Early Research with HIV

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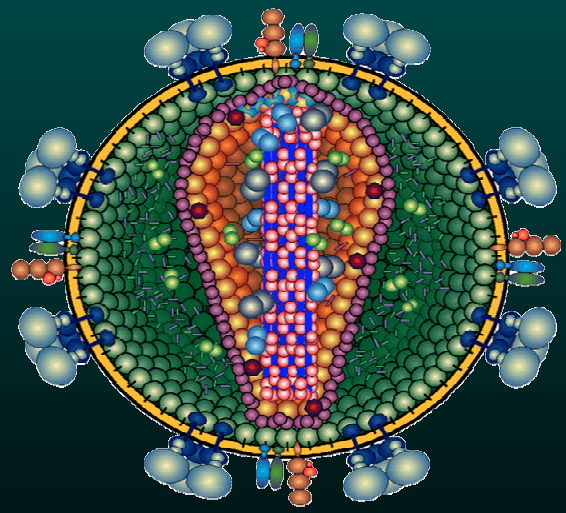

Retroviral Vaccine Section

NCI-Frederick

- Established 1972 by a Presidential directive to convert the former DoD Biological Defense Research Laboratories into “a leading center for cancer research.”
- Government-owned Contractor-operated (GoCo) facility
- FFRDC – Federally Funded Research and Development Center.
- Former BW labs (BL-2 and BL-3) and an FFRDC

Retroviral Vaccine Section


NATIONAL CANCER INSTITUTE
NATIONAL INSTITUTES OF HEALTH
DEPARTMENT OF HEALTH & HUMAN SERVICES

Chronology of HIV-1 Production

Requested to produce HIV-1 infected cells to assist in development of a screening assays for the nation's blood supply.

April 9, 1984 - Picked up HIV-1 infected cells and began production

Retroviral Vaccine Section


WARNING

Chronology of HIV-1 Production

April 9, 1984 - Picked up HIV-1 infected cells and began production

June 19, 1984 - Provided 100 liters of virus-infected cells to 5 companies charged with developing assays to detect HIV-1 infection in blood

March 1985 - FDA Approved assays to test blood used in transfusions

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1984 - ~7200 people infected with HIV-1 by blood transfusions

1989 - < 450 people infected with HIV-1 by blood transfusions

Retroviral Vaccine Section

Viral Vaccines (Human)

Disease	Virus
Smallpox	Vaccinia
Rabies	Rabies Virus
Yellow Fever	Yellow Fever Virus
Poliomyelitis	Poliovirus
Measles	Measles Virus
Mumps	Mumps Virus
Influenza	Influenza Virus
Respiratory Disease	Adenovirus
German Measles	Rubella Virus
Hepatitis	Hepatitis B Virus

Retroviral Vaccine Section


BECKMAN

Infection of chimpanzees by human T-lymphotropic retroviruses in brain and other tissues from AIDS patients.

Gajdusek DC, Amyx HL, Gibbs CJ Jr, Asher DM, Rodgers-Johnson P, Epstein LG, Sarin PS, Gallo RC, Maluish A, Arthur LO, et al.

Lancet. 1985, 8419:55-6

- Chimpanzees were readily infectable with HIV-1
- Titered HIV-1 in Chimpanzees
- Provided this infectious stock of HIV-1 to investigators world-wide

Characterization of envelope and core structural gene products of HTLV-III with sera from AIDS patients.

Robey WG, Safai B, Oroszlan S, Arthur LO, Gonda MA, Gallo RC, Fischinger PJ.

Science. 1985, 228:593-5.

- Showed that the major envelope protein of HIV-1 was gp120
- The quantity of gp120 was under represented on purified virus
  - Gp120 was purified from spent media and cell membranes
- Provided and immunogen (gp120) for sub-unit vaccine

Retroviral Vaccine Section

Challenge of chimpanzees (Pan troglodytes) immunized with human immunodeficiency virus envelope glycoprotein gp120.

Arthur LO, Bess JW Jr, Waters DJ, Pyle SW, Kelliher JC, Nara PL, Krohn K, Robey WG, Langlois AJ, Gallo RC, et al.

J Virol. 1989, 63:5046-53.

- Monomeric gp120 vaccine was not protective in vaccine/challenge model.

Retroviral Vaccine Section

Cellular proteins bound to immunodeficiency viruses: implications for pathogenesis and vaccines.

Arthur LO, Bess JW Jr, Sowder RC 2nd, Benveniste RE, Mann DL, Chermann JC, Henderson LE.

Science. 1992, 258:1935-8.

- **Cellular proteins were incorporated into the membrane of immunodeficiency viruses**

Retroviral Vaccine Section

Macaques immunized with HLA-DR are protected from challenge with simian immunodeficiency virus.

Arthur LO, Bess JW Jr, Urban RG, Strominger JL, Morton WR, Mann DL, Henderson LE, Benveniste RE.

J Virol. 1995, 69:3117-24.

- **Immunization with human cellular proteins (HLA-DR) protected macaques from challenge with SIV propagated in human cells.**
- **HLA-DR immunization did not protect from challenge with SIV propagated in macaque cells.**
- **Suggested that protection seen with inactivated viral vaccine studies were due to cellular component of the vaccine.**

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Findings from our lab unfortunately suggested that inactivated virus vaccines may not work for immunodeficiency virus vaccines

1. Finding gp120 in low concentrations on purified viruses and in high levels in spent culture fluid suggested shedding of this glycoprotein from the virus during purification.

2. Cellular proteins on viruses were responsible for the protection in the vaccine/challenge experiments.

Retroviral Vaccine Section

Viral Vaccines (Human)

Disease	Virus	Type of Vaccine
Smallpox	Vaccinia	Live, Attenuated
Rabies	Rabies Virus	Inactivated
Yellow Fever	Yellow Fever Virus	Live, Attenuated
Poliomyelitis	Poliovirus	Inactivated (Salk) Live, Attenuated (Sabin)
Measles	Measles Virus	Inactivated Live, Attenuated
Mumps	Mumps Virus	Live, Attenuated
Influenza	Influenza Virus	Inactivated
Respiratory Disease	Adenovirus	Live, Attenuated
German Measles	Rubella Virus	Live, Attenuated
Hepatitis	Hepatitis B Virus	Virus-like particle

Retroviral Vaccine Section

Envelope glycoprotein incorporation, not shedding of surface envelope glycoprotein (gp120/SU), is the primary determinant of SU content of purified human immunodeficiency virus type 1 and simian immunodeficiency virus.

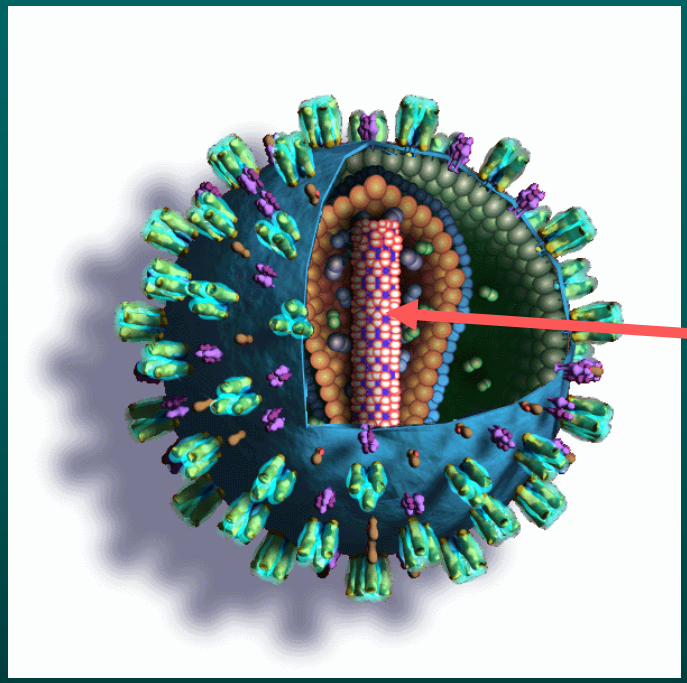
Chertova E, Bess Jr JW Jr, Crise BJ, Sowder II RC, Schaden TM, Hilburn JM, Hoxie JA, Benveniste RE, Lifson JD, Henderson LE, Arthur LO.

J Virol. 2002, 76:5315-25.

- **Gp120 is difficult to remove from purified viruses**
- **Incorporation of the transmembrane glycoprotein (gp41) determines the quantity of gp120 on the virus.**

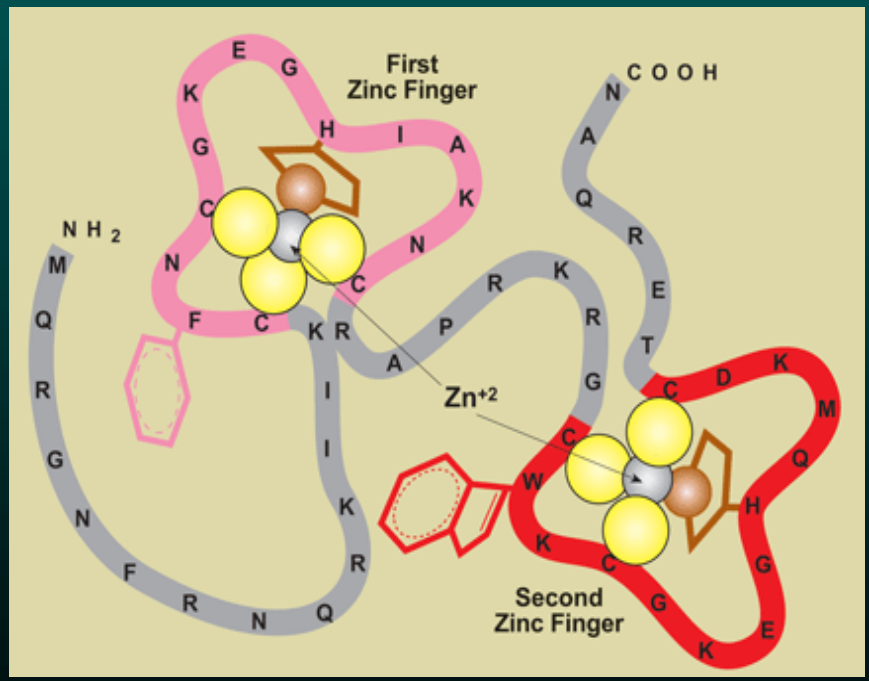
Retroviral Vaccine Section

Nucleocapsid Protein Required For Multiple Steps in the Retroviral Life Cycle



- RNA Packaging
- Reverse Transcription
- Integration

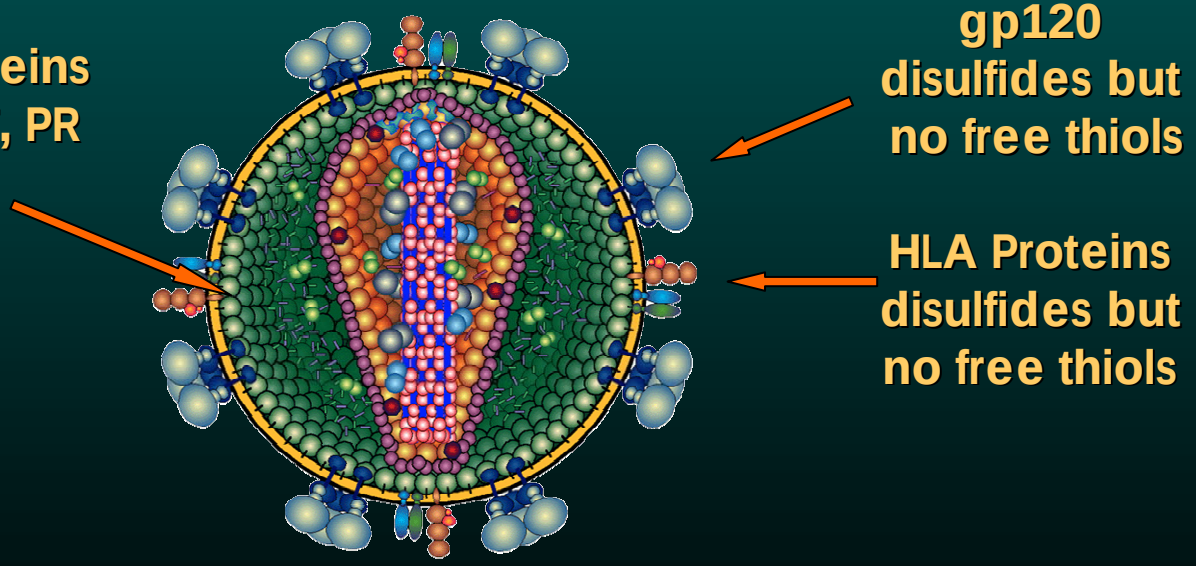
NC protein



- Retroviral Zn Fingers required for NC functions
- Modification of Zn Fingers eliminates infectivity
  - Mutagenesis
  - Chemical

Retroviral Vaccine Section

- **Interior Proteins - Reducing Environments**
  - Internal Proteins - Cys in zinc finger coordination and free thiols
- **Exterior Proteins - Oxidizing Environments**
  - Surface proteins - Cys residues are oxidized (disulfide bonds)



Retroviral Vaccine Section

Inactivation of human immunodeficiency virus type 1 infectivity with preservation of conformational and functional integrity of virion surface proteins.

Rossio JL, Esser MT, Suryanarayana K, Schneider DK, Bess JW Jr, Vasquez GM, Wiltrout TA, Chertova E, Grimes MK, Sattentau Q, Arthur LO, Henderson LE, Lifson JD.

J Virol. 1998, 72:7992-8001

Chemical inactivation of retroviral infectivity by targeting nucleocapsid protein zinc fingers: a candidate SIV vaccine.

Arthur LO, Bess JW Jr, Chertova EN, Rossio JL, Esser MT, Benveniste RE, Henderson LE, Lifson JD

AIDS Res Hum Retroviruses. 1998, 3:S311-9

Retroviral Vaccine Section

Properties of Inactivated Virions

- Conformationally and functionally intact Env
  - Immunoprecipitation with gp120 MAbs (conformational epitopes)
  - CD4-dependent binding
  - Mediate “Fusion-from-without”
- In vivo immunogenicity
- Not detectably infectious
  - In vitro and in vivo

Retroviral Vaccine Section

ACKNOWLEDGEMENTS

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Only 5 other laboratories in the world are working on HIV-1 killed virus vaccines. **Three use processes which destroy the envelope proteins**.

- Active Collaboration
  (Two orthogonal methods of killing - FDA)

- The only research laboratory in the world producing and purifying virus at this scale
  (Supplying killed SIV and HIV-1 as a reagent world-wide)

- Evaluating killed SIV as a vaccine

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