

Abstract

Tobacco mosaic virus (TMV) is one of the most investigated viruses and its attributes and structure are therefore well-known. In this work, we have chosen TMV as a biotemplate for the adjustable-length particles production in plants. The viral RNA and coat protein of TMV self-assemble into particles under physiological conditions. The particle length depends on the length of packaged RNA. The encapsidation signal that is necessary for preferential viral RNA packaging by coat protein disks is known and characterized since the 1980's.

In this work, we have proposed a two-component system based on a *Nicotiana benthamiana* plants infection with packaging competent defective RNA (dRNA) and a helper virus RNA which provides all the components necessary for dRNA replication and packaging. The encapsidation signal in the helper virus sequence was removed to avoid formation of particles of incorrect length. Some of our helper viruses contained a coat protein with modified region of the particle's inner channel. This modification should allow specific binding of metal atoms within the core of the rod shaped particle. Several variants of dRNA and helper viruses were prepared to identify individual areas important for the replication, encapsidation and nanoparticle stability. We focused on the particle formation ability in plants, identification of RNA contained in the particles and distribution of particle lengths in the population.

We have surprisingly discovered that the encapsidation signal elimination from the viral genome does not affect the particle assembly, only the virus sensitivity to higher temperatures. This interesting discovery had an impact on our proposed system when the helper virus particles substantially prevailed over the target-length particles. We constructed a virus with a metal-binding peptide that is exposed in the particle's inner channel. This virus retains its infectivity and the ability to create particles. The length distribution of particles, however, revealed that the specificity of such a modified coat protein for the viral RNA was impaired. The individual components of the proposed system seem to be very promising and could have broad potential use both in scientific research and in biotechnology applications. Further research will be necessary to use the dRNA vectors and modify the system components to ensure the uniform particle length. Further research might also improve our understanding of the mechanisms of viral RNA encapsidation in plants.

Key Words: Tobacco mosaic virus, TMV, nanoparticles, origin of assembly, encapsidation signal