Major advances in science, especially in formerly becalmed fields, create not only new insights but excitement as well. Such is the case with the discovery by Peter Agre and his colleagues (1) that the protein called aquaporin-CHIP (AQP-CHIP) serves as the water channel in the plasma membrane of human red blood cells. AQP-CHIP thus functions to mediate net movements of water into and out of cells, depending on the direction of the net osmotic force (see reference 1 and references therein). Its occurrence in different cell types and the recognition of certain of its molecular relatives have also been established. The paper contained in this issue of the *Journal* is an extension of this kind of work defining the timed appearance in red cells and kidneys of functional AQP-CHIP water channels that occurs during growth and development (1).

By way of background, the existence of water channels in red cell membranes was first deduced from the finding that the ratio of the osmotic (P_f) to the diffusional (P_{dw}) permeability of cells to water was not only greater than 1 but approximated 10 (2, 3). A pivotal development with regard to the mechanisms involved was provided by the finding that the red cell water channels could be closed by mercurials, such as parachloromercuribenzene sulfonate (PCMBS), resulting in a reduction in the P_f/P_{dw} ratio to 1, the same as that exhibited by lipid bilayers (4). In addition, the remaining flux, represented as a slightly lower P_{dw} value, turned out to be equivalent to the water permeability of pure lipid bilayers (see reference 3 for a lucid and critical account of the basics of this field). The foregoing results, as well as other types of studies, in pointing to the reality of water channels, set the stage for the discovery of the AQP-CHIP protein and provided needed criteria for its identification. It should also be evident that the notion that plasma membrane structure involves proteins inserted into a lipid bilayer is nowhere better supported than by studies of the water permeability of human red blood cells.

AQP-CHIP is known to have a molecular mass of 28.5 kD (in its unglycosylated form) and evidently resides as a tetramer in the membrane. The human red blood cell harbors some 200,000 copies of AQP-CHIP per cell. Purified AQP-CHIP has been incorporated into liposomes and its cloned gene has been expressed in xenopus oocytes. In both of these preparations osmotic water permeability was only observed when AQP-CHIP was present and their respective values of P_f were comparable to those obtained with intact cells. In addition, PCMBS inhibited the osmotic water permeability to its control level (no AQP-CHIP) in both preparations. Importantly, the permeability of AQP-CHIP in reconstituted liposomes was limited to water. AQP-CHIP is impermeable not only to ions, including protons, but to polar solutes, such as urea, as well. Evidently water transport through AQP-CHIP is by a single-file mechanism, in analogy with water movement through gramicidin A (3). The energy of activation of water flow through AQP-CHIP is consistent with this view. The number of water molecules in the channel is considered to be approximately 10 as defined by the ratio of P_f/P_{dw} (3). It is to be hoped that detailed information will be forthcoming about the structure of the water channel, how water moves through it, and the identification of the amino acid residues that define the channel.

The results presented in the current paper (1) show by immunochemical and functional means that AQP-CHIP is essentially absent in neonatal red cells of the rat. After birth AQP-CHIP appears in the red cells and increases within several weeks to the adult level of expression. The neonatal kidney, while displaying low levels of AQP-CHIP expression, has a parallel increase in the amount (and distribution) of AQP-CHIP in the proximal tubules and the descending thin limbs of the loops of Henle, commensurate with the kidney's ability to form concentrated urine. The basis for this concordant differentiation in the two types of tissue is unknown, as well as are the details concerning the synthesis and delivery of AQP-CHIP to the surface membranes and how these events are controlled. There is an additional question of how the switch from fetal to adult red cell lines is accomplished. It would of course be interesting to know the stage in hematopoiesis where AQP-CHIP first appears. While similar questions could be directed toward the differentiation of other cell types, the lung would seem to offer special relevance. If, as suggested (5), the lung at birth clears fluid from the alveolar and bronchial spaces by synthesis of Na,K-ATPase to increase the fluid reabsorption by increasing the transcellular transport of Na, then fluid reabsorption may be coordinately enhanced by the presence and/or insertion of AQP-CHIP water channels.

What is the function of water channels in red blood cells? The authors (1) suggest that the water channels act to promote the rehydration of the cells after their shrinkage in the hypertonic environment of the renal medulla. Rapid rehydration would return the cells to their normal volume, optimizing their deformability for transit in the microcirculation. This explanation represents the flipside of Macey's proposal (4) regarding the role that the red cell's high (facilitated) permeability to urea plays as the cells circulate through the medulla. Since urea's entry would reduce the extent of cell shrinkage, rehydration would be enhanced on leaving the medulla. This situation has been modeled (4) for cells with low and high urea transport during their circulation through the medulla. While in the latter case the cells appear to return to their original volume when they return to the cortex, the cells in the former case swell beyond their isotonic volume (and possibly to their hemolytic volume), depending on the amount of urea that entered the cells in the medulla. If this model is correct it would seem that the need for water channels is linked to the cells possessing a rapid urea transport system. So it would be interesting to know the predictions depending on the presence and absence of water channels with and without high urea transport, incorporating more recent estimates of blood flow in the medulla (6). In this regard, the red cells of individuals lacking the Kidd blood group antigens also lack the facilitated urea transport system (7), but whether or not AQP-CHIP is present is not known.

Another approach to studying the relationship between water channels in red cells and renal function is to make a quantitative comparison between the amount of AQP-CHIP

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(and/or their PCMBS-sensitive values of P_f) in red cells of different mammals as a function of the renal concentrating ability of their kidneys (cf. reference 8). It would also be of interest to know the comparative content of AQP-CHIP (as well as their relative P_f's) among different vertebrate species, for they are known to show considerable variation (4, 9). Perhaps this kind of information will provide new insights into water channel function in red blood cells.

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