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Modification of grapevine virus A genome for vector development

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The development of new vectors based on plant viruses does not discontinue for the goal of creation a more efficient vector with high expression of heterologous proteins. We attempted to develop such vector based on genome of grapevine virus A (GVA). For creation of this viral vector we inserted gene encoding coat protein of Apple chlorotic leafspot virus (ACLSV) before ORF4 of GVA. PCASS vector carrying the complete genome of the grapevine virus A (pCASSgva) was used for creation a viral vector based on GVA. The viral genome was modified by introducing a CP gene of ACLSV before ORF 4 within restriction sites XmaI and XbaI. The overlapping region of 3'- terminus ORF3 and 5'- terminus of ORF4 was intact. The CP gene of ACLSV was placed under control of CP subgenomic promoter of GVA. The modified viral genome was subcloned into a pCambia 2300 binary vector. The expression of the CP of ACLSV in agroinfiltrated *N.benthamiana* leaves after 3-4

days of infection was confirmed by using western blotting. Agroinfiltration of transgenic plants carrying CP of GVA was not successful, we assume due RNA-silencing. It will be investigated the expression level of the viral vector and its usefulness as a vector for the expression of avian influenza hemagglutinin.

Speaker Biography

Gritsenko D A is a PhD- student at Kazakh National University named after al- Farabi. She performs her diploma work at Institute of Plant Biology and Biotechnology. The title of diploma is "Development of Viral Vector for Heterologous Protein Expression in Plants". She developed 2 vectors based on genome of Grapevine virus A by using main strategies for vector engineering such as "deconstructed virus" and "full virus". Currently, these vectors were investigated for successful expression of eGFP and coat protein of Apple chlorotic leafspot virus. Moreover, she developed transgenic plants carrying coat protein of GVA for increasing of target protein yield since GVA cannot move between cells in non-encapsidated form.

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