Mechanogated Channels in *Xenopus* Oocytes: Different Gating Modes Enable a Channel to Switch From a Phasic to a Tonic Mechanotransducer

OWEN P. HAMILL AND DON W. MCBRIDE JR.

*Department of Physiology and Biophysics, The University of Texas Medical Branch, Galveston, Texas 77555*

Critical to the survival of any cell is its ability to sense and respond appropriately to changes in its environment. In the case of the mechanical environment, there are both static and dynamic components that the cell may be required to selectively detect. Such detection takes place in the presence of a dynamic background of mechanical stimulation arising from Brownian motion, gravitational force, and various forces generated within a cell (e.g., due to molecular motors and cycles of cytoskeletal polymerization and depolymerization) that maintain cell shape and also mediate shape changes during growth and adhesion. In addition to such background forces, a cell may experience other mechanical perturbations, ranging from steady indentations to high-frequency vibrations, and from osmotic challenges to fluid shear stresses. Detection and appropriate responses to such perturbations may be critical for the function and, perhaps, survival of the cell. Therefore, cells require biological mechanotransducers that can extract specific information regarding relevant mechanical stimuli while filtering out irrelevant stimuli.

A variety of mechanosensitive processes have been identified including (i) mechanosensitive enzymes such as adenylate cyclase (Watson, 1990) and phospholipase A₂ (Jukka *et al.*, 1995), (ii) mechanosensitive transmitter release (Chen and Grinnell, 1995), (iii) mechanosensitive gene activation (Sadoshima *et al.*, 1992), and (iv) the widely expressed class of mechanogated (MG) membrane ion channels (Martinac, 1992). Of these different processes, the MG channels have proven the most amenable to detailed biophysical study. This has been due, in part, to the development of high-resolution patch-clamp recording (Hamill *et al.*, 1981) and fast pressure-clamp stimulating techniques (McBride and Hamill, 1992, 1993, 1995; Hamill and McBride, 1995a). We have used these techniques to study the dynamic properties of single MG channels, mainly focusing on the cation-selective channel endogenously expressed in *Xenopus* oocytes (Hamill and McBride, 1992, 1994, 1995a, 1995b; Zhang *et al.*, 1996). This channel is blocked by amiloride and its analogs, aminoglycoside antibiotics and gadolinium (for review see Hamill and McBride, 1996b). One of the most interesting kinetic features of the channel is that its gating mode can be shifted from a highly nonstationary, phasic or "high pass" mode to a stationary, tonic or "low pass" mode (i.e., the input-filter characteristics change). When in the phasic mode the MG channel activity exhibits rapid and complete adaptation (i.e., the channels close) despite the presence of maintained mechanical stimulation. This adaptation is highly voltage dependent and is similar to that seen in audiovestibular hair cells (Crawford *et al.*, 1991). For example, at -100 mV the decay time constant of adaptation is about 100 ms, while at +100 mV it is more than 2 s. However, unlike the hair cell, this adaptation does not depend on either extracellular or intracellular Ca⁺⁺ nor on the polarity of stimulation (i.e., suction or pressure). We find that the adaptation in the oocyte MG channel is due to a shift of the stimulus-response relation (Boltzmann) to the right (i.e., towards higher pressures) with no change in shape of the relation. This adaptation reduces response saturation.
while preserving the differential sensitivity to transient changes in mechanical stimulation.

When the MG channel is in the tonic mode, the open channel probability becomes time independent but increases with increasing suction or pressure stimulation. However, the sensitivity of the MG channel to mechanical activation is decreased. Although voltage-dependent adaptation is clearly absent in this mode, the voltage dependence of MG channel lifetime is preserved and single channel conductance and ion selectivity remain unaltered. The switching between gating modes can be mechanically induced in the patch-clamp configuration and is most likely due to a physical decoupling of the membrane from the underlying cytoskeleton, which presumably contains the required viscoelastic elements. This idea of decoupling is supported by the observed development of a clear space within the patch pipette between the membrane and the underlying cytoplasm; development of this space accompanies the mechanically induced channel mode switching.

The channel mode switching from phasic to tonic, as studied here, probably reflects a pathological situation associated with patch-clamp recording (Hamill and McBride, 1997). Nevertheless, it does indicate a mechanism by which a single molecular mechanotransducer may alter its input filter characteristics (i.e., go from a transient to a steady-state detector) of mechanical signals. It remains to be determined whether the remodeling of the cortical cytoskeleton that occurs during cell development and differentiation (e.g., see Vale, 1991; Sardet et al., 1994) may also modulate membrane mechanosensitivity by such a mechanism.

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Literature Cited


