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Semen decontamination: From concept to application

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INTRODUCTION:
The evaluation of novel disinfection procedures, to eliminate a variety of bacterial and viral pathogens from human semen, was initiated in 2002. *In vitro* spiking of semen from sero-negative males with e.g. human immunodeficiency virus (HIV) and hepatitis C virus using various density gradients/layers followed. The ability of the ProInsert™, Nidacon Int. to reduce viral copies >1.5 x 10^5 times/or lower than the lowest level of detection (LLD) was encouraging. Experimental processing of semen from HIV+ males followed in 2007.

Aim:
An overview on the initiation and outcome of the decontamination program, lessons learned, as well as subsequent experimentation will be discussed (2002-2014).

METHODS:
More than 500 semen samples from >100 HIV+ patients have been analysed since 2007. Male patients enrolled in the program provided 2 to 4 semen samples, depending on the quality of the processed sperm samples. The initial semen sample was used for diagnostic purposes, and submitted for microbiological evaluation combined with reverse transcription PCR quantitative and qualitative tests. The second sample was processed for therapeutic use, by means of discontinuous density gradient centrifugation using a ProInsert™ according to the manufacturer's specifications. Different versions of the ProInsert™ were tested during 2009-2012, combined with on-going experimentation.

RESULTS:
Due to budget restrictions and capacity, a maximum of 2 decontamination procedures were performed per week (114 semen procedures per year). The males were on average 38.5 years of age (26 min & 54 yrs. max), mostly on anti-retroviral therapy (66%) with an HIV sero-negative female partner (68%). The patients presented with CD4 counts (117 min & 1338 max cells/µl), with no statistical difference ($p>0.05$) in semen parameters for the CD4 categories of above and below 350 cells/µl. The blood plasma viral load (BPVL) ranged between LLD to 249,015 and the neat seminal viral load (SVL) between LLD and 1,443,350 copies/ml. Just over 50% of patients presented with undetectable BPVL, of which 30% had a positive SVL. PCR validation errors were present in <10% of cases. Proviral DNA was found in 2% of post-processed sperm samples and none of the samples tested positive for RNA

CONCLUSION/DISCUSSION:
In 2014, 60% of patients were "in-house" and 40% were from private practices. Same day viral validation would preclude cryopreservation of samples until validation results are available. On the other hand, since intra-cytoplasmic sperm injection and molecular viral validation are expensive and not readily available in developing countries, we have to strive towards a superior but simplistic risk-reduction semen decontamination procedure.