Preventing Donor Derived Infections Through Virology Screening Reducing Risk and Increasing Organ Availability

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OUTLINE

1. *Background*
   - Infections
   - Risks
2. *Testing Protocols Australia*
   - Current guidelines
3. *NAT and Serology testing*
   - Window period (WP)
   - Testing Algorithm at SEALS
   - *Parallel testing*
   - *Results over a 20 month period*

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Estimated Number of Persons with Chronic Blood-borne Virus Infections 2000

Region	Population (millions)	Chronic infections (millions)		
		HIV	HCV	HBV
Africa	749	22.7	22.5	59.3
Asia	3,585	7.3	107.5	286.8
Latin America	504	1.7	15.1	10.3
Europe	729	0.8	21.8	10.9
Oceania	30	0.0	0.9	2.4
North America	305	0.9	9.1	1.9
Total	5,902	33.4	176.9	371.6

Global burden of BBV

Cumulative number of infections by HBV, HIV, HCV: 1/10th of world population

Mortality	related to	HIV	2.1 million
		HBV	1 - 1.2 million
		HCV	~> 1 million
Chronic infections	Cost of HCV therapy		~£8,500 / year
	Cost of HAART		$ 12,000 - 24,000 / year (less in developing countries)
	Cost of HBV therapy		$3,500 (lamivudine)
			$7,400 - 9,000 (adefovir, entecavir, telbivudine)
			$24,000 peginterferon
Liver transplant	$ 100,000 - 400,000 in developed countries		
	$ 18,500 in developing countries		
Testing	> $ 1.3 Billion		

Risk of acquisition of a BBV through organ transplantation

- The prevalence of the virus in the donor population
- The viral load in the donor
- The specific organ transplanted
- The efficiency of virus transmission after contact with blood and tissues

Organ donation in Australia

Population	~22 million
DOD	~247 donors (2009)
DODR	11.3 donors/million popn
Tx from DOD	1,187~ (2009)
Waiting List	1650 patients (2009)
Dialysis Px	2,337 patients (2009)

Prevalence of BBV in Australia

Virus	Infected Population in Australia	Prevalence rate (%)	Prevalence rate in High Risk (%)
HIV	17,000	0.085	0.17
HBV	88,000	0.44	28
HCV	210,000	1.05	36
Total	315,000	1.57	64.17

Prevalence among organ donors at SEALS NAT Laboratory

Virus	HIV		HCV		HBV		
BBV Marker	Anti-HIV	HIV RNA	Anti-HCV	HCV RNA	HBsAg	Anti-HBc	HBV DNA
Prevalence at SEALS (%)	0.00	0.00	1.92	0.00	0.64	7.05	0.64

Increased Risk Donors

- Aiming to increase donation rate
- Testing protocols not nationally agreed yet, but in progress
- Draft BBV policies in place in a number of States, managed by State Organ Donation Agencies
- *Donor assessment NSW*
  1. *Clinical assessment then BBV assays*
  2. *Prospective NAT*

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Organs for Transplant Time to NAT?

HBV DNA real time PCR

- TGA Not a requirement yet (? Living donors)
- TMA
  - Not available within timeframe (NAR)
  - $40-50 per HIV/HCV mpx test
- PCR Turnaround time 6 hours
  - $700-800 per HIV+HCV assay
  - ?spread cost across the number of O&Ts
- **RT-PCR**
  - **Turnaround time <3 hours**
  - **Registration status of assays**
  - **TPX configurations for developing world ($20)**
- Feasible options for organ donation
  - Validation
  - Status of cadaveric validation

Universal Viral Screening Markers

[NSW DOH Organ Donation and Transplantation Managing Risks of transmission of HIV, HCV and HBV]

Serology:
- Anti-HIV-1/2
- Anti-HCV
- Anti-HTLV-I/II
- HBsAg
- Anti- HBc
- Anti-HBs
- anti-EBV
- anti-CMV
- Syphilis antibody (TPHA)

NAT:
- HIV-1 RNA
- HCV RNA
- (HBV DNA)
- Prospective in "increased risk"
- Retrospective

Donors with identified risk factors
- MSM
- IV Drug Users
- Incarceration in previous 12 months
- Sexual partners of above
- Unexplained fever /weight loss/ LAD/cough etc
- Partner with HIV/HBV/HCV
- Prostitution
- STD in past 12 months
- Cosmetic body piercing/tattooing
- (cocaine snorting)
- Physician concern

TSANZ CONSENSUS STATEMENT ON ELIGIBILITY CRITERIA AND ALLOCATION PROTOCOLS

- Submitted to AOTDTA in July 2010
- Hepatitis B testing and use of hepatitis B positive donor
- Hepatitis C testing and use of hepatitis C positive donor organs
- Use of prospective NAT for increased risk donors

International Guidelines

- **GUIDANCE ON THE MICROBIOLOGICAL SAFETY OF HUMAN ORGANS, TISSUES AND CELLS USED IN TRANSPLANTATION UK, Feb 2011**
  - Clear guidelines for laboratories testing donations, including parallel testing
  - Well defined testing algorithms for interpretation of results and management of repeat reactive results
  - Donor blood/plasma samples must be archived for at least 10 years in a retrievable manner and testing records for a period of 30 years,

Current Testing Protocols in Australia

Blood donor screening
- Serology testing for HIV1/2, HBV, HCV, HTLV I/II, Syphilis, Malaria, CMV
- NAT testing for HIV1, HCV

Organ donor screening
- Serology testing for HIV1/2, HBV, HCV, HTLV I/II, Syphilis
- Serology for CMV, EBV, T. gondii
- From October 2009, BBV SEALS NAT HIV-1, HCV, HBV for organ donors in NSW and Victoria

Optimal use of NAT in SOT

Being assessed through evaluation of on-going requests and results of testing increased risk donors

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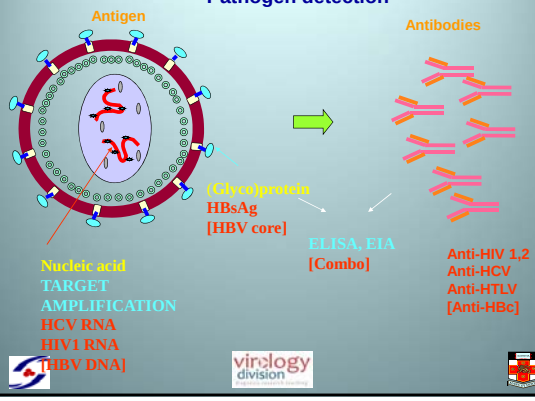
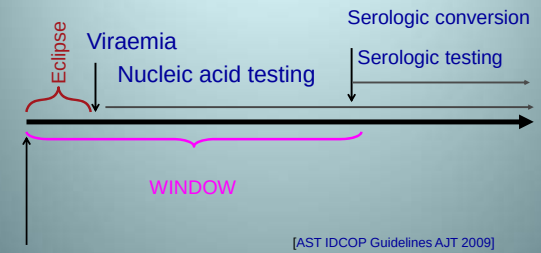
Pathogen detection



Screening for Infectious Diseases

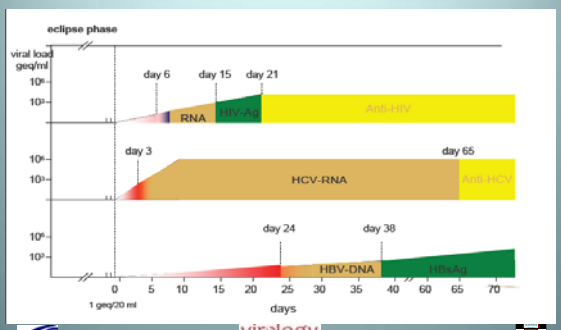


[AST IDCOP Guidelines AJT 2009]

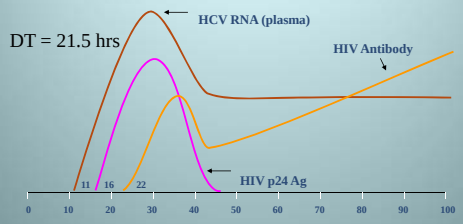
Stages of Infectivity

- Surrogate for infectivity is viraemia detectable by NAT (PCR)
- Can define four stages of viraemia
  - Pre-ramp up
  - Ramp-up (exponential increase)
  - Plateau phase
  - Post-sero-conversion phase

What is NAT Infectious Window



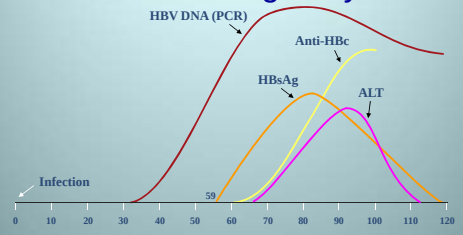
HIV markers during early infection



Theoretical Infectivity	Day 0
HIV RNA	Day 11
HIV p24 Ag	Day 16
HIV Antibody	Day 22

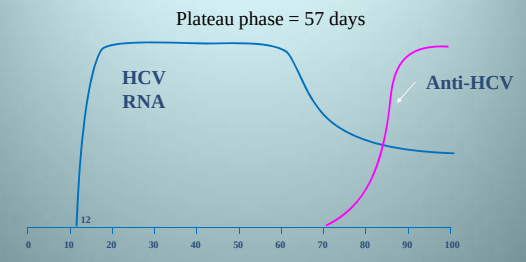
5 Days
6 Days

HBV markers during early infection



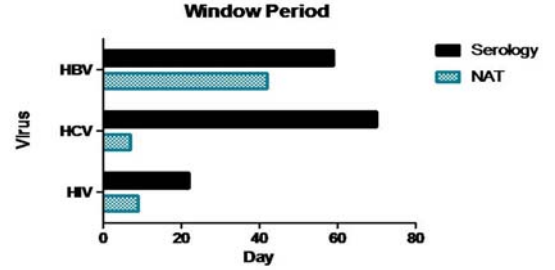
Infection	Day 0
HBV DNA	Variable, up to 31 days prior to HBsAg by ID NAT(9-11 days by MP NAT)
HBsAg	Day 59; disappears Day 120

HCV markers during early infection



Infection	Day 0
HCV RNA	Day 12
HCV Antibody	Day 70

NAT and Serology Window Periods



Limitations of Serological Tests

- Longer window period than NAT
- Do not distinguish between HCV present or past infection
- HBV scape mutants are not detected
- Occult HCV and HBV infections are not detected

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Current processes for screening

Nat Tests done at SEALS

- HCV
- HIV-1
- HBV
- 3 Molecular tests run in parallel
- LOD 10 IU/ml, TAT 8 hrs
- Additional tests being developed:
- CMV Serology
- EBV Serology
- BK/JC
- Toxoplasma

Technical laboratory aspects

- NAT Service
- Increased-risk donors
- Residual risks
- Conclusions

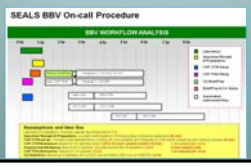
NAT/BBV Laboratory

Provide blood donor screening for HBV, HCV and HIV-1 in organ and tissue donations

- ➢Prospective/Urgent NAT for increased-risk donors (NSW & VIC)
- ➢Retrospective NAT screening in average-risk donors (NSW)

•Use of 3 different molecular tests

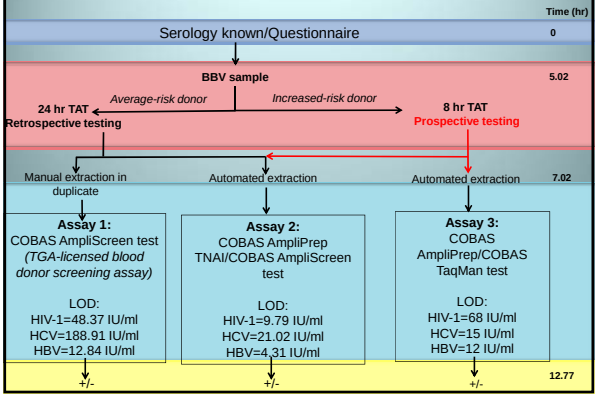
- ➢TGA licensed COBAS AmpliScreen assay
- ➢COBAS AmpliPrep TNAI/AmpliScreen assay
- ➢COBAS AmpliPrep/TaqMan assay



Prospective NAT

- 1st October 2009 to 30th August 2010
- N = 35 prospective NAT performed at SEALS, POWH
- Average delivery time from collection of blood to hospital = 5.02 h (range 1 – 15 h)
- Average TAT from specimen receipt to reporting of results = 7.75 h (range 7 – 10 h)

Algorithm for discordant result confirmation



Estimated probability that a donor is truly infection positive on given discrepant NAT assay results

PCR Result			Probability of True Positive			Reported as
CAS	TNAI	TaqMan	HBV	HCV	HIV	
+	-	-	0.0004	0.0007	<0.0001	Neg
-	+	-	0.5212*	0.2676	0.0107	Neg
-	-	+	0.0544	0.2975	0.0001	Neg
+	+	-	0.9972	0.9699	0.5922*	Pos
+	-	+	0.9489	0.9740	0.0095*	Pos
-	+	+	0.9999	0.9999	0. 9861	Pos

Costs

- Total costs for organ donor screening for HIV-1, HCV and HBV
  - ➢Prospective NAT = $2,200 (salary $550, consumables $1650)

Summary of BBV testing at SEALS since 1st October 2009 to 30th April 2011

Sample type	No. of donors tested by NAT	No. of organs retrieved
Routine (Average-risk donor)	121	443
Prospective (Increased-risk donor)	35 (NSW & VIC)	98
Total	**156**	**541**

Donor no.	Serology results				NAT results			Organ(s) retrieved
	HBs Ag	HBc Ab	HCV Ab	HIV-1/2 Ab	HBV DNA	HCV RNA	HIV-1 RNA	
1	-	-	+	-	-	-	-	3
2	-	-	-	-	-	-	-	5
3	-	+	-	-	-	-	-	Not medically suitable
4	+	+	-	-	+	-	-	Positive serology
5	-	+	+	-	-	-	-	1
6	-	-	-	-	-	-	-	2
7	-	+	+	-	-	-	-	3
8	-	-	-	-	-	-	-	1
9	-	+	-	-	-	-	-	4
10	-	-	-	-	-	-	-	3
11	-	+	-	-	-	-	-	4
12	-	-	-	-	-	-	-	2
13	-	+	-	-	-	-	-	2
14	n/a	n/a	n/a	n/a	-	-	-	n/a*
15	-	+	-	-	-	-	-	3
16	-	-	-	-	-	-	-	3
17	-	+	-	-	-	-	-	2
18	-	+	-	-	-	-	-	5
19	-	-	-	-	-	-	-	1
20	-	-	-	-	-	-	-	2
21	-	-	-	-	-	-	-	3
22	-	+	-	-	-	-	-	0*
23	-	-	-	-	-	-	-	5
24	-	-	+	-	-	-	-	0
25	-	-	-	-	-	-	-	6
26	-	-	-	-	-	-	-	4
27	-	-	-	-	-	-	-	4
28	-	-	-	-	-	-	-	1
29	-	-	-	-	-	-	-	5
30	-	-	-	-	-	-	-	4
31	-	-	-	-	-	-	-	4
32	-	-	-	-	-	-	-	5
33	-	-	-	-	-	-	-	4
34	-	-	-	-	-	-	-	6
35	-	-	-	-	-	-	-	
TOTAL								98

Recommendations for Laboratories performing NAT

Issue	Recommendation	Status at SEALS
Standards labs	Competency	☑
	Proficiency	☑
	Staff training	☑
Sampling and transport	Not haemodiluted	Stated in request form
	Dedicated samples	☑
	Standardised transport	☑
Laboratory Procedures	Single unit testing	☑
	Rapid TAT	☑
	Discrimination of initial positive results	☑

Hilmar *et al.* Am J Transplant 2010;10:889-899

Emerging issues

- Imported infections/zoonoses/mosquitoes
  - WNV
  - Bat lyssavirus, Hendra/Menangle/Nipah
  - Dengue, Murray Valley encephalitis
- Long-term immunosuppression + cancer
  - EBV
  - Endogenous retrovirus
  - HHV-8

Anti-HBc (+) donors

Occult Hepatitis B Infection (OBI)

Residual risk

Conclusions

- NAT screening
  - Diagnosis in 7-8 hr
  - Enabled the use of organs from donors with positive serology but not active viral infection
- New testing algorithms and additional pathogens need to be considered (i.e. CMV, EBV) for donor screening
- Improvements
  - Development of National policy for tests done, protocols and informed consent
  - Active surveillance (Biovigilance)
  - Improved knowledge by health professionals involved in OD&Tx
  - Post diagnostic counselling

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