Arenaviruses

- **Arenaviruses Include Clinically Important Human Pathogens:**
  - **Lassa Virus (Lassa fever disease)**
  - **South American Hemorrhagic Fever Virus**

- **The Prototypic Arenavirus LCMV: A Rosetta Stone For The Study Of Virus-Host Interactions And Associated Diseases**

- **LCMV Is Likely A Neglected Human Pathogen:**
  - **High Sero-Prevalence of LCMV In Humans**
  - **Contribution To Congenital Disorders**
  - **Transplant-Associated Disease**

Lassa Virus (LASV) And Its Impact In Public Health

- **LASV causes Lassa Fever, a hemorrhagic fever disease endemic to a vast geographic region with a large (180 million) population at risk of infection**

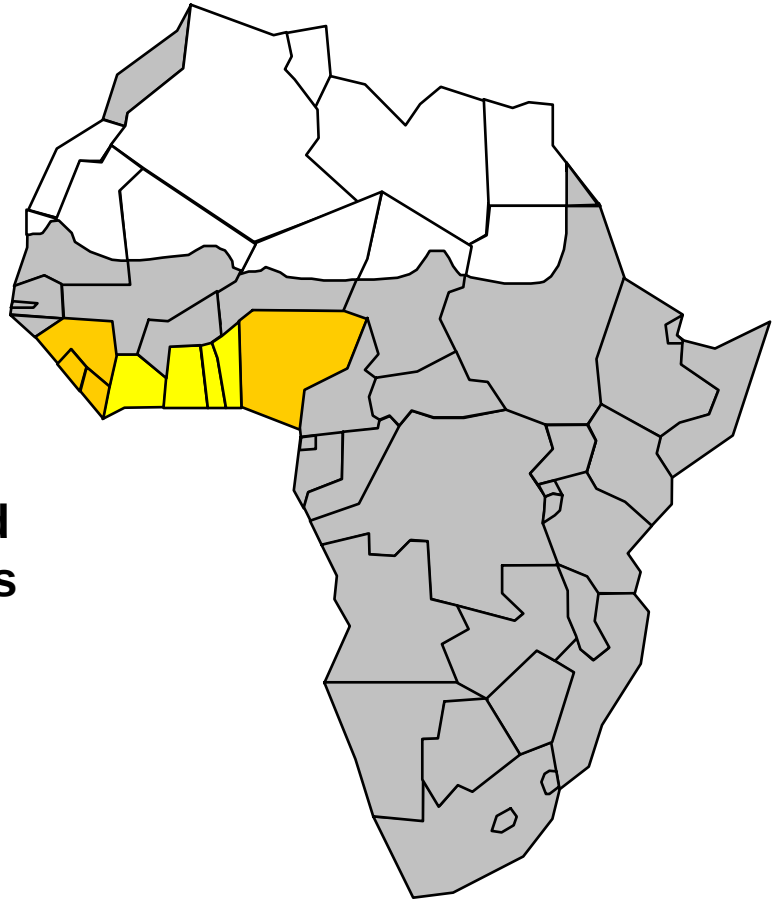
- **Over 500,000 infections per year associated with high morbidity and significant mortality (> 5,000 deaths/yr)**

- **Unfeasibility to control and eradicate LASV natural reservoir (Mastomys)**

- **Lack of licensed vaccines and current therapy limited to the use of the ribavirin, which is only partially effective and associated with significant side effects**

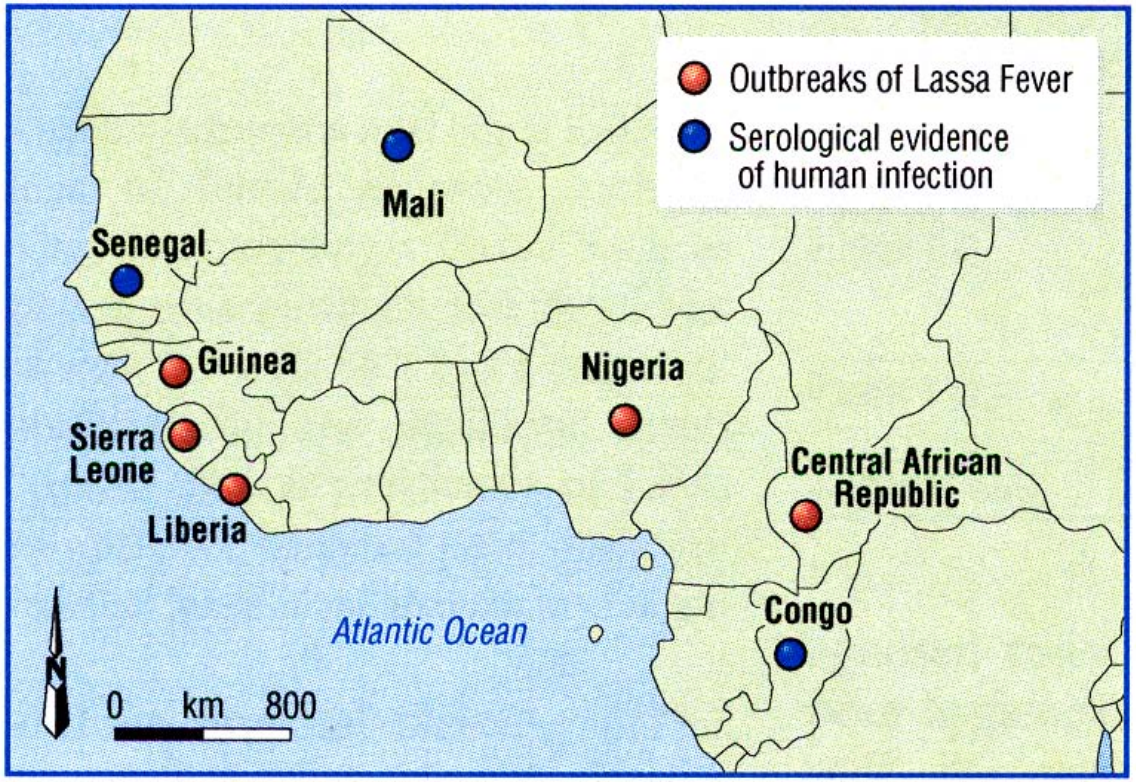
- **Travel to and from endemic areas has resulted in importation of LF cases in metropolitan areas in non-endemic areas**

- **LASV poses also a biodefense concern**



High Prevalence Of LASV In Western Africa

In some regions of Western Africa > 20% of adults are seropositive for Lassa virus, making Lassa fever a leading cause of death among adults.



Adults seropositive for Lassa virus:

Nigeria: 21.3%

Sierra Leone: 4-53%

Guinea: 8-52%

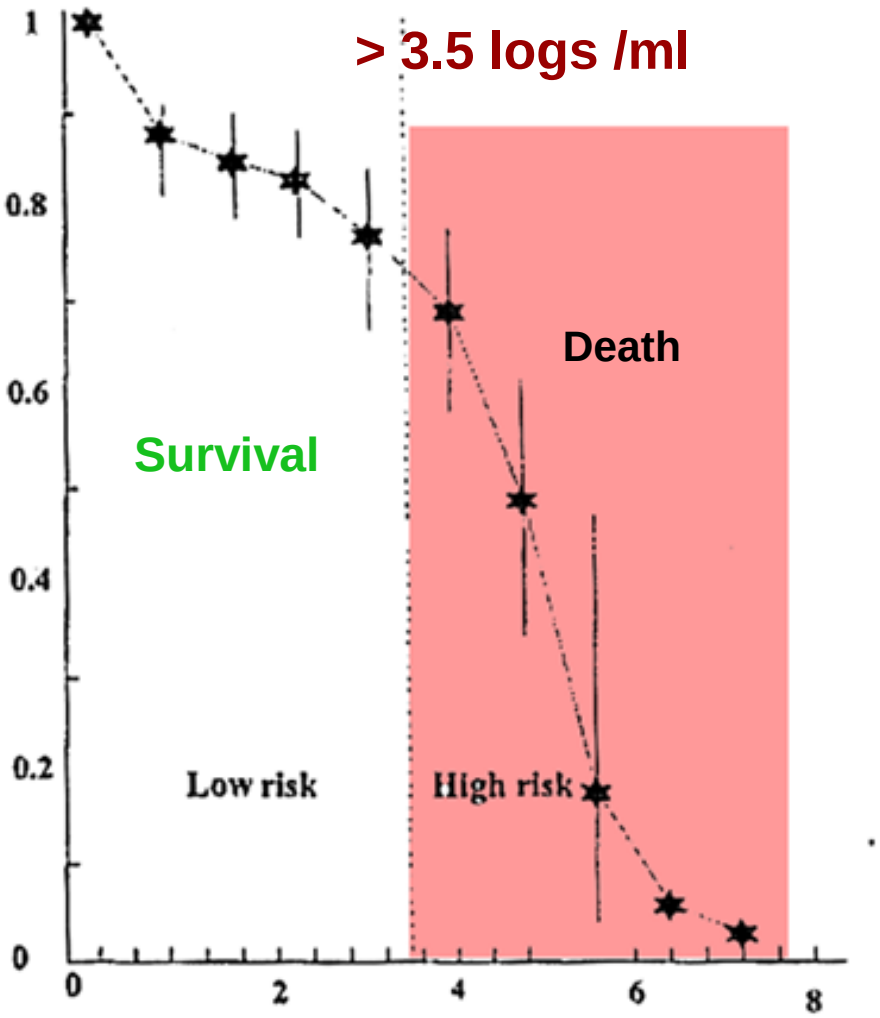
Clinical Disease: Lassa Fever

- **Incubation period 15-21 days**
- **Early symptoms are unspecific: fever and general malaise including: neurological and gastrointestinal symptoms**
- **20% of patients develop facial edema and mucosal bleeding**
- **Coagulation abnormalities and increased vascular permeability**
- **Hemorrhagic shock syndrome**
- **Death from multi-organ failure**
- **Convalescent patients suffer frequently from deafness**

High Viremia In Lassa Fever Patients Correlates With Fatal Outcome

Factors associated with poor prognosis:

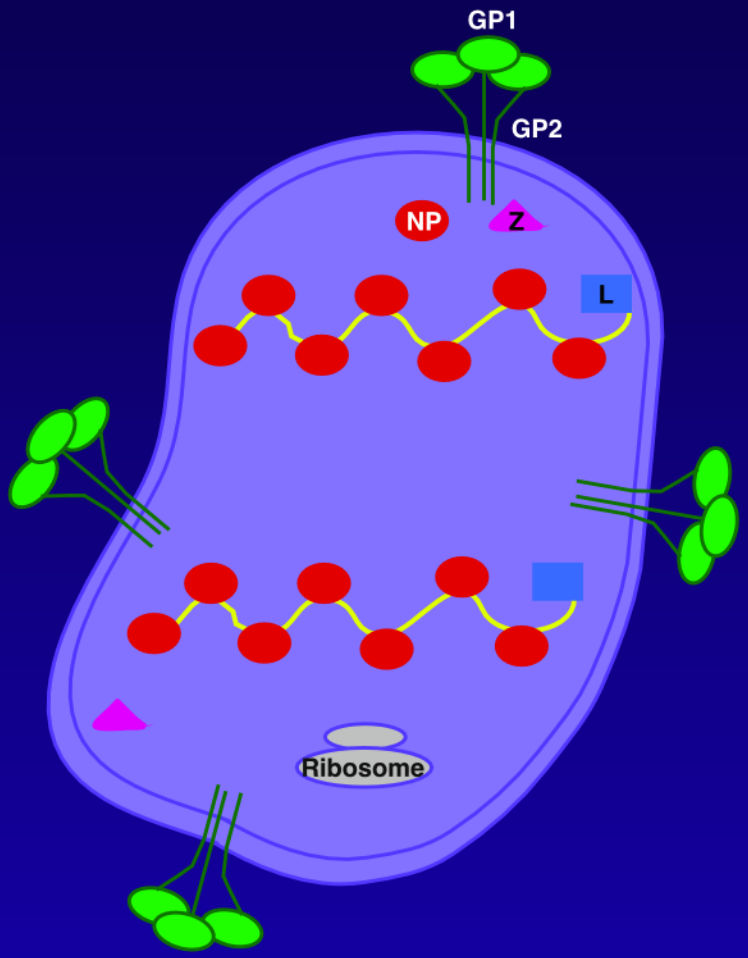
High viremia
Facial edema
Lung edema
Bleeding



Titers of LFV in surviving and dying patients

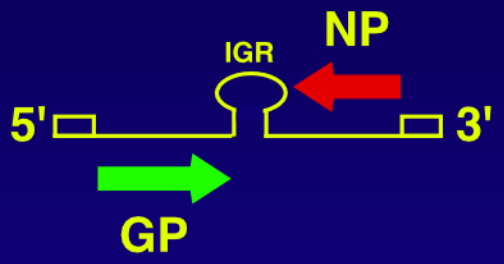
LCMV

Virion Structure



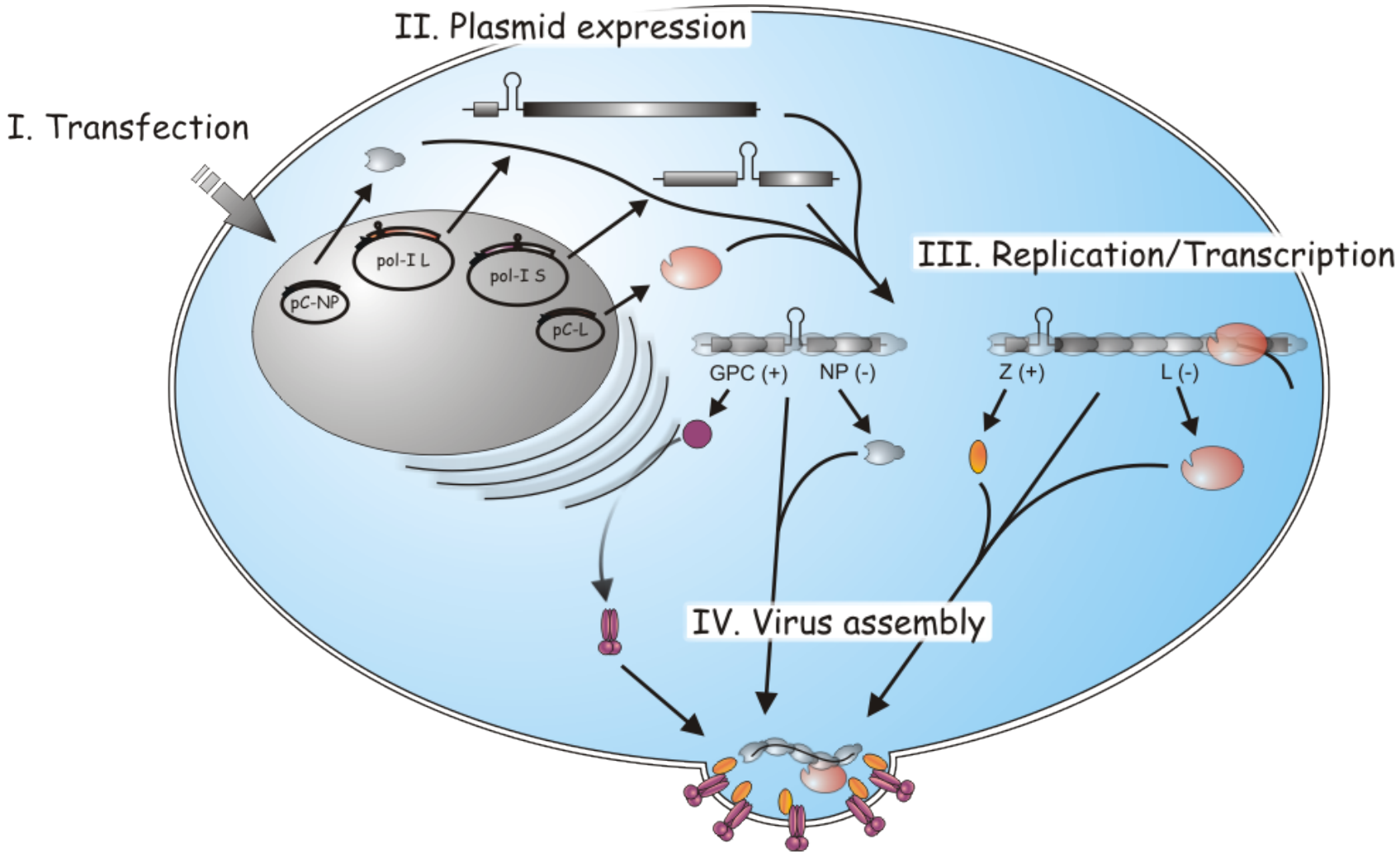
Genome Organization

S RNA (3.5 kb)

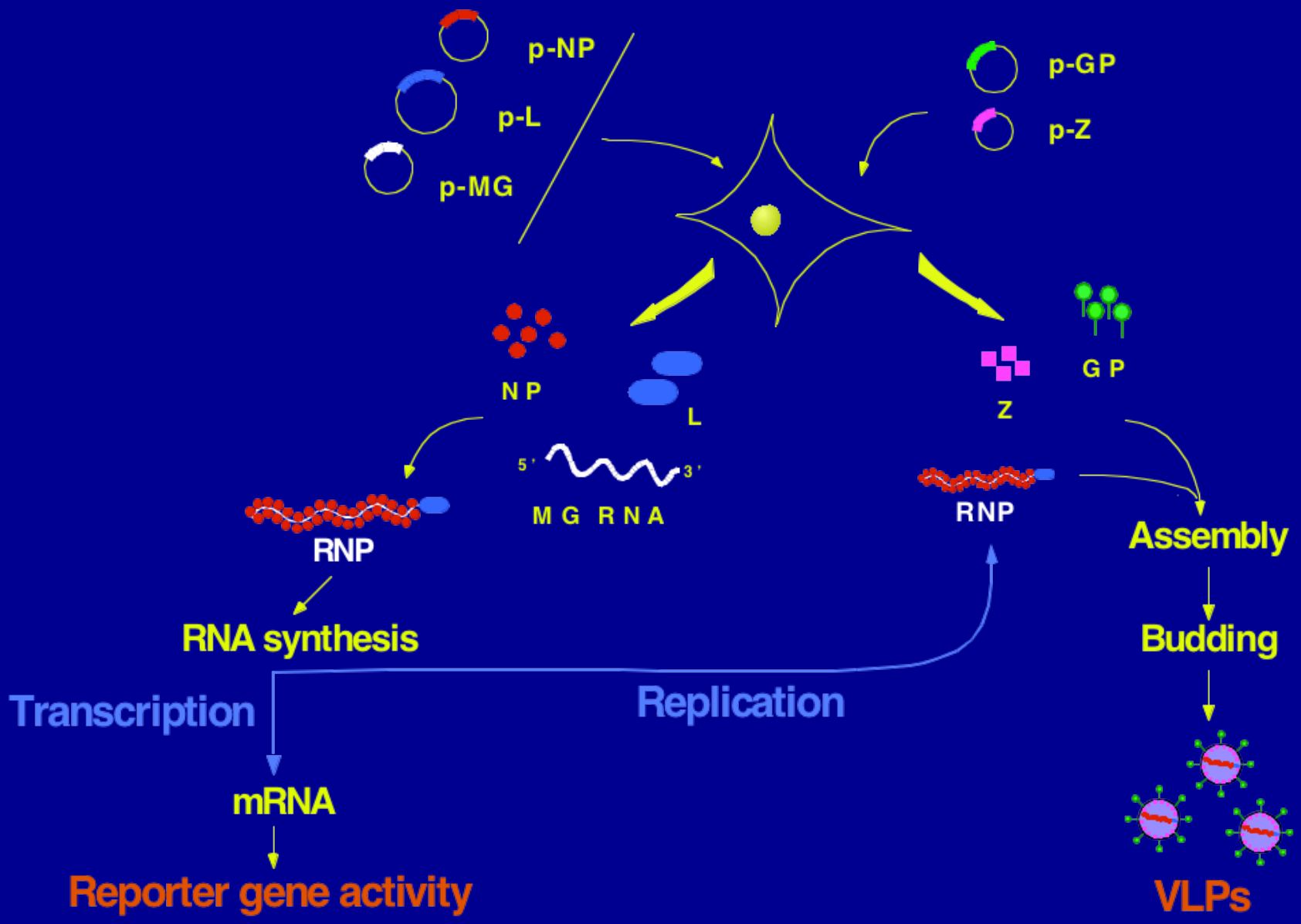


L RNA (7.2 kb)

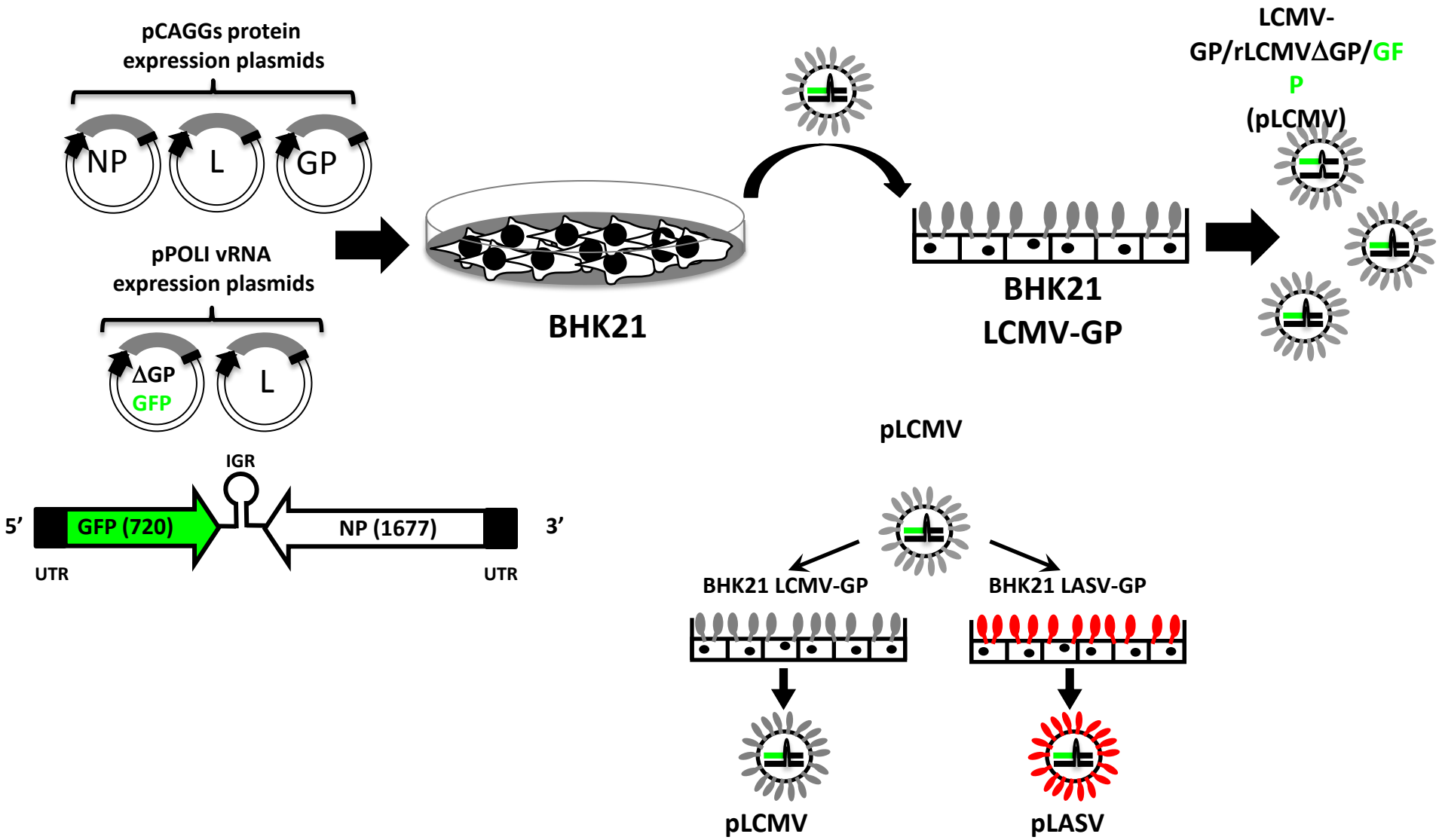
Arenavirus Reverse Genetics Provides Us With A Novel And Powerful Approach To Study Arenavirus-Host Interactions

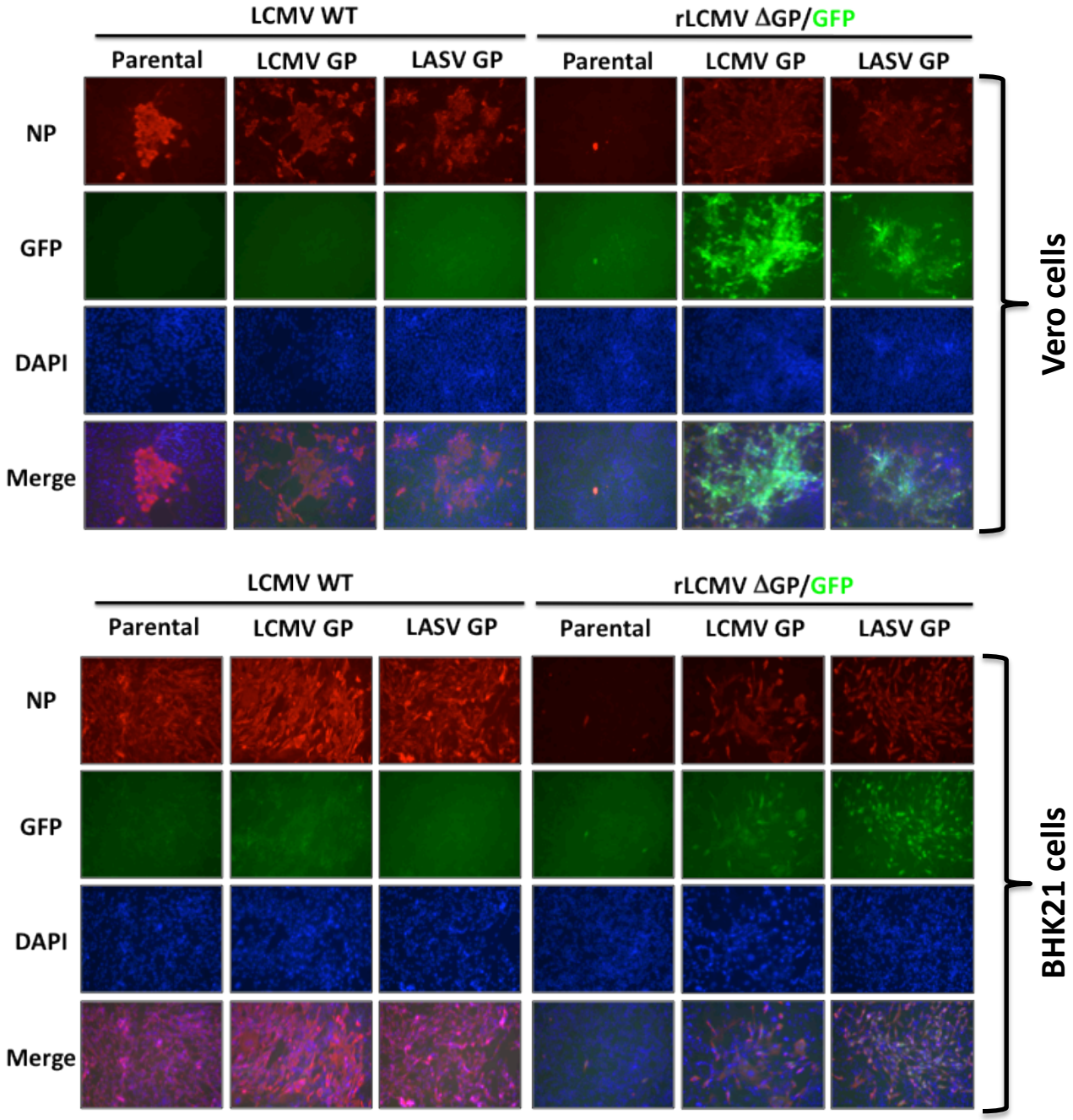


LCMV Minigenome Rescue Assay

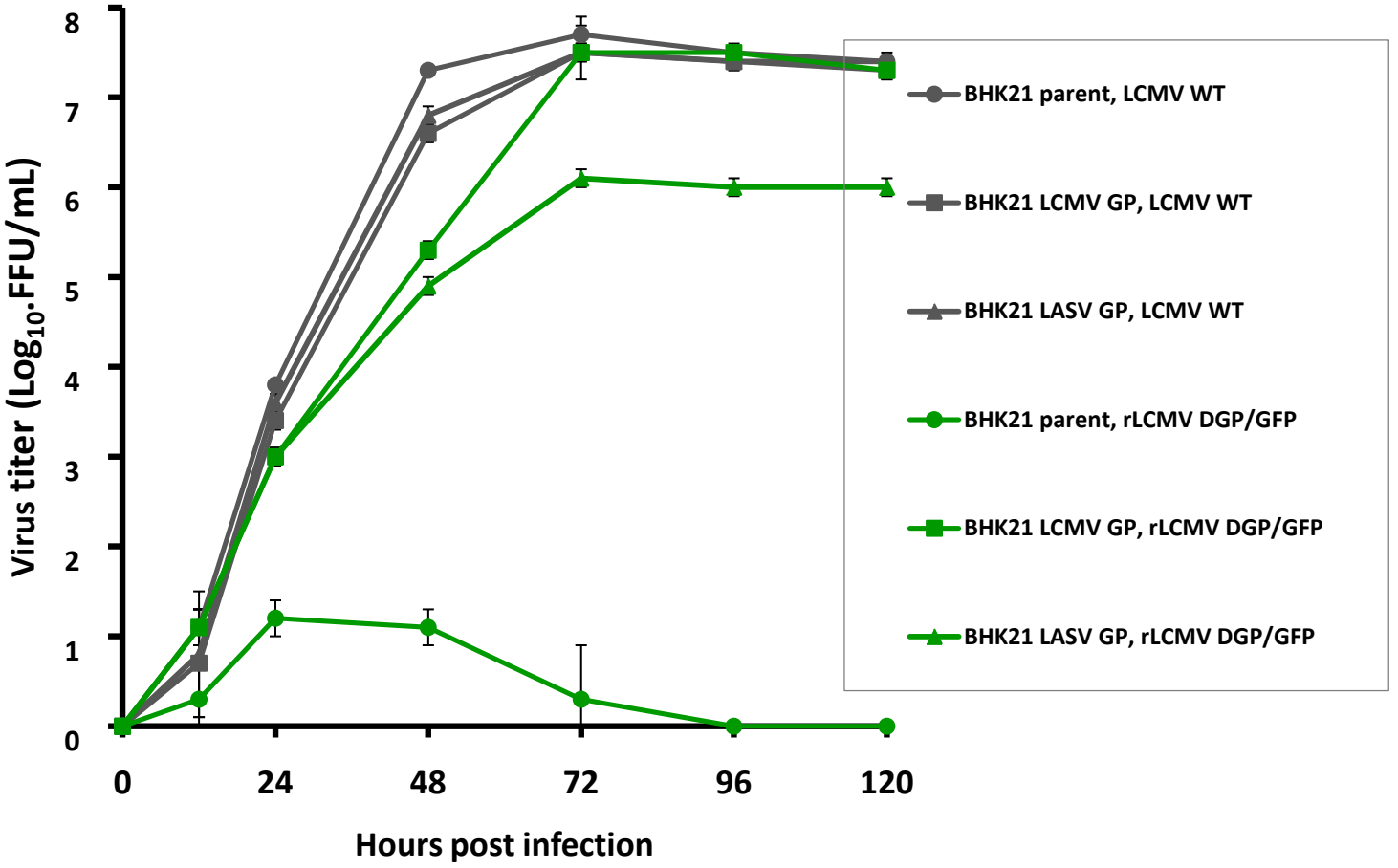


Generation Of Single-Cycle Infectious LCMV

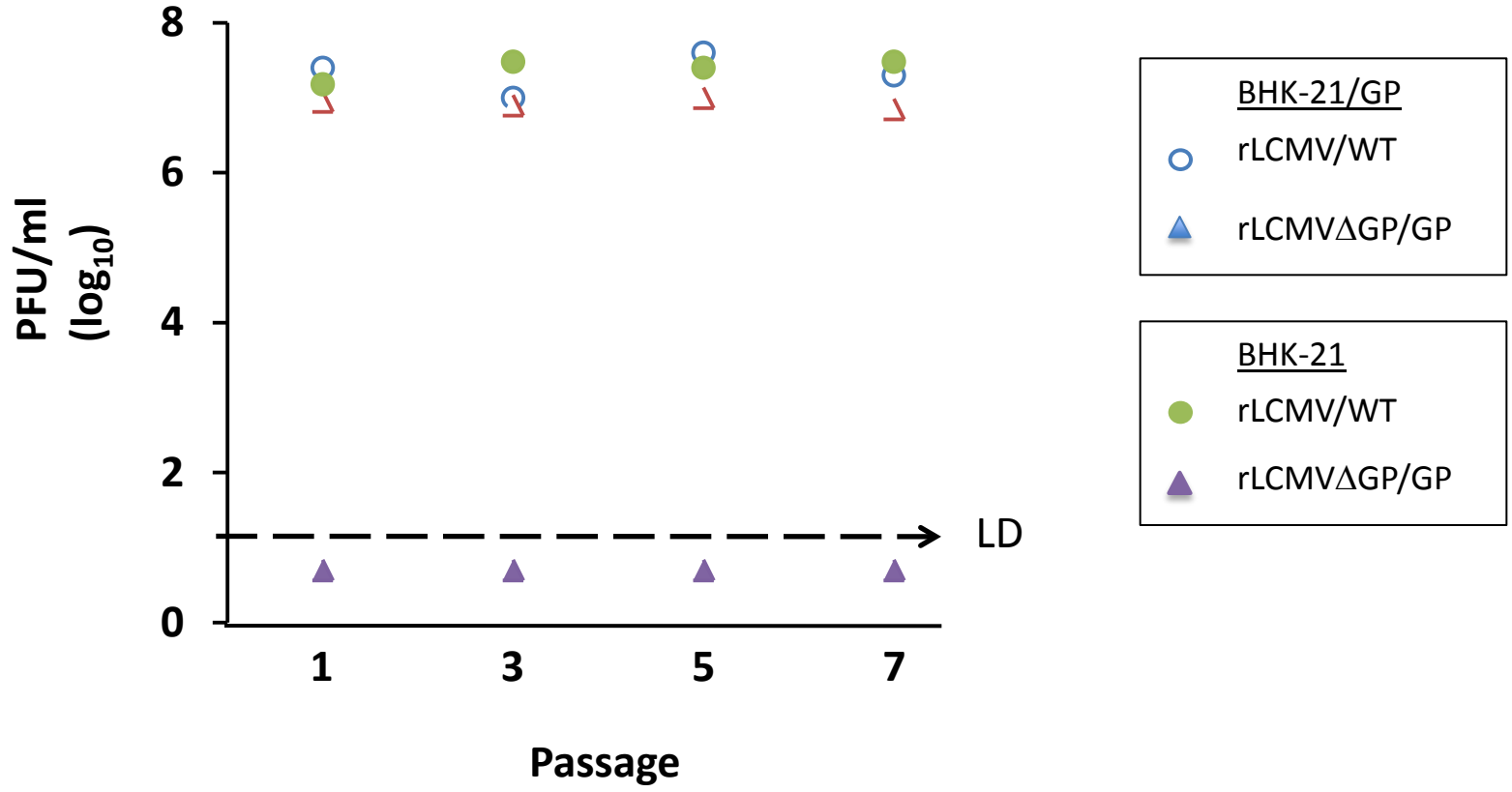



LCMV WT
rLCMV ΔGP/GFP
Parental
LCMV GP
LASV GP
Parental
LCMV GP
LASV GP
NP
GFP
DAPI
Merge
Vero cells
LCMV WT
rLCMV ΔGP/GFP
Parental
LCMV GP
LASV GP
Parental
LCMV GP
LASV GP
NP
GFP
DAPI
Merge
BHK21 cells

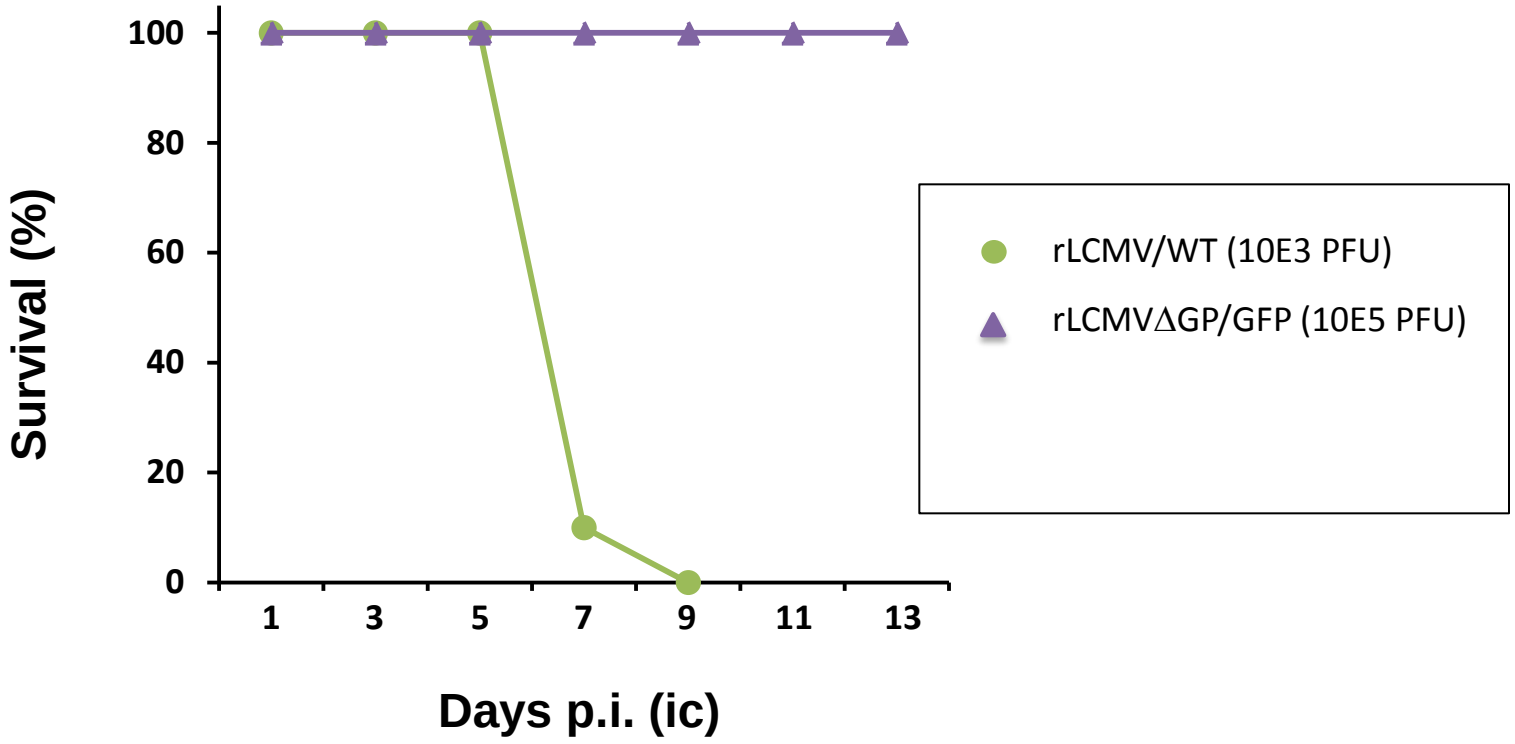
Growth Properties Of rLCMVΔGP/GFP



rLCMVΔGP/GFP Propagation Is Restricted To Cells Expressing GP



rLCMVΔGP/GFP Is Non-Virulent In Vivo



Steps In The Generation Of rLASVΔGP/GFP

Step 1: Generate pol1 expression plasmids to direct intracellular synthesis of LASV L and SΔGP/GFP genome RNA species (BSL2)

Step 2: Generate and functionally test (minigenome assay) expression plasmids for LASV NP and L (BSL2)

Step 3: Rescue of rLASVΔGP/GFP (BSL4)

Step 4: Characterization of biosafety properties of of rLASVΔGP/GFP (BSL4)

1) Restricted growth to cells expressing GP
2) Virulence in animal models of LASV induced disease

Some General Biosafety Procedures

- Dedicate space for experiments involving the use of LASV pol1 L and pol1SDGP/GFP plasmid
- Complete LASV S genome will not be generated as part of this project
- Cells used for these studies will undergo strict QC procedures to rule out possible contamination with LCMV
- Only appropriately trained laboratory personnel will participate in these studies