DOI: 10.1113/jphysiol.2002.024398
www.jphysiol.org

PERSPECTIVES

Does aquaporin-1 pass gas? An opposing view

A. S. Verkman

Departments of Medicine and Physiology, Cardiovascular Research Institute, University of California, San Francisco CA 94143-0521, USA

verkman@itsa.ucsf.edu

The possibility that aquaporin-1 (AQP1) facilitates CO_2 transport across biological membranes is an intriguing one with potentially numerous consequences in mammalian physiology. However, such an unorthodox conjecture must pass tests of physical reasonableness and be validated by incisive yet simple-to-understand experimental methods. The conjecture that AQP1 passes CO_2 is unorthodox because biomembranes are in general highly gas permeable, so much so that unless extraordinary experimental approaches are used CO_2 permeability is limited not by the intrinsic membrane permeability to CO_2 but by 'unstirred layers' outside of the membrane (Geers & Gros, 2000).

Can CO_2 pass through AQP1, and if so, can the increase in intrinsic membrane CO_2 permeability conferred by AQP1 significantly augment CO_2 transport in real tissues? It is not possible on theoretical grounds to exclude the possibility that some CO_2 can pass through AQP1 or other proteins. Recent molecular dynamics simulations of water movement through AQP1 suggest, however, that water molecules twist and turn as they wind through the narrow, single-file pore in AQP1 monomers. Whether the larger CO_2 molecule can do so is unknown. Of greater theoretical concern is that membranes where AQP1 is expressed – erythrocytes, lung endothelia and kidney proximal tubule – are highly CO_2 permeable. CO_2 transport is probably rate limited in these tissues by unstirred layers, and so is unlikely to increase even if AQP1 were infinitely permeable to CO_2.

What about the experimental evidence to support or refute the idea that AQP1 is a CO_2 channel? As detailed in the accompanying review in this issue of *The Journal of Physiology* by Boron and colleagues (Cooper *et al.* 2002), the principal evidence is that CO_2 permeability in *Xenopus* oocytes expressing large amounts of AQP1 is ~40 % greater than that of control oocytes (Nakhoul *et al.* 1998). These are technically complex experiments in which oocytes are first microinjected with cRNA. At a later time they are microinjected with carbonic anhydrase to catalyse CO_2/HCO_3^-/pH equilibration. The oocytes are then impaled for a third time with a pH-sensing microelectrode to measure cytoplasmic acidification in response to addition of CO_2 to the bathing solution. In a different protocol designed to test whether CO_2 permeability through AQP1 could enhance NBC-mediated Na^+/HCO_3^- cotransport by locally shunting CO_2, zinc-dependent oocyte acidification was measured after microinjection with a mixture of cRNAs encoding NBC, two carbonic anhydrase isoforms and AQP1. The results of this complex study were interpreted in terms of AQP1-dependent CO_2 shunting, however the possibility of NBC-adjacent CO_2 transport seems unlikely because CO_2 diffuses rapidly in cytoplasm and there is no evidence for physical coupling of NBC and AQP1, which are probably expressed in very different amounts.

As detailed in Yang *et al.* (2000) and in a new study reported in this issue of *The Journal of Physiology* (Fang *et al.* 2002), we investigated whether CO_2 transport by AQP1 could be detected under physiological and super-physiological conditions in erythrocytes, lung, kidney and reconstituted proteoliposomes. Organ studies were done comparing normal mice and transgenic mice lacking AQP1. Carbon dioxide permeability in erythrocytes and kidney proximal tubule membranes was very fast (equilibration time a few milliseconds) and was not reduced in AQP1 knockout mice. CO_2 permeability in AQP1-reconstituted proteoliposomes was not different from that in control liposomes, even though water permeability was >100-fold different. In each of these systems intrinsic membrane CO_2 permeabilities should be even higher due to unstirred layer effects. CO_2 blow-off by the lung was not AQP1-dependent under physiological conditions in living mice, and airspace-capillary CO_2 permeability was not affected by AQP1 deletion under super-physiological conditions in a rapidly perfused lung preparation.

From these considerations and experimental results it appears unlikely that CO_2 transport by AQP1, if it occurs, is important in mammalian physiology. However, the possibility that small gases such as CO_2, NH_3 and NO are transported by proteins should not be ignored. The relatively non-selective aquaglyceroporins, or other abundant proteins such as Rh factors, should be evaluated for their gas transporting ability.

Cooper, G. J., Zhou, Y., Bouyer, P., Grichtchenko, I. I. & Boron, W. F. (2002). *Journal of Physiology* **542**, 17–29.

Fang, X., Yang, B., Matthay, M. A. & Verkman, A. S. (2002). *Journal of Physiology* **542**, 63–69.

Geers, C. & Gros, G. (2000). *Physiological Reviews* **80**, 681–715.

Nakhoul, N. L., Davis, B. A., Romero, M. F. & Boron, W. F. (1998). *American Journal of Physiology* **274**, C543–548.

Yang, B., Fukuda, N., Van Hoek, A., Matthay, M. A., Ma, T. & Verkman, A. S. (2000). *Journal of Biological Chemistry* **275**, 2686–2692.

Presented at The Journal of Physiology Synthesium on Water Transport Controversies, Christchurch, New Zealand, 30 August, 2001.