

Crystallization and X-ray diffraction of virus-like particles of a grouper betanodavirus

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The grouper is a high-value fish in seafood market, whereas grouper nervous necrosis virus (GNNV) causes near 100% mortality in larvae and juveniles. We expressed GNNV virus-like particles in *Escherichia coli* and crystallized it by sitting-drop vapor diffusion method. The crystals grew to the size of 0.22-0.27 mm in one week and diffracted by X-rays to 7.5 Å resolution. The data were indexed in a primitive rhombohedral crystal system. Preliminary processing of the DGNNV VLPs diffracting data suggests that the crystal belong to space group $R32$ with $a = b = 353.00$ Å, $c = 800.40$ Å, $\alpha = \beta = 90^\circ$, $\gamma = 120^\circ$. A total of 299 images were collected from a single VLP crystal by 60° diffracting range, 0.2 oscillation angle, and 10s exposure. 60,035 observed reflections were reduced to 23,268 unique reflections with an overall R_{merge} of 18.2% and a completeness of 93.2%. Self-rotation function maps were calculated using the program POLARRFN from the CCP4 suite; spherical angles of $\kappa = 72^\circ$, 120° , and 180° were for fivefold, threefold and twofold symmetry axes. The 3D atomic model of the asymmetric subunit were predicted. This is the first attempt to solve betanodavirus structure in addition to electron cryomicroscopy.