lateral rat descending colonic mucosa and showed that recovery of fluorescence within the crypt lumen was the same for widely different molecular weight dextrans and abolished by inhibition of epithelial Na transport. These studies provide evidence that fluid flow in colonic crypts is convective.3. The generation of a sufficiently strong suction pressure at crypt openings requires a highly hypertonic compartment, postulated to be localized in the pericryptal space surrounding crypts. Confocal microscopy performed in vivo in mice using a low affinity ratiometric Na sensitive dye confirmed the presence of a pericryptal hypertonic compartment with a local Na concentration of 200-400 mM, which would be sufficient to provide the necessary osmotic pressure for convective flow into crypts. Inhibition of Na transport significantly reduced pericryptal Na concentration as did blockade of crypt opening using paraffin oil suggesting that pericryptal hypertonicity is dependent on salt transport from the crypt lumen.

Colonic fluid transport can be altered by in a number of states resulting in changes in colonic crypt function. High doses of ionizing radiation result in a reduction in fluid absorption. Examination of colonic mucosa post-irradiation showed that this reduction in fluid absorption was associated with increased leakiness of colonic crypt cells and the loss of cell adhesion molecules. Additionally it was shown that the pericryptal sheath was significantly disrupted after radiation and these changes were preceded by the release of apoptotic enzymes and signaling molecules. The loss of the pericryptal sheath and crypt epithelial adhesion molecules coincided with increased permeability of FITC dextrans out of crypts. In contrast to irradiation, low dietary Na results in increased fluid absorption through increases in plasma aldosterone and angiotensin II. Investigation of the effects of low Na diet in rat descending colon revealed a significant trophic effect on myofibroblast cells and extracellular components of the pericryptal sheath in contrast to rat proximal colon. These changes suggest that increased pericryptal barrier function may be contribute to increased fluid absorption after low Na diet. Several in vivo and in vitro studies of colonic crypt function both in health and disease now provide evidence that crypts play an important role in colonic fluid absorption. Further studies are required to measure important determinants such as crypt luminal pressure gradients and the role of active secretion in states of disease on crypt and pericryptal function.


**REGULATION OF AQUAPORIN 2 FUNCTION**

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Body fluid balance is controlled predominantly by hormonal regulation of renal collecting duct function. The water permeability of the collecting duct is controlled by vasopressin, which causes the shuttling of aquaporin 2 (AQP2) water channels from intracellular vesicles to the apical plasma membrane of the cells. This shuttling is mediated by cAMP and activation of protein kinase A; phosphorylation of AQP2 seems to be a key step leading to its exocytic insertion into the plasma membrane. When vasopressin levels fall, AQP2 is retrieved endocytically. This acute shuttling of AQP2 is modulated by changes in AQP2 expression: vasopressin infusion, or chronic dehydration, increase AQP2 expression, while water loading decreases it. This modulation can be partly explained by vasopressin effects mediated by cAMP, but it is now clear that other factors are also involved. A number of other hormones and local factors are known to modulate antidiuresis, although in many cases the mechanisms behind this remain unclear. We have been particularly interested in possible roles of prostaglandins, angiotensin, and bradykinin. Prostaglandin E2 and angiotensin II both appear to have some ability to increase cAMP and hence mimic the effects of vasopressin, while bradykinin antagonises the effects of vasopressin. We are currently investigating the signalling cascades underlying these effects. Pathological disorders of water balance have been shown to be associated with changes in both AQP2 expression and shuttling. In particular, many acquired forms of nephrogenic diabetes insipidus are associated with a decrease in AQP2 levels, which may be profound, while some, but not others, also show impaired trafficking of AQP2. In seeking treatments for such disorders, it is important to understand both why the disorder has arisen, and possible ways to bypass it. We hope that in the long term we can find stimuli that will alter both expression and shuttling of AQP2 independent of vasopressin, and that this will lead to new treatments for water balance disorders, and potentially for other problems such as hypertension.

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Where applicable, the experiments described here conform with Physiological Society ethical requirements.