

Truncated forms of viral VP2 proteins fused to EGFP assemble into fluorescent parvovirus-like particles

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[摘要] 13 December 2006. Fluorescence correlation spectroscopy (FCS) monitors random movements of fluorescent molecules in solution, giving information about the number and the size of for example nano-particles. The canine parvovirus VP2 structural protein as well as N-terminal deletion mutants of VP2 (14-, 23-, and 40 amino acids) were fused to the C-terminus of the enhanced green fluorescent protein (EGFP).

[关键词] VP2 protein; fluorescent parvovirus

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Fluorescence correlation spectroscopy (FCS) monitors random movements of fluorescent molecules in solution, giving information about the number and the size of for example nano-particles. The canine parvovirus VP2 structural protein as well as N-terminal deletion mutants of VP2 (14-, 23-, and 40 amino acids) were fused to the C-

terminus of the enhanced green fluorescent protein (EGFP). The proteins were produced in insect cells, purified, and analyzed by western blotting, confocal and electron microscopy as well as FCS. The non-

truncated form, EGFP-

VP2, diffused with a hydrodynamic radius of 17 nm, whereas the fluorescent mutants truncated by 14, 23 and 40 amino acids showed hydrodynamic radii of 7, 20 and 14 nm, respectively. These results show that the non-

truncated EGFP:VP2 fusion protein and the EGFP:VP2 constructs truncated by 23 and by as much as 40 amino acids were able to form virus-like particles (VLPs). The fluorescent VLP, harbouring VP2 truncated by 23 amino acids, showed a somewhat larger hydrodynamic radius compared to the non-

truncated EGFP:VP2. In contrast, the construct containing EGFP-

VP2 truncated by 14 amino acids was not able to assemble into VLP assembling structures. Formation of capsid structures was confirmed by confocal and electron microscopy. The number of fluorescent fusion protein molecules present within the different VLPs was determined by FCS. In conclusion, FCS provides a novel strategy to analyze virus assembly and give information about the size of the particles.

