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Aquaporins in Clinical Medicine

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Abstract

The aquaporins are a family of membrane water channels, some of which also transport glycerol. They are involved in a wide range of physiological functions (including water/salt homeostasis, exocrine fluid secretion, and epidermal hydration) and human diseases (including glaucoma, cancer, epilepsy, and obesity). At the cellular level, aquaporin-mediated osmotic water transport across cell plasma membranes facilitates transepithelial fluid transport, cell migration, and neuroexcitation; aquaporin-mediated glycerol transport regulates cell proliferation, adipocyte metabolism, and epidermal water retention. Genetic diseases caused by loss-of-function mutations in aquaporins include nephrogenic diabetes insipidus and congenital cataracts. The neuroinflammatory demyelinating disease neuromyelitis optica is marked by pathogenic autoantibodies against astrocyte water channel aquaporin-4. There remain broad opportunities for the development of aquaporin-based diagnostics and therapeutics. Disease-relevant aquaporin polymorphisms are beginning to be explored. There is great promise in the development of small-molecule aquaporin modulators for therapy of some types of refractory edema, brain swelling, neuroinflammation, glaucoma, epilepsy, cancer, pain, and obesity.

Keywords

water transport; neuromyelitis optica; cell migration; cancer; obesity

INTRODUCTION

The aquaporins (AQPs) are a family of “water channels” that are conserved throughout the animal and plant kingdoms, and in lower organisms. There are 13 mammalian aquaporins, which are widely distributed in specific cell types in many organs and tissues. Their primary function is to facilitate water transport across cell plasma membranes; some AQPs (called aquaglyceroporins) also transport glycerol. Much of our understanding of AQP physiology has come from phenotype analysis of AQP knockout mice. Mouse phenotype studies confirmed the suspected involvement of AQPs in the urinary concentrating mechanism and glandular fluid secretion, and led to the discovery of unanticipated roles of AQPs in brain water balance, cell migration, cell proliferation, neural activity, pain, epidermal hydration, and ocular function. This review focuses on the clinical and translational aspects of AQP biology and highlights the possibilities for AQP-based diagnostics and therapeutics.

DISCLOSURE STATEMENT

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PRIMER ON AQUAPORIN STRUCTURE, FUNCTION, AND TISSUE EXPRESSION

The AQPs are relatively simple in their structure and function. Unlike ion channels and solute transporters, the AQPs do not show gating, saturation, or membrane potential-dependence behavior. Many mammalian AQPs, including AQPs 1, 2, 4, 5, and 8, function primarily as water-selective transporters. Cells expressing AQPs on their plasma membrane have a few-fold up to ~50-fold higher osmotic water permeability than membranes that do not contain AQPs. Aquaglyceroporins, the AQPs that transport both water and glycerol, include AQPs 3, 7, and 9. AQP9 may transport other small, polar solutes as well, including amino acids, sugars, and even arsenite (1, 2). There is controversial evidence that some AQPs also transport various other small molecules and gases, including carbon dioxide, ammonia, nitric oxide, and hydrogen peroxide (3–6), although the physiological relevance of AQP-facilitated transport of molecules other than water and glycerol remains unproven. As AQPs are generally expressed constitutively in plasma membranes, regulation of their function is mainly at the transcriptional level. There are many descriptive studies of regulated AQP expression in response to various stresses, such as AQP4 upregulation in brain infection and trauma. However, with the notable exception of AQP2 in the urinary concentrating mechanism, discussed below, the biological significance of transcriptional and post-translational regulation of AQPs remains unclear.

X-ray crystal structures exist for several mammalian AQPs. All AQPs form tetramers in membranes in which monomers, each of ~30-kd molecular size, contain six transmembrane helical domains and two short helical segments surrounding cytoplasmic and extra-cellular vestibules (7, 8). The vestibules are connected by a narrow aqueous pore allowing single-file water transport in which water selectivity is conferred by electrostatic and steric factors (9). The aquaglyceroporins have a less constricted pore than the water-selective AQPs, with a larger proportion of hydrophobic pore-lining residues. AQP0 and AQP4 can form supramolecular crystalline arrays (10, 11).

The mammalian AQPs are expressed in various epithelia and endothelia involved in fluid transport, as well as in other cell types such as epidermis, adipocytes, and skeletal muscle. In kidney, AQPs 1–4 are important in the urinary concentrating mechanism (12). AQPs also facilitate transepithelial fluid secretion in exocrine glands and other secretory epithelia (13). In the central nervous system (CNS), AQP4 in astrocytes is involved in water balance and neuroexcitatory processes (14). In the eye, AQPs in cornea, lens, ciliary epithelium, and retina are involved in ocular surface hydration, corneal and lens transparency, intraocular pressure regulation, and visual signal transduction (15). In skin, AQP3 in epidermal keratinocytes is involved in skin hydration and epidermal proliferation (16). AQPs are also expressed widely in various cell types in lung (17) and the gastrointestinal tract (18), although their expression in these systems appears to have little functional consequence. AQP tissue expression without demonstrable physiological function may represent a remnant from early mammals, or, more trivially, may mean that the appropriate stress to expose pathology has not been identified.

ROLE OF AQUAPORINS IN HEALTH AND DISEASE

Epithelial Physiology

The major AQPs expressed in kidney include AQP1 in proximal tubule and thin descending limb of Henle epithelia, and in descending vasa recta endothelia; AQP2, the vasopressin-regulated water channel, in collecting-duct apical membrane and intracellular vesicles; and AQPs 3 and 4 at the basolateral membrane of collecting-duct epithelia (12, 19) (Figure 1*a*). Water transport across kidney tubules and microvessels is required for reabsorption of water

filtered by the glomerulus, for countercurrent multiplication and exchange mechanisms, and for vasopressin-regulated water permeability in collecting duct. Urinary concentrating function is defective in mice lacking AQPs 1–4 (20–23) and in humans with mutations in AQP1 (24) or AQP2 (25). Defective urinary concentrating function in AQP1 deficiency is the consequence of impaired near-isosmolar fluid absorption in proximal tubule and impaired countercurrent mechanisms (26–28), resulting in reduced medullary hyperosmolality; in deficiency of AQPs 2–4 there is impaired vasopressin-regulated water permeability in collecting duct (20, 21). Reduced AQP2 expression is often associated with acquired forms of nephrogenic diabetes insipidus (NDI), such as that due to lithium therapy (29). AQPs thus play a central role in the regulation of urinary salt and water excretion. AQP inhibitor “aquaretic” therapy of refractory edema is an intriguing yet so far unrealized possibility.

AQPs are also expressed in various secretory epithelia including exocrine glands, choroid plexus, and the ocular ciliary epithelium (Figure 1*b*). AQP5 deletion in mice impairs fluid secretion by salivary (30) and airway sub-mucosal (31) glands, resulting in reduced secretion of a relatively hyperosmolar fluid. AQP1 deletion in mice reduces intracranial pressure by slowing cerebrospinal fluid secretion by choroid plexus (32), and reduces intraocular pressure by slowing aqueous humor secretion by ciliary epithelium (33). Topical AQP1 inhibitors, when available, may be useful for therapy of intraocular hypertension in glaucoma. As shown in Figure 1*b*, active, near-isosmolar fluid transport involves water movement across a highly water-permeable epithelium in response to osmotic gradients produced by salt transport. Reduced epithelial cell water permeability results in the secretion of a relatively low volume of a hyperosmolar fluid. AQPs are also expressed in epithelia in airways and alveoli in lung, sweat and lacrimal glands, and several gastrointestinal organs. However, AQP deletion in these epithelia, though reducing their osmotic water permeability, does not have demonstrable physiological consequences, probably because the rate of transepithelial fluid transport in these epithelia is much lower than in kidney proximal tubule or glandular epithelia (13).

Brain Swelling

AQP4 is expressed in astrocytes throughout the CNS, particularly at interfaces between brain parenchyma and cerebrospinal fluid in the ventricular and subarachoid compartments (Figure 1*c*). There are two major types of brain edema that can occur independently or together. In cytotoxic (cellular) brain edema, as in water intoxication, water moves into the brain through an intact blood–brain barrier in response to osmotic driving forces. Mice lacking AQP4 show improved clinical outcome and reduced brain water accumulation compared to wild-type mice in water intoxication and in other models where cytotoxic brain edema is prominent, including ischemic stroke and bacterial meningitis (34, 35). Transgenic AQP4 overexpression in mice worsens brain swelling in water intoxication (36). In vasogenic (leaky-vessel) brain edema, as in brain tumor edema, water moves into the brain by bulk fluid flow through a leaky blood–brain barrier and exits the brain through the AQP4-rich glia limitans lining brain ventricles and the brain surface. When these water exit routes are impaired in obstructive hydrocephalus, water exit through the blood–brain barrier becomes more significant. AQP4 knockout mice manifest worse clinical outcome and greater brain water accumulation than wild-type mice in brain tumor edema and in other models of vasogenic edema including intraparenchymal fluid infusion, cortical-freeze injury, and brain abscess (37, 38). AQP4-null mice also manifest accelerated brain swelling in obstructive hydrocephalus (39). As a bidirectional water channel, AQP4 facilitates brain water accumulation in cytotoxic edema and clearance of excess brain water in vasogenic and interstitial edema. In spinal cord, AQP4 deficiency is associated with reduced swelling and improved clinical outcome in a model of compression injury, where cytotoxic edema

predominates (40), but with greater swelling in a model of spinal cord contusion injury, where vasogenic edema predominates (41). Recent data show greatly attenuated autoimmune neuroinflammation in AQP4-deficient mice in experimental autoimmune encephalomyelitis (42), where it was concluded that AQP4 deficiency reduces astrocyte swelling and cytokine release, as well as local cytotoxic edema. These findings imply a central role of AQP4 in brain and spinal cord water balance, which is relevant to tumor, infection, trauma, stroke, hydrocephalus, and neuroinflammation.

Cancer—AQPs in Cell Migration and Proliferation

The involvement of AQPs in cell migration has implications for tumor angiogenesis, local invasion, and metastasis. Tumor microvessels strongly express AQP1, and many tumors express AQPs, with AQP expression correlating with tumor grade in glioblastomas and some other tumors (43). The discovery of AQP involvement in cell migration followed from the finding of impaired tumor growth and angiogenesis in mice lacking AQP1, and impaired migration of AQP1-deficient aortic endothelial cells in culture (44). Further evidence, including AQP polarization to the leading end of migrating cells, increased lamellipodial dynamics in AQP-expressing cells, osmotic gradient-dependent cell migration, and AQP-dependent cell migration in different cell types and with different AQPs, suggested a possible mechanism for AQP-facilitated migration (45). As shown in Figure 1*d*, actin depolymerization and ion influx increase cytoplasmic osmolality at the leading edge of a migrating cell, driving water influx through the plasma membrane. Water influx causes expansion of the adjacent plasma membrane by increased hydrostatic pressure, which is followed by actin repolymerization to stabilize the membrane protrusion. In support of this idea is the observation that regional hydrostatic pressure changes within cells do not equilibrate throughout the cytoplasm on scales of 10 μm and 10 sec, and could thus contribute to the formation of localized cell-membrane protrusions (46). Regardless of the exact biophysical mechanism, AQP-facilitated cell migration appears to be a general phenomenon relevant not only to angiogenesis but also to tumor spread, glial scarring, wound healing, and likely other phenomena including immune-cell chemotaxis. AQP1 expression in tumor cells increases their ability to extravasate across blood vessels and to invade locally (47). AQP4 expression in brain astrocytes increases their migration toward a chemotaxic stimulus and increases glial scarring (48, 49). AQP3 expression in skin and cornea facilitate wound healing (50, 51).

Also of relevance to cancer is the involvement of aquaglyceroporin AQP3 in cell proliferation, which was discovered in various AQP3-expressing cell types, including skin, colon, and cornea. AQP3-deficient mice manifest impaired cutaneous wound healing (50), colonic epithelial cell regeneration (52), and corneal wound healing (51). A remarkable tumor phenotype was found in AQP3-null mice, which showed complete resistance to formation of skin tumors in response to a tumor initiator-promoter protocol that produced multiple tumors in wild-type mice (53). Biochemical studies in epidermal cells showed impaired cellular glycerol metabolism and biosynthesis in AQP3 deficiency, with reduced ATP content and impaired MAP kinase signaling (Figure 2*a*). These studies implicated AQP3-facilitated glycerol transport as a key determinant of cell proliferation in some cell types. The possibility of AQP3 inhibition to prevent or treat skin and certain other tumors is intriguing because AQP3 inhibition is predicted to reduce both tumor-cell migration and proliferation.

Epilepsy—AQPs in Neuroexcitation

AQP4 is expressed in neural tissues in supportive cells adjacent to excitable cells, including glia (but not neurons) in brain, Müller cells (but not bipolar cells) in retina, supportive cells (but not hair cells) in inner ear, and support cells (but not olfactory receptor neurons) in

olfactory epithelium. Involvement of AQP4 in neuroexcitatory phenomena was demonstrated by electrophysiological studies showing impairment in vision (54), hearing (55), and olfaction (56) in AQP4-deficient mice. Also, the threshold for seizure initiation is reduced in AQP4 deficiency, and seizure duration and intensity are increased (57). Possible mechanisms for these phenomena supported by experimental data include delayed K^+ reuptake by astrocytes in AQP4 deficiency following neuroexcitation (57, 58), and mild extracellular space (ECS) expansion (59, 60) (Figure 1*e*). Slowed K^+ reuptake in brain following neuroexcitation would prolong seizure duration, as found experimentally. The link between K^+ reuptake by astrocytes and AQP4 water permeability is not known. One widely speculated possibility, functional interaction between AQP4 and the inwardly rectifying K^+ channel, Kir4.1, was ruled out by patch-clamp analysis (61). We postulate an alternative, "pseudosolvent drag" mechanism in which AQP4-dependent water permeability enhances K^+ transport. Excess K^+ released into brain extracellular space (ECS) from neurons during neuroexcitation is taken up largely by the AQP4-containing astrocytes, driving osmotic water influx and consequent ECS shrinkage, which maintains the electrochemical driving force for K^+ reuptake. Reduced astrocyte water permeability in AQP4 deficiency would reduce ECS contraction and slow K^+ reuptake. Together with evidence of altered AQP4 expression in human epileptic brain (62), AQP4 modulation has been proposed as an adjunctive treatment for epilepsy. In the peripheral nervous system, AQP1 is expressed in dorsal root ganglion neurons that carry sensory signals through small-diameter nonmyelinated C-fibers. Mice lacking AQP1 manifest impaired nociception to inflammatory thermal and cold pain (63), which may involve AQP1 interaction with the $Na_v1.8$ Na^+ channel as well as water-cation coupling in the ECS of peripheral nerve bundles.

Skin

AQP3-facilitated glycerol transport in skin is an important determinant of epidermal and stratum corneum hydration (16). Mice lacking AQP3, which is normally expressed in the basal layer of proliferating keratinocytes in epidermis, manifest reduced stratum corneum hydration and skin elasticity, as well as impaired stratum corneum biosynthesis and wound healing (64). The reduced skin hydration and elasticity in AQP3 deficiency is caused by impaired epidermal-cell glycerol permeability (Figure 2*b*), resulting in reduced glycerol content in the stratum corneum and epidermis (65). Because of the humectant property of glycerol, reduced stratum corneum glycerol reduces its water content. Because of its role in biosynthesis and intracellular energetics and signaling, as mentioned above, reduced epidermal glycerol impairs epidermal cell proliferation in wound healing (50). Topical or systemic glycerol administration corrected these defects (66), which provided a scientific rationale for the inclusion of glycerol in topical and medicinal skin formulations. Dysregulation of AQP3 expression has been found in skin diseases associated with altered epidermal proliferation (67, 68); however, changes in AQP3 expression are probably a secondary consequence of the underlying pathology rather than a primary cause of disease.

Obesity

The aquaglyceroporin AQP7 is expressed in the plasma membrane of adipocytes. AQP7-deficient mice manifest progressive increase in fat mass and adipocyte hypertrophy as they age, with accumulation of glycerol and triglycerides in adipocytes (69, 70). Biochemical studies support the conclusion that adipocyte hypertrophy in AQP7 deficiency is the consequence of reduced plasma membrane glycerol permeability, with cellular glycerol and triglyceride accumulation, and glycerol kinase upregulation (Figure 2*c*). These findings suggest adipocyte glycerol permeability as a novel regulator of adipocyte metabolism and whole-body fat mass, raising the possibility of modulation of adipocyte AQP7 expression and/or function to alter fat mass. AQP9 has been proposed as an important route for hepatic

glycerol uptake (71), and AQP7 and AQP9 as metabolic regulators in diabetes and obesity (72), although convincing experimental evidence is lacking.

AQUAPORINOPATHIES

AQP Loss-of-Function Mutations and Nephrogenic Diabetes Insipidus

Loss-of-function mutations in AQPs can cause human disease, albeit very rarely. Mutations in AQP2 produce non-X-linked nephrogenic diabetes insipidus (NDI), both by a recessive mechanism in which a mutant AQP2 protein causes defective cellular processing and/or function, and by a dominant mechanism in which mutant AQP2 prevents plasma membrane targeting of wild-type AQP2 (25, 73). NDI caused by AQP2 mutation (incidence ~1 in 20 million births) is characterized by severe polyuria and polydipsia that are refractory to antidiuretic hormone. Current therapy involves replacing water losses and minimizing urinary water loss with thiazides to impair urinary diluting ability. The possibility of pharmacological chaperone therapy for some forms of NDI caused by AQP2 mutations is supported by evidence from cell and mouse models (74, 75), and gene replacement/stem cell therapy remains a theoretical possibility. For other AQPs only a handful of subjects have been identified with loss-of-function mutations. The few subjects who lack functional AQP1, who were identified by blood-group screening, are phenotypically normal but manifest defective urinary concentrating function when deprived of water (24), similar to AQP1-null mice. Because human deficiencies of AQPs 1, 3, or 7 are so rare and phenotypes so variable, little useful information is available about the roles of these AQPs in humans. Mutations in the major intrinsic protein (MIP, AQP0) of the lens cause congenital cataracts (10). Recent results suggest, however, that the primary function of MIP in lens involves cell–cell adhesion and gap–junction channel regulation rather than water transport (76). Disease-causing mutations of other AQPs in humans have not been described.

AQP4 and Neuromyelitis Optica

The involvement of AQP4 in neuromyelitis optica (NMO), a neuroinflammatory demyelinating disease, was quite unexpected. NMO and multiple sclerosis share some similarities, but NMO primarily affects optic nerve and spinal cord, causing blindness and paralysis, and has characteristic pathological and clinical features that distinguish it from multiple sclerosis (77). NMO is a rare disease in Caucasians (incidence ~1 in 100,000) but is more common in Asians. As in other autoimmune diseases, females are affected more frequently than males (7:1 ratio). The defining feature in NMO is the presence of serum autoantibodies (NMO-IgG) directed against extracellular epitopes on AQP4 (78). NMO-IgG seropositivity is highly specific for NMO, and, in some studies, NMO-IgG levels correlate with disease activity. There is emerging evidence for a pathogenic role of NMO-IgG in NMO, as administration of human NMO-IgG produces NMO-like pathology in rats with pre-existing neuroinflammation (79). Naïve mice injected intracranially with human NMO-IgG and complement develop characteristic NMO lesions, with neuroinflammation, loss of glial fibrillary acidic protein (GFAP) and AQP4 immunore-activity, myelin loss, and perivascular deposition of activated complement (80). It is thought that IgG binding to AQP4 in astrocytes initiates an inflammatory cascade involving recruitment of leukocytes (granulocytes, macrophages, NK cells, lymphocytes), cytokine release, and complement and NK cell–mediated astrocyte damage. The consequent neuroinflammation and myelin loss produce neurological deficits. The events involved in the initiation of NMO-IgG production and CNS penetration remain unknown, as do the reasons why NMO pathology is much more prevalent in spinal and optic nerve than in brain and why it is absent in peripheral AQP4-expressing organs.

Current NMO therapies are directed toward reducing the inflammatory response (immunosuppression) and the NMO-IgG load (B cell depletion and plasmapheresis). A complement-targeted monoclonal antibody therapy is in clinical trials. An intriguing possibility is the development of monoclonal antibody or small-molecule blockers of the binding of pathogenic NMO-IgG AQP4, the presumed initiating event in NMO pathogenesis. Recent proof-of-concept studies have demonstrated that high-affinity, non-pathogenic, recombinant NMO antibodies (aquaporumabs) can block NMO-IgG binding to AQP4 and prevent consequent cell killing and development of NMO lesions in ex vivo and in vivo animal models. Among autoimmune diseases, NMO is uniquely suitable for blocker therapy because of its well-defined and physically small target, AQP4.

AQUAPORIN-BASED DIAGNOSTICS AND THERAPEUTICS

AQP Diagnostics

The most prominent example of an AQP antibody-based diagnostic is assay of serum AQP4 autoantibody (NMO-IgG) in NMO (78). The most recent data suggest that NMO-IgG seropositivity is nearly 100% sensitive and specific for NMO (81). Circulating AQP autoantibodies may be of utility for diagnosis of other diseases and possibly involved in their pathogenesis, such as, perhaps, AQP3 autoantibodies in autoimmune skin diseases and AQP5 autoantibodies in Sjögren's syndrome. Assay of AQP protein in bodily fluids and tissue specimens may have diagnostic value. The best example is assay of AQP2 immunoreactive protein in urine for distinguishing among various etiologies of NDI (82). The rationale for urinary AQP2 assay is the shedding, by an exosomal mechanism, of AQP2 protein when expressed at the luminal membrane of kidney collecting duct. However, diagnostic assay of urinary AQP2 has received little attention because alternative, reliable methods are available to evaluate NDI. The possibility of shedding of other AQPs in urine, such as AQP1 in proximal tubule injury, has only recently received consideration, as has shedding of AQPs in other bodily fluids, such as in aqueous humor or cerebrospinal fluid. Another potential role for AQP protein–based diagnostics is in evaluating AQP expression in tissue specimens. For example, AQP expression in tumor cells has been correlated with tumor grade (43), and altered AQP expression has been reported in epilepsy (62) and in ocular (15) and skin (83) diseases. Whether diagnostically informative or unique information can be obtained by such measurements remains to be seen. Last, the possibility of functionally significant AQP polymorphisms has only begun to receive attention, and it remains too early to predict the ultimate usefulness of such information. A few recent studies have investigated possible polymorphisms in AQPs associated with stroke, migraine, temporal lobe epilepsy, diabetes, and obesity (84–88), although no compelling associations have been identified.

Potential for AQP-based Therapeutics

There are compelling opportunities, yet so far little progress, in the area of AQP-based therapeutics. One area, as mentioned above, is the possibility of monoclonal antibody or small-molecule blockers of NMO-IgG binding to AQP4 in NMO. Challenges in clinical development of NMO blocker therapy will be in developing antibodies or small molecules with suitable CNS penetration and in establishing efficacy criteria for a rare disease with highly variable natural history. The other major opportunity is in the development of small-molecule modulators of AQP function, including inhibitors of AQP water/glycerol transport function and transcriptional up-regulators of AQP expression. The AQPs appear to be a refractory target for drug discovery; although there have been attempts to identify small-molecule AQP inhibitors, the published data have been questioned, and no useful compounds have emerged to date. In addition to the challenges in compound discovery, challenges in their development include engineering of AQP isoform-selective inhibitors,

and, for some indications such as brain edema and epilepsy, the potentially narrow therapeutic window.

Notwithstanding these challenges, the bench science in AQP biology suggests multiple potential indications of AQP modulators. The requirement of AQPs for the formation of a concentrated urine suggests that AQP inhibitors, or aquaretics, would reduce urine concentration, producing a water > salt diuresis. AQP1 inhibitors are predicted to have utility in diuretic-refractory edematous states, such as severe congestive heart failure, where conventional salt-blocking diuretics are of limited efficacy. Inhibitors of AQP4 are predicted to reduce brain swelling in cytotoxic edema, potentially offering neuroprotection following some types of brain and spinal cord injury, ischemic stroke, and infection. However, the predicted slowed clearance of excess brain water in vasogenic edema with AQP4 inhibition would mandate great care in the timing of therapy. Inhibitors of AQPs in tumor cells and microvessels are predicted to reduce tumor spread and angiogenesis, offering adjunctive tumor chemotherapy. Inhibition of AQP4-facilitated glial cell migration is predicted to inhibit glial scar formation following brain and spinal cord injury, promoting axonal regeneration and improving long-term neurological outcome. Topical inhibitors of AQP1 in the eye may reduce intraocular pressure in glaucoma, and inhibitors of AQP3 in the skin may reduce skin cancer. AQP1 inhibitors may be useful in pain management. Compounds that increase AQP function, acting by increasing AQP expression, are predicted to have efficacy in reducing fat mass in obesity, in accelerating brain water clearance in vasogenic edema, in promoting wound healing and tissue regeneration following injury, and in inhibiting cataractogenesis. Although studies in transgenic mice or naturally occurring human AQP mutations offer proof of concept for these indications, human clinical trials will ultimately establish the suitability of AQP-based therapeutics in human disease.

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SUMMARY POINTS

1. The aquaporins are a family of conserved membrane channels that transport water, and in some cases small solutes such as glycerol as well.
2. Water-selective aquaporins are involved in epithelial fluid transport, brain swelling, cell migration, and neuroexcitation; water/glycerol-transporting aquaporins (aquaglyceroporins) are involved in skin hydration, cell proliferation, and adipocyte metabolism.
3. Aquaporins are strongly expressed and functionally important in kidney, central nervous system, eye, skin, and exocrine glands. Aquaporins are expressed but probably not functionally important in lung, gastrointestinal organs, and muscle.
4. Human aquaporinopathies include neuromyelitis optica, an autoimmune neuroinflammatory disease caused by anti-AQP4 antibodies; nephrogenic diabetes insipidus, caused by AQP2 mutation; and congenital cataracts, caused by AQP0 mutation.
5. Aquaporin-based drug therapy has potential utility for some types of refractory edema, brain swelling, neuroinflammation, glaucoma, epilepsy, cancer, pain, and obesity.

FUTURE ISSUES

1. Identification and development of small-molecule aquaporin-selective inhibitors.
2. Development of monoclonal and small-molecule blocker therapy for neuromyelitis optica.
3. Elucidation of the relevance of aquaporin polymorphisms to human diseases.
4. Clarification of the relevance and diagnostic utility of altered aquaporin expression to human diseases.
5. Determination of precise molecular mechanisms of aquaporin-dependent cell migration and proliferation, neuroexcitation, neuroinflammation, adipocyte metabolism, and pain.

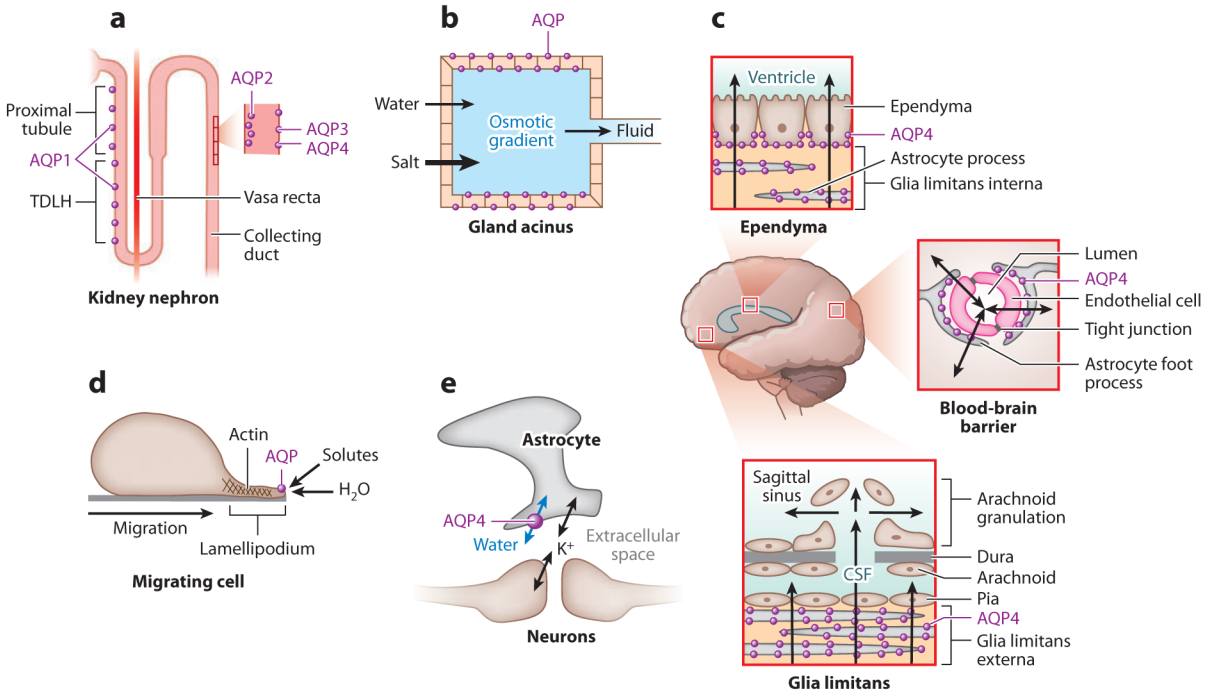

Figure 1.

Roles of water-selective aquaporins (AQPs, *shown in purple*). (*a*) Kidney nephron. High transepithelial water permeability in proximal tubule, thin descending limb of Henle (TDLH), vasa recta, and collecting duct is required for urinary concentrating function. (*b*) Epithelial fluid secretion (exocrine glands, ICP/IOP, etc.). High transepithelial water permeability facilitates active, near-isosmolar fluid secretion. (*c*) Brain water balance (cytotoxic and vasogenic edema). High water permeability across blood–brain and blood–CSF barriers facilitates water movement into and out of brain. (*d*) Cell migration (angiogenesis, tumor metastasis, glial scarring, etc.). AQP-facilitated cell migration involves water entry into protruding lamellipodia in migrating cells. (*e*) Neuroexcitation (neurosensory function, seizure activity, etc.). AQP4-facilitated water transport in astrocytes during K^+ reuptake following neuroexcitation causes extracellular space contraction, maintaining the driving force for K^+ reuptake. Abbreviations: CSF, cerebrospinal fluid; ICP, intracranial pressure; IOP, intraocular pressure.

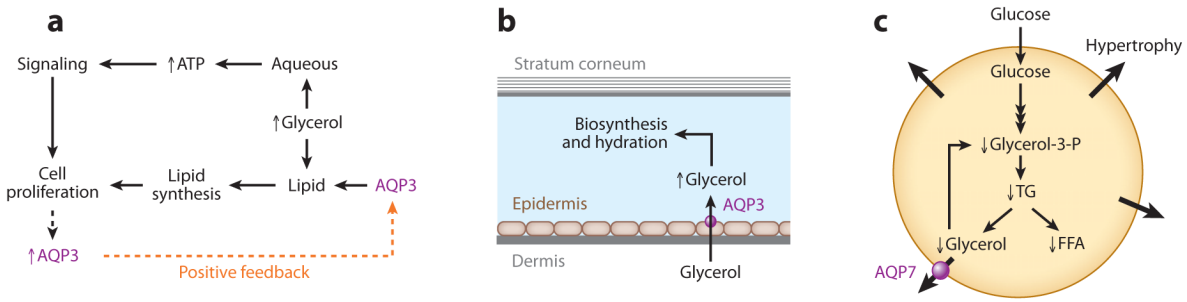

Figure 2.

Roles of water/glycerol-transporting aquaporins (aquaglyceroporins). (*a*) Cell proliferation (tumor growth, wound healing, etc.). AQP3 maintains high cellular glycerol for generation of ATP and biosynthesis. (*b*) Skin hydration. AQP3 maintains high stratum corneum glycerol, which acts as a humectant to retain water. (*c*) Adipocyte metabolism. AQP7 facilitates glycerol exit from adipocytes, preventing intracellular glycerol and triglyceride accumulation. Abbreviations: AQP, aquaporin; ATP, adenosine triphosphate; FFA, free fatty acid(s); TG, triglyceride(s).