ASSISTED REPRODUCTION WITH SPERM FROM HIV-INFECTED MEN

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Retroviruses and reproduction

The venereal transmissibility of human immunosuppressive virus (HIV) and human T-cell leukemia virus, I (HTLV-I) impacts reproductive medicine in a number of ways. The possibility of HIV transmission by donor semen has received prompt and appropriate attention, with most centers using only cryopreserved semen stored according to quarantine guidelines for monitoring possible seroconversion of donors. As the incidence of HIV-positive pregnant women rises, obstetric units are evolving safeguards for medical staff attending deliveries. Counseling women at risk for HIV infections about the possible outcome to offspring is becoming increasingly necessary. Some centers for assisted reproductive technology require mandatory screening of patients for HIV antibody as a safeguard for their laboratory and medical staff.

However, a consequence of the venereal transmission of human retroviruses that has not received widespread attention is the impact on reproduction per se. That the impact will be a negative one is due to
First electron micrograph of HIV in semen

1983:

Reported:
First Int’l Aids Congress, 1985; AIDS 1: 419, 1988
The Need:

By 1987, 10,000 men and boys with hemophilia in the U. S. infected with HIV through clotting factors

Thousands of men and women infected by blood transfusion and IV drug abuse

1990: First Congress on “AIDS and Reproduction,” Genoa, Italy
“Sperm washing” and IUI advocated by Augusto Semprini

1991: Centers for Disease Control: “IUI with sperm from HIV infected men not proven safe”
The Goal:

Develop methods to reduce, hopefully eliminate, transmission of HIV from infected male to uninfected female attempting pregnancy.

The Hope:

HIV infection in the male would be regarded as a form of male factor infertility.

The Plan:

Develop sensitive assays for HIV and HIV-infected cells to eliminate the use of sperm from semen with detectable virus.
Semen HIV assays:

**Semen specimen**

- **One-Half**
  - Washed with TYB, cryopreserved, quarantined

- **One-Fourth**
  - Fixed, immunostained (CD45, panleukocyte marker)

- **One-Fourth**
  - Qiagen column, RT-PCR, triple-bracket-nested amplification of HIVgag
CD45+ leukocytes (red-brown)

Immature germ cells (blue)

Mature sperm (dark blue)
RT-PCR steps:

Semen cells and particles, but not sperm heads, lysed to release RNA and DNA:

- Reverse transcription to convert HIV RNA to HIV DNA
- First round of multiplex PCR: co-amplify 341 base pair segment of HIVgag and a 268 base pair segment of beta-globin
- Second round of multiplex PCR: co-amplify 144 base pair nested segment of HIVgag and a 172 base pair nested segment of beta-globin
- Third round of PCR: amplify 105 base pair nested segment of HIVgag
RT-PCR steps:

- **RT → PCR**: 2 ul
  - 341 bp HIV gag + 268 bp b-globin
  - 144 bp HIV gag + 172 bp b-globin
  - 105 bp HIV gag

Gel electrophoresis:

- L 1 2 3 4 5 6 Pos Bl 1 2 3 4 5 6 Pos Bl L

20 copies HIV RNA

1 infected cell

*Biotechniques 15: 128, 1993*

*Fertil Steril 60: 576, 1993*
HIV in sequential semen specimens:

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Month of study

U = undetectable
• = HIV positive
Program steps:

Set up as a clinical trial; all couples treated uniformly until at least 100 births; then convene a workshop to alter as indicated

1. Interview with HIV counselor
   a) Health status of male:
      regular visits to infectious disease specialist;
      no “minimum” blood viral load;
      explain complex nature of HIV in semen;
   b) Health status of female:
      at risk for HIV infection (and possibly Hep C/B);
      need for fertility evaluation;
      need for follow-up HIV antibody testing;
   c) Fertility clinic referral, if needed
   d) Need to test baby sometime during the first year
Program steps:

(2) Submit semen specimens for testing, using overnight transport kit if not in Boston area

(3) Sperm from half of specimen is cryopreserved, remainder of specimen is tested for leukocytes, HIV virus, and HIV-infected cells

(4) If HIV detected, sperm discarded; fresh specimen is tested

(5) If two specimens in a row test positive for HIV, man may be referred to urologist, if suspect GU tract infection; or to infectious disease physician if adjustment to antiviral therapy is indicated.
Program steps:

(6) Cryopreserved sperm from two specimens with undetectable viral burden stored until shipped to collaborating fertility treatment center

(7) Pregnancy attempted by IVF or oligospermia cup (NOT IUI)

(8) Female partner tested for HIV antibody at 3 weeks, 3 months and 6 months after pregnancy attempt -- whether or not pregnancy achieved

(9) Baby tested for HIV antibody within first year of life
Program began in 1996
  Controversial, caught in hospital merger, relegated to a public charity

First baby born in 1998
  (Stephen Smith, “Fertility Race,” Minnesota Public Radio)
Conceiving Ryan
by Stephen Smith of Americanradioworks
Minnesotta Public Radio
2000-2006:

608 semen specimens from 262 men tested for HIV

107 (19%) semen specimens tested positive, discarded

151 couples proceeded to ART

69 pregnancies, 62 babies born, 10 sets of twins
As of September, 2007:

- 26 collaborating fertility clinics
- 71 Babies born, 8 ongoing pregnancies
- All Moms and Babies test negative for HIV
Future Plans:

Seek FDA/CDC approval for semen testing protocol

Seek insurance recognition of HIV infection as male factor infertility

Correlate data, not an “infertile” population

Provide semen test for Hep C/B

FUNDING NEEDED
To help, or for information:

lab@bedfordresearch.org

(all inquiries strictly confidential)