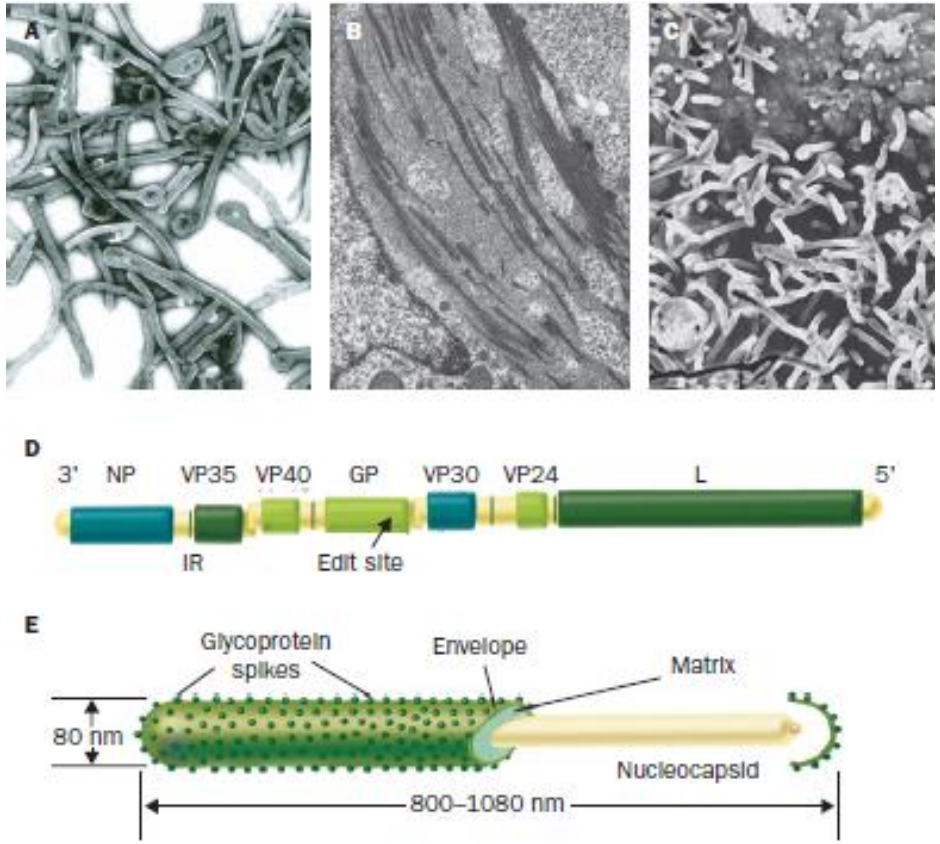
Virology and clinical aspects of EVD

ID and PH Forum

13 Aug 2014

Virology



- Filoviruses
- First recognized in 1967
- Non-segmented, negative-sense, single-stranded RNA viruses
- 5 different species (the ***Zaire***, Sudan, Ivory Coast, Bundibugyo, and Reston agents)

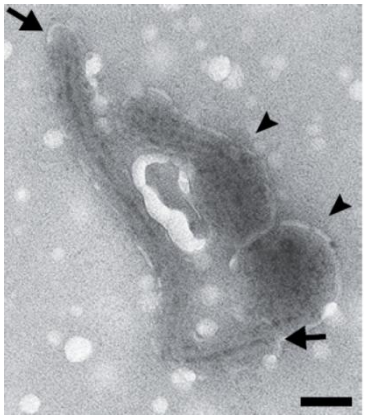
NP and VP35: nucleoproteins
VP30: nucleocapsid
VP24 and VP40: matrix proteins
GP: glycoprotein
L: large protein (RNA dependent RNA polymerase)

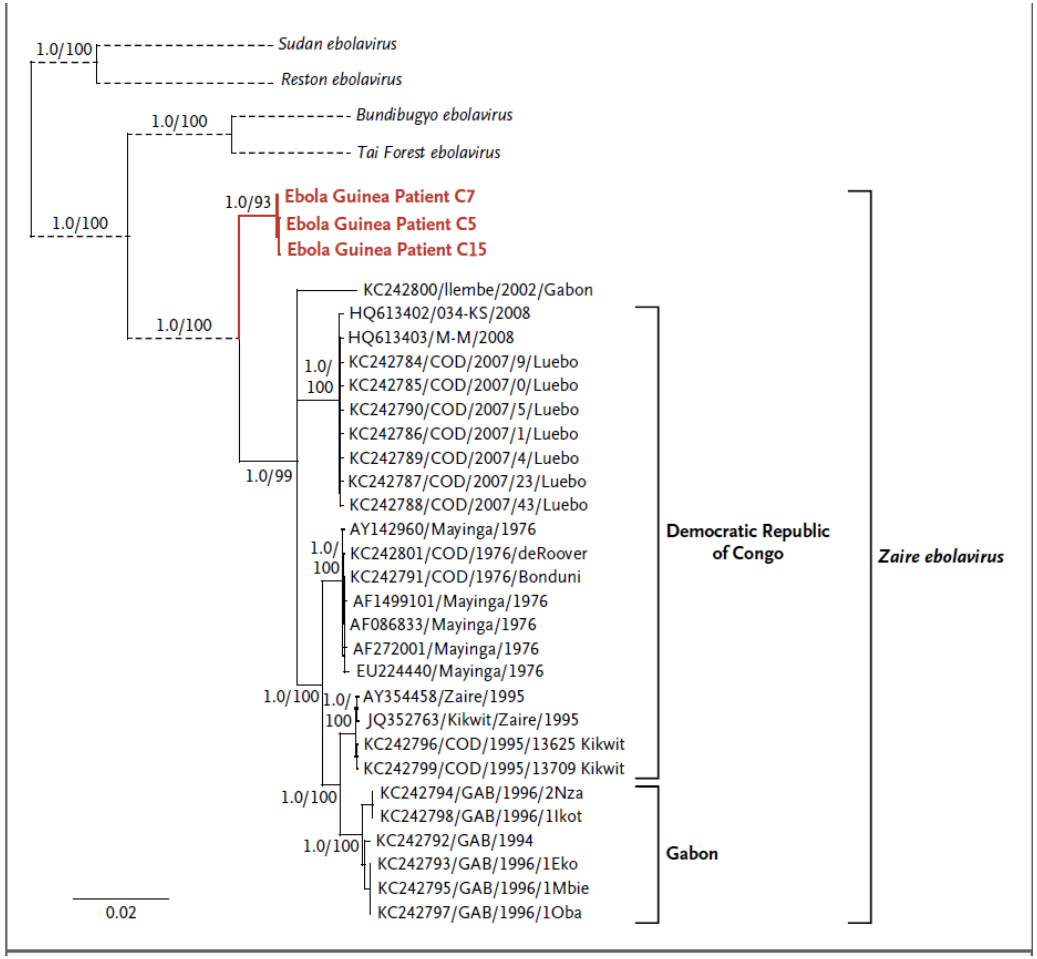
Mahanty S. Lancet ID 2004

The NEW ENGLAND JOURNAL of MEDICINE

BRIEF REPORT

Emergence of Zaire Ebola Virus Disease in Guinea — Preliminary Report



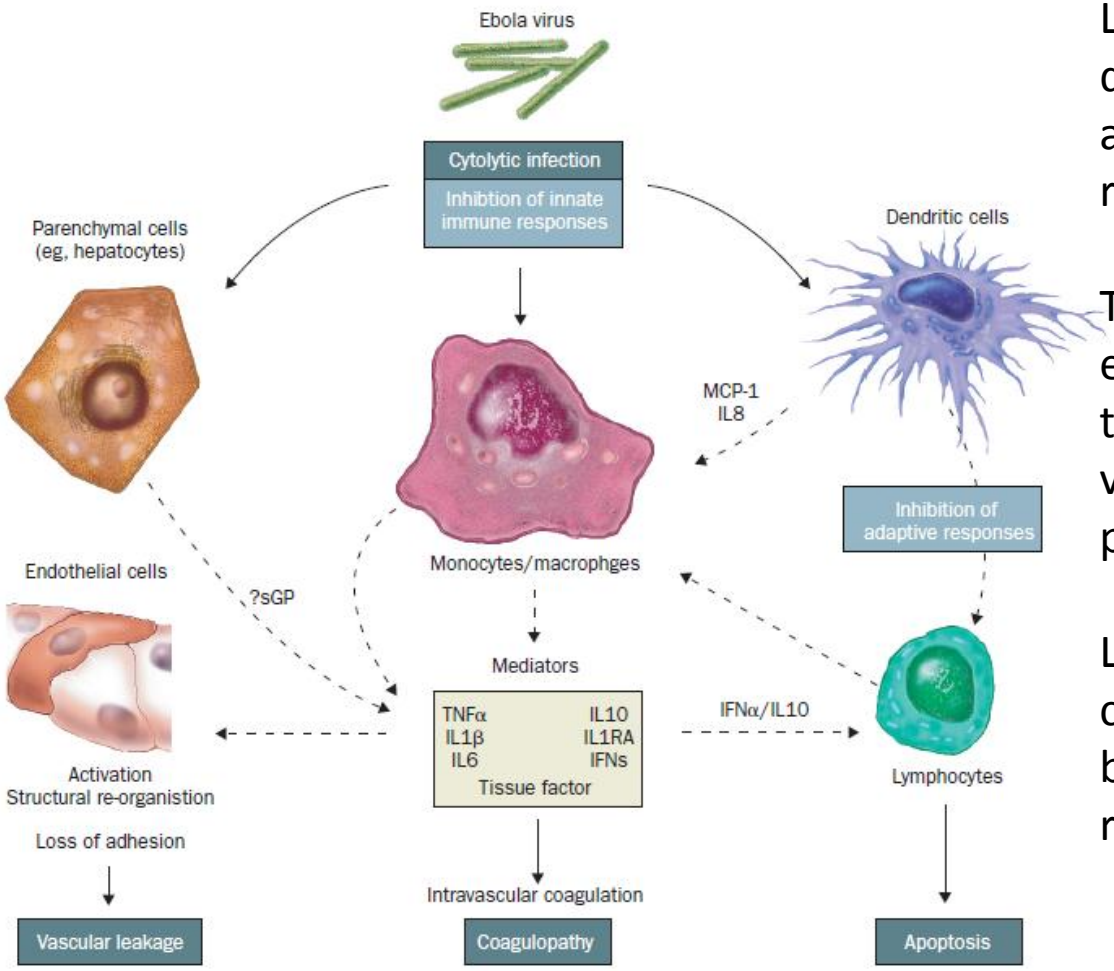


A separate clade for the Guinean EBOV strain in sister relationship with other known EBOV strains

EBOV strain from Guinea has evolved in parallel with the strains from the Democratic Republic of Congo and Gabon from a recent ancestor and has not been introduced from the latter countries into Guinea

Baize S, et al. NEJM Apr 2014

Pathogenesis of EVD



Lysis of monocytes/macrophages, dendritic cells, and hepatocytes and suppresses innate immune responses

The severe illness (including DIC and endothelial damage) results from the combined effects of widespread viral cytolysis and massive release of proinflammatory mediators

Lymphocyte apoptosis may contribute to immunosuppression by weakening adaptive immune responses.

Mahanty S. Lancet ID 2004

Transmission of infections (1)

- Through exposure to bats: experience from Uganda
    - Colebunders R, et al. J Infect Dis. 2007

- Through exposure to wild animals: experience from Gabon
    - Georges-Courbot MC, et al. Emerg Infect Dis. 1997

Transmission of infections (2)

- Human to human:
  - no EVD has been reported in persons whose contact with an infected person occurred only during the incubation period
  - risk for person-to-person transmission is greatest during the latter stages of illness when virus loads are highest
  - epidemiologic studies in humans do not indicate that VHF is readily transmitted from person to person by the airborne route
  - no evidence that filoviruses are carried by mosquitoes or other biting arthropods

Detection of virus at different body sites

- Monkeys experimentally infected with Ebola virus (Reston strain), fever and other systemic signs of illness preceded detection of infectious virus:
  - pharynx by 2-4 days
  - in the conjunctivae and on anal swabs by 5-6 days
  - in the nares by 5-10 days

Jahrling PB, et al. Arch Virol Suppl 1996

Experience from outbreak at Kikwit, Democratic Republic of the Congo, 1995

- 173 household contacts of the primary cases
- 28 (16%) developed EHF
- All secondary cases had direct physical contact with the ill person (rate ratio [RR], undefined; P<.001)
- Among those with direct contact, exposure to body fluids conferred additional risk (RR, 3.6; 95% confidence interval [CI], 1.9-6.8)
- After adjusting for direct contact and exposure to body fluids, adult family members, those who touched the cadaver, and those who were exposed during the late hospital phase were at additional risk
- None of the 78 household members who had no physical contact with the case during the clinical illness were infected (upper 95% CI, 4%)

Dowell SF, et al. J Infect Dis. 1999;179 Suppl 1:S87.

Transmission of infections (3)

- Examples of nosocomial transmission:
  - Contaminated syringes
    - Piot P, et al. Ebola Virus Haemorrhagic Fever, Pattyn, S (Ed), Elsevier/North-Holland, Amsterdam 1978
  - Operation of an infected patient
    - Khan AS, et al. J Infect Dis. 1999
- Other possible scenarios of transmission:
  - Lab workers
  - Bioterrorism

Clinical manifestations (1)

Initial:

- Abrupt onset of fever, chills, and general malaise
- Other signs and symptoms include weakness, severe headache, muscle pain, nausea, vomiting, diarrhea, and abdominal pain, nonproductive cough and pharyngitis

Within 1st week:

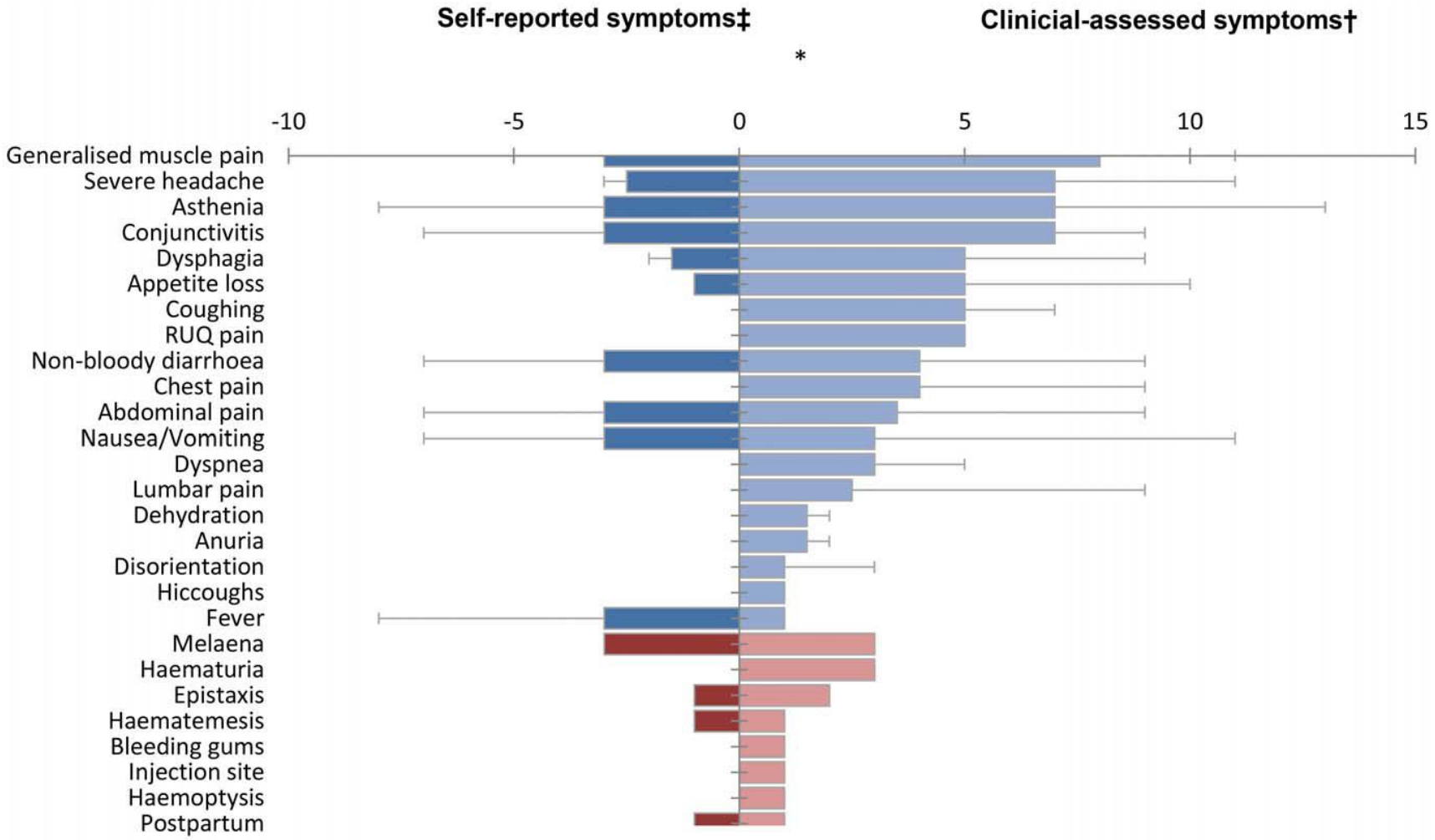
- progressive worsening of prostration, stupor, and hypotension
- signs of impaired coagulation: conjunctival hemorrhages, easy bruising, and failure of venipuncture sites to clot. Gross bleeding is common only in moribund patients
- nonpruritic maculopapular rash

Death from multiple organ failure and haemorrhage usually occurs by the second week of illness

Clinical manifestations (2)

- Survivors:
  - Slow recovery from organ dysfunction, profound weight loss and slow regain in body weight
  - Late complications:
    - musculo-skeletal symptoms most common
    - Others: arthralgia, ocular diseases, parotitis, orchitis, hearing impairment, pericarditis, amenorrhoea

Median duration in days of symptoms from self-reported onset until clinical outcome among 26 symptomatic laboratory confirmed EVD (11 died), Bundibugyo District, Uganda (November 2007–February 2008)



Roddy P, et al. PLOS One 2012

Lab findings

- Early: Leukopenia and lymphopenia, thrombocytopenia
- In days: transaminase elevations and coagulation abnormalities
- Terminal illnesses: acute kidney injury, metabolic acidosis

Lab confirmation of EVD

- PHLC provides RT-PCR testing (prior arrangement is required)
- Specimen: serum/plasma (EDTA blood is preferable)
- TAT for Negative/preliminary positive results: 3 hours after specimen reception at PHLC

Differential diagnoses

- ***Malaria***
- Other DDx
  - Typhoid fever
  - Yellow fever
  - Other viral haemorrhagic fever, e.g. Lassa fever, -
  - dengue fever
  - Staphylococcal or streptococcal infection +/- toxic shock syndrome
  - Gram-negative sepsis
  - Meningococcemia
  - leptospirosis

Prognostic factors (1)

43 cases (17 died) diagnosed on the basis of positive acute-phase diagnostic samples, Bundibugyo District, Uganda, 2007

	Case-patients confirmed by acute-phase sample, n = 43		
Characteristic	No. survived, n = 26	No. died, n = 17	p value
Mean age, y (range)	33 (12–50)	42(20–70)	0.039†
Male sex (%)	16 (62)	8 (47)	>0.100‡
Mean incubation period, d (95% CI)§	5.7 (4.4–7.0)	7.4 (5.4–9.3)	>0.100†
Signs and symptoms, no. reporting/no. available (%)¶			
Fever	26/26 (100)	16/16 (100)	>0.100
Fatigue	22/23 (96)	14/14 (100)	>0.100
Headache	21/25 (84)	14/15 (93)	>0.100
Nausea/vomiting	24/26 (92)	13/15 (87)	>0.100
Abdominal pain	23/26 (88)	13/14 (93)	>0.100
Muscle/joint pain	19/23 (83)	12/14 (86)	>0.100
Diarrhea	24/26 (92)	13/15 (87)	>0.100
Anorexia/weight loss	19/23 (83)	12/15 (80)	>0.100
Difficulty swallowing	10/23 (43)	6/15 (60)	>0.100
Rash	9/26 (35)	5/15 (33)	>0.100
Difficulty breathing	6/23 (26)	8/14 (57)	0.085
Hiccups	4/23 (17)	6/15 (40)	>0.100
Bleeding#	11/26 (42)	9/17 (53)	>0.100

MacNeil A, et al. EID 2010

Prognostic factors (2)

- Peripheral blood samples obtained from patients during an outbreak of Ebola virus (Sudan species) disease in Uganda in 2000
- Ebola virus RNA levels (virus load) were found to be much higher in infected patients who died than patients who survived the disease
- Infected patients who died had more profound leucopenia, reduced numbers of T cells, CD8+ T cells, and evidence of impaired humoral immune response

Sanchez A. et al. J Virol. 2004

Clinical management

- No specific therapy of proven benefit
- Supportive treatment
- Strict adherence of infection control measures is essential to prevent transmission

Q & A for ZMapp, CDC, US

- Experimental treatment for use with individuals infected with Ebola virus.
- A combination of three different monoclonal antibodies that bind to the protein of the Ebola virus.
- The manufacturer reports that there is a very limited supply, so it cannot be purchased and is not available for general use.
- The drug has not gone through clinical trials, meaning its safety and effectiveness has not yet been tested in humans.

http://www.cdc.gov/vhf/ebola/outbreaks/guinea/qa-experimental-treatments.html

Combination of monoclonal antibodies against ebolavirus glycoproteins

- Principle of therapy: slow the replication cycle of the virus, allowing the host's immune system to clear the infection
- Reduce mortality in non-human primate model of infections
  - Pettitt J, et al. Sci Transl Med. 2013
  - Qiu X, et al. Sci Transl Med. 2013

TKM- Ebola

- A combination of modified small interfering RNAs (siRNAs) targeting the L polymerase (EK-1 mod), viral protein VP24 (VP24-1160 mod), and VP35 (VP35-855 mod) were formulated in table nucleic acid-lipid particles (SNALPs)
- Conferred post-exposure protection in non-human primate model
  - Geisbert TW, et al. Lancet 2010
- Phase I trial was commended in Jan 2014

Other Novel treatment reported in literature

- AVI-7357: antisense Phosphorodiamidate Morpholino Oligomers (PMOs) targeting the VP24 gene of Ebola virus. (in vitro studies)
- BCX4430, a novel synthetic adenosine analogue, inhibits viral RNA polymerase function, acting as a non-obligate RNA chain terminator. (in vitro and animal studies)
- p38 MAPK pyridinyl imidazole inhibitors (in vitro studies)
- T-705 (favipiravir), nucleotide analog that selectively inhibits the viral RNA dependent RNA polymerase or causes lethal mutagenesis upon incorporation into the virus RNA (in vitro and animal studies)
- Amiodarone, a multi-ion channel inhibitor and adrenoceptor antagonist, is a potent inhibitor of filovirus cell entry at concentrations that are routinely reached in human serum during antiarrhythmic therapy (in vitro studies)

Studies on other therapies

- Ribavirin X
- Interferon α: treatment of non-human primates with human interferon-alpha-2b, only slightly delayed the onset of illness and death
  - Bray M. J Gen Virol. 2001
- Interferon β: increased survival time of non-human primates infected with a lethal dose of Ebola virus, although it failed to alter mortality
  - Smith ML, et al. JID 2013

Convalescent blood products (1)

Outbreak at Kikwit, Democratic Republic of the Congo, 1995

- 8 patients were transfused with blood donated by 5 convalescent patients
- All were seriously ill with severe asthenia, 4 presented with hemorrhagic manifestations, and 2 became comatose as their disease progressed.
- Only 1 transfused patient (12.5%) died; this number is significantly lower than the overall case fatality rate (80%)

Mupapa K, et al. JID 1999

Convalescent blood products (2)

Outbreak at Kikwit, Democratic Republic of the Congo, 1995

- Subsequent epidemiologic study with conditional probability analysis to examine the effectiveness of whole blood transfusion from convalescent patients on survival
- The main predictor of survival in the proportional hazards model was age.
- No statistical evidence of a survival benefit of transfusion of blood from convalescent patients was evident after adjusting for age, sex, and the days since onset of symptoms (P ≥ 0.1713)

Sadek RF, et al. JID 2009