Plant viral particles as nanoscaffolds for controlled positioning of enzymes on solid supports

**Introduction**

Mimicking metabolon formation in vitro on scaffolds is a promising way to study known metabolomic pathways and to create artificial cascade reactions for efficient in vitro synthesis. The aim of this research is to assemble an enzymatic cascade on the surface of virus particles for the synthesis of piceid, a resveratrol derivative, which may have potential pharmacological applications.

**Approach**

To achieve an enzyme-nano carrier (ENC) that allows placement of enzymes within close proximity, a novel approach was developed, utilizing the immunoglobulin (IgG)-binding peptide Z33. This method was tested by the non-covalent linkage of fluorescent proteins to the viral surface. Imaging was done using immunolabelling and correlative microscopy studies.

**Immunolabelling**

Virus particles coated with Z33mCFP were labeled via antibodies with attached gold colloids of 10 nm and observed by Transmission Electron Microscopy.

**Correlative microscopy**

A. **Light microscopy** ZYMV particles were adsorbed on copper grids and coated with anti-ZYMV antibodies and Z33mYFP. Samples were imaged by fluorescence.

B. **Transmission electron microscopy (TEM)** The same grids were subsequently stained and observed via TEM. The same virus particle pattern was observed.

**Conclusions and future research**

A novel approach to link proteins to the surface of virus particles has been demonstrated for using viruses as enzyme nano-carriers (ENCs). Genetic constructs are underway to fuse 4-coumarate-CoA ligase 2 (4CL2) and Stilbene synthase (STS) with Z33. An artificial metabolon consisting of enzymes of the piceid pathway will be assembled on virus particles.

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