LASSA FEVER

Background

- Caused by the Old World arenavirus of the same name:
  - Spherical or pleomorphic virions, generally 110-130 nm in diameter
  - Genome contains single-stranded RNA with 2 segments (both ambisense)
  - Size: 11 kbp
  - Viral particles contain host ribosomes, which appear as dense granules 20–25 nm in diameter and give viruses "sandy" appearance
  - Lipid envelope
  - Distinct club-shaped or spike projections on viral envelope composed of glycoproteins
- Incubation period: 5-16 days
- Lethal dose:
- Prodrome: Illness beings gradually with fever, weakness, generalized malaise.
- Mode of infection/transmission:
  - Predominantly airborne through virus-containing aerosols of rodent excreta
  - Person-to-person (eg, contact with blood or body fluids)
  - Percutaneous through accidental needlesticks or reuse of injection equipment
  - Possibly person-to-person airborne (in at least one instance, transmission may have occurred in hospital setting from patient with extensive pulmonary involvement)
  - Sexual transmission (virus has been found in semen)
- The severity and clinical symptoms depend on viral virulence, route of exposure, dose and host factors.

Common physiology changes caused by the viral infection:

- Hemorrhagic manifestations - bleeding
- Hepatic involvement – hepatic failure

Pathogenesis:

- Organs/tissues affected: lymphoid tissue (spleen and lymph nodes), liver, adrenal gland, endothelium. A close relationship between the severity of clinical illness and the distribution of infection and organ titers was observed in squirrel monkeys infected with Lassa virus
- Lesions are generally not severe enough to account for terminal shock and death
- Lymphoid tissues are severely damaged > lymphoid depletion
- Defects in coagulation system
- High viremia (large number of viruses in the blood)
- Immunosuppression
- Cells that support replication of the virus: monocytes, macrophages, dendritic cells, endothelial cells, hepatocytes, adrenal cortical cells

Infection mechanism:

- Not completely clear; it is believed to be initiated in the nasopharyngeal mucosa (inhaled)
- Virus gets into the circulation/blood by …
- Entry into host cells: probably by receptor-mediated endocytosis
- Monocytes, macrophages and dendritic cells are the early targets of the virus; endothelial cells are infected later, proximal to death; infected cells are not typically destroyed
- Virtually all tissues become infected
- Death seems to be caused by ineffective/insufficient immune response early > high viral loads > high levels of proinflammatory mediators (cytokines, chemokines) in the late stages > toxic/lethal effects
- No DIC (disseminated intravascular coagulation) in Lassa fever

**Drug Therapy**: Ribavirin (a nucleoside analog) has some activity against Arenaviruses

**VIRUS LOADS**

*Guinea pigs* were inoculated s.c. with a 3.4 log10 PFU dose of Lassa virus (Jahrling 1982)\(^6\). Development of Lassa viremia in lethally infected strain 13, outbred guinea pigs and in surviving outbred guinea pigs was monitored for ~22 days post infection (Figure 1). They also monitored the viremia development in surviving outbread guinea pigs for 180 days after infection (Figure 2).

Plasma viral titers registered based on this work for the guinea pig model:
- Incubation stage: 2 log10 PFU/ml (~average day 4)
- Late stage: 4 log10 PFU/ml (~average of plateau)

Tissue titers for the guinea pig model during the incubation period (day 4) and in the late stage of the disease were recorded based on Figure 4 and Figure 5 (Jahrling 1982)\(^6\), respectively.

<table>
<thead>
<tr>
<th>Disease stage</th>
<th>Serum</th>
<th>Lung</th>
<th>Spleen</th>
<th>Pancreas</th>
<th>Lymph node</th>
<th>Adrenal</th>
<th>Kidney</th>
<th>Salivary gland</th>
<th>Liver</th>
<th>Heart</th>
<th>Brain</th>
</tr>
</thead>
<tbody>
<tr>
<td>Incubation</td>
<td>2</td>
<td>4.5</td>
<td>5.5</td>
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<td>6.8</td>
<td>4.2</td>
<td>1.5</td>
<td>4</td>
<td>3.6</td>
<td>2.5</td>
<td>&lt;0.6</td>
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<tr>
<td>Late</td>
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<td>6.8</td>
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<td>6.4</td>
<td>5.8</td>
<td>5.7</td>
<td>5.5</td>
<td>5.4</td>
<td>5.3</td>
<td>4.2</td>
<td>3.5</td>
</tr>
</tbody>
</table>

In another study, *guinea pigs* were inoculated ip with live virus (no dose provided, Walker et al, 1975)\(^7\). Lassa virus titers in various organs were determined and presented in table 2, using log10 TCID50 units. The Log10 TCID50 values are ~similar to the log10 PFU/g or /ml values from the study of Jahrling et al.\(^6\).
Monkeys
Cynomolgus and rhesus monkeys are comparable in size (~5-7 kg); squirrel monkeys are much smaller (~ 1 kg).

Cynomolgus monkey (Geisbert 2005)

Four cynomolgus macaques (M. fascicularis) were immunized by i.m. injection with a single dose of approximately $2 \times 10^7$ PFU of VSVDG/LVGPC, and two control animals received VSVDG/ZEOBVG (same route and dose). All six animals were subsequently challenged intramuscularly on day 28 postvaccination with a high dose ($1 \times 10^8$ PFU) of Lassa virus. The two control animals (vaccinated with irrelevant VSVDG/ZEOVGP) started to show clinical signs of illness on day 3, when one of the animals had a fever (defined as a temperature over 104°F). By day 10, both control animals developed macular rashes and anorexia, and one animal had severe facial edema, which is prognostic for a poor outcome in humans. These control animals succumbed to the Lassa virus challenge and were euthanized on day 11 and day 13, respectively.

Rhesus monkeys (Callis 1982)

Presence of virus concentrations above serum levels were found in the liver, lung, adrenal gland, pancreas, spleen, kidney, and lymph node. Viral titers were also elevated above serum levels in pleural fluid, but titers were equal to or below serum levels in the brain and spinal cord.

Serum levels in monkeys based on Figure 3B of Geisbert 2005 and Table 1 of Callis 1982:
- stage 1: avg. of day 5 values of non-vaccinated $\approx 3 \log_{10}$ PFU/ml (based on data shown in Figure 3B, Geisbert 2005)
- stage 2: avg. values on day 10: $\approx 6.9 \log_{10}$ PFU/ml

Squirrel monkeys

Four squirrel monkeys were inoculated intramuscularly with Lassa virus (dose not published) and were sacrificed 7, 12, 14, and 28 days later. The 4 animals represented various stages in the infection course: Animal A was well at 7 days after inoculation and was considered to represent the incubation period of infection; animal B was moribund at 12 days; animal C was clinically recovered from a 2-day illness at 14 days; and animal D was convalescent from a 10-day illness at day 28. Organ titers (in $\log_{10}$ of tissue culture median infective dose TCID$_{50}$ per gram or ml of tissue or body fluid) of Lassa virus in squirrel monkeys were extracted from Table 1 (Walker 1975).

Note: $\log_{10}$ TCID$_{50}$ considered to be equivalent of $\log_{10}$ PFU.
Hamster

5- to 6-weekold female Syrian golden hamsters were infected with $10^4$ PFU of pirital virus (PIRV), a rodent model of Lassa fever. Daily mean ± SD virus titers in blood of three hamsters were sampled after experimental infection with PIRV (Pirital virus) – rodent model of Lassa fever. Virus titer in serum was highest at day 5 (7.4 log10 PFU/ml), and decreased slightly to about 6.8-6.9 log 10 PFU/ml during days 6-8 (Figure 2, Sbrana 2006)\textsuperscript{10}.

Human

Data comes from postmortem examination of 21 virologically documented cases of Lassa fever (Walker et al.)\textsuperscript{11}. Average viral loads were calculated on all cases excluding fetuses (Table 1 of Walker et al.).

*Unit for viral load:* *Reciprocal log median tissue culture infectious dilution* = the median of the reciprocal of the dilution (of test material) that produces cytopathic (cell killing) effect (Log\textsubscript{10} TCID\textsubscript{50})

<table>
<thead>
<tr>
<th></th>
<th>N total</th>
<th>Mean</th>
<th>Standard Deviation</th>
<th>Minimum</th>
<th>Maximum</th>
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<tbody>
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<tr>
<td>Liver</td>
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<tr>
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<tr>
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<tr>
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References


