Analysis of the proteome of a Modified Vaccinia virus Ankara-based vaccine provides insight into the effects of transgenic modification on virus

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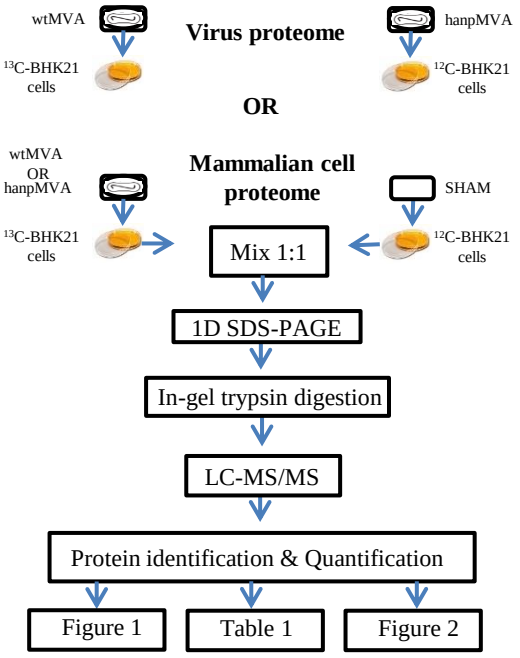
Introduction

- Modified Vaccinia virus Ankara (MVA) is an attenuated strain of Vaccinia virus
- Vaccinia virus and MVA were used as vaccines in the 1980s for the eradication of smallpox
- Since smallpox eradication MVA has further been developed as a live virus vaccine vector for verterinary and human diseases
- The verterinary MVA-based vaccines have had some success, but the human vaccines are still under clinical trials
- Several of the failures at clinical trials are attributed to low efficacy as well as issues of safety
- MVA vaccine vectors are known as good expressors of inserted transgenes, but information is lacking on the expression of indigenous virus proteins following transgene insertion
- The aim of this work was to evaluate the global protein expression of wild type MVA (wtMVA) and a vaccine vector, hanpMVA containing haemaglutinin (ha) and nucleoprotein (np) cDNA inserts, during infection of BHK-21 cells

Materials & Methods

Scheme describes the Stable Isotope Labelling of Amino Acid in Cell Culture (SILAC) for determination of changes in virus and mammalian cell proteomes



Results

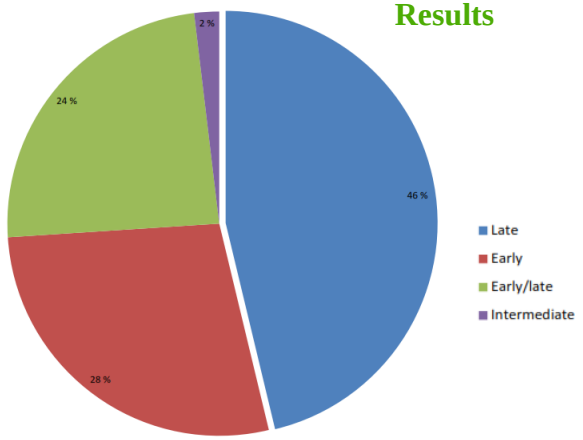


Figure 1: Modulated proteins of virus classified according to temporal expression. Relative to wild type (wtMVA) strain, 54 proteins of hanpMVA were identified among the differentially expressed proteins (37 up-regulated; 17 down-regulated). The proteins were classified according to their time of appearance (temporal expression) during the virus life cycle, i.e. Late, Early, Early/Late & Intermediate proteins

Table 1: Protein post-translational modifications identified in wtMVA & hanpMVA

Protein	Modification		Peptide Sequence	Site in Pepti	Ion Score	Exp Value
	wtMVA	hanpMVA				
A36R	ND	Phosphorylation	SNPFITELNNK	S1	54,23	0,0003708
B19R	ND	Phosphorylation	YNCYVHYDDVR	Y1	40,69	0,0043764
F7L	ND	Phosphorylation	SDINTLDIK	T5	29,53	0,0728191
I1L	ND	Phosphorylation	SSIKDSMYVIPDELIDVLK	S1	32,65	0,0789886
G8R	ND	Acetylation	YEEKCCGR	K4	31,77	0,0286733
I1L	ND	Acetylation	IPVDLVKSSFVK	K7	58,75	2,88E-05
E3L	ND	Acetylation	SKIYIDER	K2	43,06	0,0028645
I1L	ND	Acetylation	DSMYVIPDELIDVLK	K15	22,02	0,7304319
F17R	ND	Acetylation	ASCVLKVDKPSSPACER	K6	52,86	0,0008843
I1L	ND	Acetylation	DSMYVIPDELIDVLKTR	K15	60,02	9,302E-05
F16L	ND	Acetylation	NLEFATWKDVIQNDEIDALVFYR	K8	16,47	4,5174954
A17L	ND	Acetylation	YYNMLDDFSAGAGVLDKDLFTEEQQQSFMPK	K17	27,64	0,6162568
B8R	ND	Methylation	ELILYDKDEIR	K7	33,14	0,0348922
F16L	ND	Methylation	CASQLDNVCTEMNK	K14	26,04	0,339729
J5L	ND	Methylation	LPYYCWYEPCK	K11	33,67	0,0563767
A35R	Methylatio	ND	FSIQDVK	K7	36	0,0102292
ND = Not detected						

Conclusion: Insertion of haemaglutinin and nucleoprotein resulted in differential expression and post-translational modification of hanpMVA protein relative to wtMVA. This led to differences in modulated host cell (BHK21 cells) proteins in response to infections by the virus strains

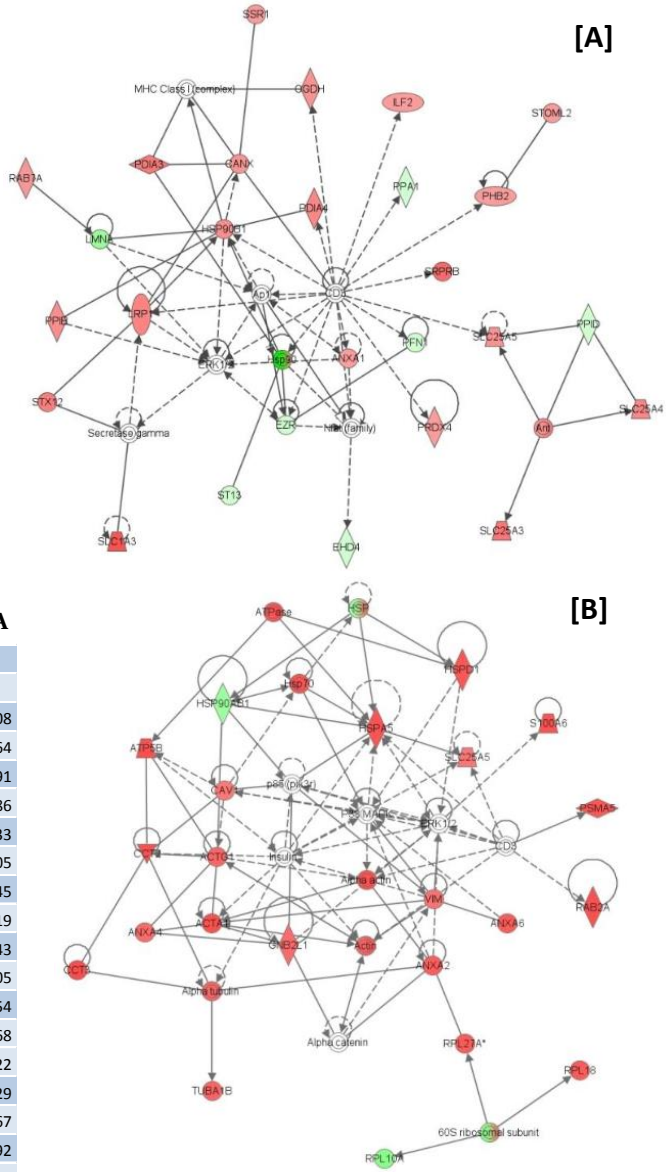


Figure 2: Disrupted pathways of protein networks of BHK-21 in response to wtMVA [A] & hanpMVA [B] infections. Green = up-regulated; Red = down-regulated.

References: Gómez CE *et al* (2011). Curr Gene Ther 11(3):189-217