Abstract

The capsids of plant ssRNA viruses represent a collection of evolutionarily optimized proteinaceous nanoparticles of different shapes and sizes that are not infectious to mammals and therefore have a high application potential. In contrast to icosahedral and rod-shaped viruses, the potential of the widespread flexible filamentous plant viruses has not yet been fully explored. Potato virus Y (PVY) belongs to the group of potyviruses, the largest group of flexible filamentous viruses, and is the most important viral pathogen of potato worldwide. The capsid/coat protein (CP) exhibits a high degree of intrinsic disorder and structural plasticity, which is strongly related to its numerous functions during the viral infection cycle. As the sole structural protein it is assembled in a left-handed helix around viral (+)ssRNA and forms flexible filamentous virions. While it remains unclear in what structural context the CP fulfils its multiple roles during the viral life cycle, the structural landscape available to the potyviral CP can be ascertained by studying recombinantly produced virus-like particles (VLPs).

Using combined structural and biophysical approaches, we have elucidated the role of intrinsically disordered regions (IDRs) in the polymorphic self-assembly of recombinant CP, which results in three distinct VLP architectures. Based on their high-resolution cryo-EM models, we successfully engineered variants with increased application potential for their use in bionanotechnology, produced via deletions at the C- and/or N-terminus, as well as carrying single-site mutations in distinct CP regions. With a wide range of variants combined with their biochemical, biophysical and structural analysis we have shown that we can control the shape, size, ability to encapsidate RNA, symmetry, stability and surface functionalization of PVY CP-derived nanoparticles. We have determined the structure of such nanoparticles, ranging from polymorphic to monomorphic filaments, rings, cubes, spherical particles and more, all with precisely defined architectural parameters. Furthermore, we have shown that we can prevent the self-assembly of CP in bacteria and allowed control of the nanoparticle formation process in space and time.

In summary, we have unraveled the vast structural landscape available to PVY CP and that the properties of the assembled nanoparticles can be tailored for specific purposes. Our results not only provide novel possibilities for the production of biodegradable nanoparticles, but could also advance future studies on the polymorphism of CP in a biological context.

Keywords: potato virus Y, coat protein, virus-like particle, self-assembly, polymorphism, cryo-EM, engineering.