M2 now serves as the prototypic member of an ever-growing family of virus-encoded ion channels, termed 'viroporins.' Examples of viroporins herald from a wide range of diverse viruses infecting humans and animals alike, including some of the most clinically and economically important viral pathogens on the planet. Although their sequences, structures and roles during viral life cycles vary, viroporins are unified by almost unanimously being essential proteins, supporting their potential as drug targets. However, despite the progress with ion channel inhibitors in other aspects of medicine, the development of novel viroporin inhibitors has rarely progressed beyond the early stages of target/hit discovery and the current repertoire of inhibitory small molecules is narrow indeed. Nevertheless, recent examples of targeted discovery are beginning to yield clinically viable compounds that should serve as pathfinders in the expansion of viroporins as drug targets and unlock their potential for the treatment of myriad viral diseases.

9.1.1 Discovery and Expansion of the Viroporin Family

Although the activity of amantadine against influenza virus entry, and at early stages in replication for some strains, was known for some time, its relationship with a virus-coded ion channel was unknown.[1] The first indications that virus infection altered host cell membrane permeability came about in the 1970s, when small-molecule antibiotics and other compounds were shown to enter virus-infected mammalian cells and inhibit translation.[2] This was initially presumed to result from the budding of nascent viral particles from the cell surface and was proposed as a means of selectively targeting virus infection using commonly available drugs. Such membrane effects have since formed the basis of many indirect methods of assessing viroporin function (see Section 9.1.3), although it is not always clear whether molecules pass directly through viral channels or enter cells indirectly as a result of viroporin-induced perturbations of cellular membrane homeostasis.

The 1990s saw a rapid expansion of viroporin research following the demonstration that M2 displayed proton channel activity in the membranes of *Xenopus* oocytes,[3] recapitulated by peptides corresponding to its *trans*-membrane region in artificial bilayers.[4] Importantly, this activity was directly linked to the amantadine-mediated blockade of influenza virus infection at both early[5,6] (entry) and late (assembly) stages[7,8] in various strains[9,10] by the requirement for virion acidification and a monensin-like dissipation of Golgi pH, respectively (see Section 9.2.1). This provided a functional context to the observed channel activity of M2 in isolation. A number of other small hydrophobic proteins were also proposed to display viroporin activity due to their effects on membrane trafficking and permeability in various systems (see Sections 9.2–9.5), including picornavirus 2B, 2BC, 3A and VP4 proteins,[11–15] alphavirus 6K,[16,17] paramyxovirus small hydrophobic (SH) protein,[18–20] rotavirus NSP4[21–23] and human immunodeficiency virus type 1 (HIV-1) Vpu.[24–26] These proteins, with M2 as the prototype, formed the core of viroporin literature and this subsequently expanded in the new millennium to