pathogens in addition to causing less severe human disease. Three proteins encoded by CoV, primarily from SARS CoV but often with homologues in other viruses, have been proposed to act as viroporins, namely E, 3a and 8a.

Peptides corresponding to the SARS CoV E protein were first shown to display cation channel activity in planar bilayers,[32] which was subsequently shown to be sensitive to HMA.[29] In turn, HMA blocked the replication of the animal coronavirus mouse hepatitis virus (MHV), but not where the entire E ORF had been deleted; such viruses were also attenuated in culture. E has been proposed to form pentameric bundles and comprise a type 1 viroporin, although its topology is a matter of some debate.[277] E channels have also been shown to be inhibited by very high concentrations of amantadine, although the relevance of this interaction in the millimolar range is not clear.[278] Solution NMR studies have yielded models of the pentameric helical bundle derived from peptides corresponding to the TM domain.[31] These were similarly inhibited by HMA in whole 293T cell patch-clamp and the drug was shown to interact with both the N-terminal and C-terminal neck of the TM domain by NMR. E is a structural component of the virion and is highly involved in both their assembly and secretion, although how channel function relates to these activities is unclear as the protein undergoes several protein–protein interactions and may adopt various topologies.[277] Interestingly, a recent report found that the lipid environment directly influenced the ionic conductance of E channels with no cation preference evident in non-charged lipid membranes, whereas negatively charged lipids induced mild cation selectivity. In addition, Asn15Ala and Val25Phe mutations located in the TM domain abrogated channel activity.[28]

In addition to E, the 3a CoV protein was shown to display channel activity.[27] The 3a formed potassium-selective channels in oocytes and recombinant protein tetramerised in membranes, stabilised through disulfide linkages. Emodin, a constituent of various plant extracts including Polygonaceae (Japanese knotweed), inhibited 3a channels with an EC_{50} of $\sim 20\,\mu M$ and this molecule also prevented the spread of a model human coronavirus at similar concentrations.[34] However, the action of this drug against multiple cellular kinases (*e.g.* $p56^{lck}$)[279–281] makes it difficult to assign specificity to 3a in such experiments. Finally, a recent report showed that ORF8a single TM peptides form pentameric cation-selective channels *in vitro*, with modelling predicting a lumen lined by cysteines, serines and threonines.[282]

It remains unclear whether one, some or all of the proteins reported to act as viroporins in CoV perform this function during human infection; however, the large genomes of CoV certainly provide sufficient coding capacity to retain multiple channel proteins. It would be of great interest to determine precisely which aspects of the CoV life cycle are sensitive to HMA and/or emodin and whether resistant viruses can be selected in culture for either compound.

9.3.3 The Small Hydrophobic (SH) Proteins of Paramyxoviridae

Three genera of the Paramyxoviridae encode small hydrophobic (SH) proteins, namely the pneumoviruses, metapneumoviruses and rubulaviruses. Of these,