21,58]

Yamamoto and coworkers reported decreased expression of AQP4, APQ5, and AQP9 in cultured rat astrocytes stressed by hypoxia.[59] Interestingly, AQP5 was transiently up-regulated upon reoxygenation, while expression of the other AQPs returned to normal.[59] Obviously, a malfunction of the AQP4 or AQP5 systems could lead directly to conditions such as cerebral edema or hydrocephaly.

Saadoun and others proposed a fascinating explanation of cerebral edema in brain confusions, bacterial meningitis, and brain tumors.[60] They studied the relationship of AQP4 in normal astrocytes with a specific K^+ channel. Under normal conditions regulation of these two channels was coupled. However, in the damaged tissues the channels were uncoupled. AQP4 was upregulated in edematous tissue astrocytes, while the K^+ channels were upregulated in the astrocytes of the damaged tissues.[60]

Figure 2. Schematic diagrams illustrating multiple AQP involvements in respiratory cells



In panel A, AQP 1 is shown assisting water movement out of the capillary epithelial cells, while AQP5 is primarily responsible for diffusion onto the surfaces facing the alveolar airspace. In panel B, AQP1 is again shown facilitating transport from the capillary through the basement membrane. AQP3 and AQP4 are involved with providing humidification to the respiratory spaces of the bronchial passages.

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Another interesting observation is the localization of AQP4 in the neurohypophysis, where osmoreceptors control the release of ADH by an undefined process.[57] Thus, AQP4 may enable the rapid changes in cell volume in response to changes in plasma osmolality that are thought to control osmoregulation.[57] When these osmoregulatory cells sense hyperosmolar plasma, they release ADH, which in turn causes AQP2-assisted water reabsorption by kidney collecting duct cells. Thus, the hyperosmolar condition is diluted back to normal, and both blood volume and pressure increase. If this view of osmoregulation proves to be correct, changes in AQP4 function or distribution could potentially result in disorders of ADH release, including CDI or the syndrome of inappropriate ADH secretion (SIADH). Ultimately, AQP4 may help control plasma osmolality and blood pressure, and malfunctions could contribute to "essential" or "idiopathic" hypertension.

CONCLUSION AND SUMMARY

Aquaporins span the cell membranes of virtually all human cells, facilitating the passive movement of water in response to osmotic gradients.[1,4-9] Malfunction of AQPs and their regulatory systems are associated with numerous important conditions. AQP1 was originally isolated from RBCs, but has subsequently been shown to exist in numerous other cells.[1,2,4-9] This channel is primarily responsible for the unregulated water reabsorption of the PCT. Hormonally regulated water reabsorption in the renal CD relies on mobilization of AQP2-mediated water movement.[4-13] In this case, pre-synthesized AQP2 resides in internalized vesicles until ADH is bound to the receptor. Binding of ADH causes the AQP2-containing vesicles to fuse with the cell membrane and increase water reabsorption. A similar on/off mechanism is thought to control AQP5 function in secretory glands, such as salivary and lacrimal glands.[4] Aquaporins 3, 7, and 9 have the unique ability to transport glycerol in addition to water.[1,4,41-44] Their limited localization in fat cells and hepatocytes suggests that their physiological role involves glycerol transport. Responsiveness of these AQPs to insulin levels and physiological conditions such as fasting and exercise suggests that they may help control glycerol transport, and thus, lipolysis and gluconeogenesis. Patients with insulin-resistance, such as Type 2 diabetes mellitus or metabolic syndrome, have been shown to have elevated levels of AQP 7 and AQP9.

AQPs are implicated, either as the primary lesion or secondarily, in numerous diseases. For example, nephrogenic diabetes insipidus can be caused by either deficient or defective AQP2 or a malfunction in response to ADH-mediated control of

AQP2 activity. Overexpression of AQP2 is indirectly involved in edematous conditions such as CHF, cirrhotic ascites formation, and pulmonary edema. AQP0 abnormalities are implicated in early-onset cataracts.[54,55] In the brain and CSF AQP4 predominates, and dysfunction of this system may result in cerebral edema and hydrocephaly. Localization of AQP4 in cells associated with osmoregulation suggests that this aquaporin might be involved with osmoregulation, and malfunctions of the system could result in such common conditions as hypertension.[56-59] Once such malfunctions are defined, they may immediately suggest an appropriate therapeutic strategy.

Although no assays detecting AQPs have yet arrived in the clinical laboratory, they will almost certainly become part of the menus within a few years. Urinary AQP2 is routinely detected by research EIA procedures, which could easily be automated.[28,29] Dysfunctions in AQP2 and its regulation system are associated with NDI and SIADH, while elevated AQP2 levels are associated with conditions such as CHF and liver cirrhosis and quantitation of AQP2 levels may help assess the condition.[46-48] Polymorphisms in AQP1 result in Colton blood group antigens, and although anti-Colton antibodies are rarely identified, they have been associated with serious cases of hemolytic disease of the newborn.[25,35] Variant AQP function could also explain autoimmune diseases such as Sjogren's syndrome.[26,27] Testing for specific autoantibodies to these AQPs may replace less-specific tests such as antinuclear antibodies.

ACKNOWLEDGEMENTS

The authors wish to thank Carol Gee of Decatur Memorial Hospital for her assistance in obtaining the case information for the Sjogren's syndrome case and Jean Gade for her editorial comments and clinical and scientific expertise.

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