IOCBSERVICE DAYS 2007

Research-Service Groups / Service Groups / Core Facilities

Virology Research Service Group

Jan Weber





Group's mission:

To aid IOCB drug discovery effort and to help research groups with projects involving work with viruses BSL3: room C.1.07 and C.1.08 BSL2: room C1.06 Offices: C.1.33 and C.1.34

Virus bank

- RNA viruses
 - ssRNA-RT
 - **HIV-1**, HIV-2
 - (-)ssRNA
 - Influenza A H1N1, H3N2
 - Influenza B
 - Hepatitis delta virus
 - Rift Valley fever virus
 - Respiratory syncytial virus
 - dsRNA
 - Reovirus type 3
- DNA viruses
 - Herpes simplex virus type 1 and 2
 - Hepatitis B virus
 - Mpox (monkeypox virus)

- (+)ssRNA
 - Coxsackie B3 virus
 - Dengue virus type 1, 2, 3, 4
 - Aichivirus A
 - Zika virus
 - Human coronavirus 229E
 - SARS-CoV-2
 - Human coronavirus OC43
 - Murine norovirus
 - Human rhinovirus 1A, 14, 16, 17, 19

- African swine fever virus
- Human adenovirus B type 3

Screening of IOCB compound library

- >10,000 compounds
- 384-well format, 8 viruses
- Initial screening in triplicate at 50 or 25 μ M concentration
- Cytotoxicity in MT-4 cells and Vero E6 cells
- Results expressed as % of inhibition are deposited into database
- Hits verification, EC_{50} and CC_{50} deposited into database



Michala Zgarbová



Eva Tikalová



Jana Trylčová

Additional service

- Antiviral activity also for nanoparticles, peptides, proteins, siRNA, polymers, coatings, textiles, etc.
- Large scale virus and infected cells preparation
- Isolation and amplification of genetic material

• ELISA (HIV, HBV, SARS-CoV-2)



Jan Hodek



Jana Trylčová



Lucie Ulrychová



Michala Zgarbová



Marcela Pávová



Jan Hodek

Additional service – cont'd

- Immunofluorescence techniques (HIV, HBV, Dengue, HSV-1, Influenza, CVB3, SARS-CoV-2)
- Influenza minigenome assay
- FACS of infected cells



Václav Janovec

- Reverse transcriptase activity assays
- Time-of-addition assay
- Selection for resistance





Marcela Pávová

Jan Hodek



• Transfection / silencing experiments

Barbora Lubyová

Eva Tikalová

Michala Zgarbová

Marcela Pávová

SARS-CoV-2

- 16 primary isolates (alpha, beta, gamma, delta, omicron)
- Cell line models
 - Vero E6 (kidney, monkey), Huh7 (liver), Caco-2 (colon), Calu-3 (lung), A549/TMPRSS2/ACE2 (lung)
- Methods
 - CPE based assay (XTT readout), Immunofluorescence assay (nucleocapsid antibody), RNA reduction assay (qRT-PCR), plaque assay, ELISA (spike Ab)



How to use our services

- Leave everything on us
- I have compounds: put them in IOCB library
- Other antiviral determinations
 - High MW compounds
 - siRNA
 - Nanoparticles
- Pilot experiments
- Short to medium projects Requests/consultations:
 contact Jan Weber, I. 420 jan.weber@uochb.cas.cz

- I want to do it myself
- Time and resources extensive projects
- To learn something new
- To wear last BSL3 fashion



Join BSL3 user team

How to become a BSL3 user?

- Initial consultations (Jan Weber, I. 420)
- Contact HR / medical examination / valid HBV vaccination
- Safety training: guidelines, safety precautions, training in PPE



Jan Weber

• Provisional access (can enter with an experienced BSL3 user)



Barbora Lubyová



Marcela Pávová



Jan Hodek



Michala Zgarbová



Eva

Tikalová



Jana Trylčová

 After demonstrating good and safe lab practice - full access (can enter alone)

Using the BSL3 facility

- IOCB covers basic supplies (PPE, plastics, tips, media)
- Available equipment for BSL3 users



6 safety cabinets



incubators



Benchtop ultracentrifuge



Plate reader Abs, Lum, Fluo



Fluorescent microscope



96-well plate FACS Guava



Cell sorter Sony MA900

Future development

- Confocal microscope in BSL3 (summer 2023)
 - For super resolution of infection in live cells
 - Petro Khoroshyy (Microscopy core)
- New pipetting robot for 384-well plates (summer 2023)
- Compound library screening (2023)
 - Mpox (monkeypox virus)
 - Respiratory syncytial virus

Thank you for your attention. Questions?