Thermostability of the Foot-and-Mouth Disease Virus Capsid Is Modulated by Lethal and Viability-Restoring Compensatory Amino Acid Substitutions

Infection by viruses depends on a balance between capsid stability and dynamics. This study has investigated biologically and biotechnologically relevant aspects of the relationship in foot-and-mouth disease virus (FMDV) between capsid structure and thermostability, and between thermostability and infectivity. In the FMDV capsid a substantial number of amino acid side chains at the interfaces between pentameric subunits are charged at neutral pH. Here a mutational analysis revealed that the essential role for virus infection of most of the 8 tested charged groups is not related to substantial changes in capsid protein expression or processing, or in capsid assembly or stability against thermally-induced dissociation into pentamers. However, the positively charged side chains of R2018 and H3141, located at the interpentameric interfaces close to the capsid 2-fold symmetry axes, were found to be critical both for virus infectivity and for keeping the capsid in a state of weak thermostability. A charge-restoring substitution, N2019H, that was repeatedly fixed during amplification of viral genomes carrying deleterious mutations, reverted both the lethal and capsid-stabilizing effects of substitution H3141A, leading to a double mutant virus with close to normal infectivity and thermostability. H3141A and other thermostabilizing substitutions had no detectable effect on capsid resistance to acid-induced dissociation into pentamers. The results suggest that FMDV infectivity requires limited local stability around the 2-fold axes at the interpentameric interfaces of the capsid. The implications for the mechanism of genome uncoating in FMDV and the development of thermostabilized vaccines against foot-and-mouth disease are discussed. IMPORTANCE This study provides novel insights into the little known structural determinants of the balance between thermal stability and instability in the capsid of foot-and-mouth disease virus, and into the relationship between capsid stability and virus infectivity. The results provide new guidelines for the development of thermostabilized empty capsid-based recombinant vaccines against foot-and-mouth disease, one of the economically most important animal diseases worldwide.