NATIONAL HEALTH LABORATORY SERVICE

GeneXpert MTB/RIF

Progress Report

July 2013

NATIONAL HEALTH LABORATORY SERVICE

Table of Contents

1. Background to Project

This project was initiated at the request of the Honorable Minister of Health, Dr Aaron Motsoaledi, in early 2011, following the World Health Organization's strong recommendation published in December 2010 which stated that "the new automated DNA test for TB be used as the initial diagnostic test in individuals suspected of MDR-TB or HIV/TB". In essence this comprises the majority of TB suspects in South Africa. A pilot study was proposed by the TB Cluster within the National Department of Health (NDoH) while a project feasibility study was being performed with due diligence.

The pilot study was initiated in microscopy centres. The NDoH requested that at least 1 instrument be placed in each province, preferably in high burden districts. Selections were made by the TB cluster, with twenty-five microscopy centres being selected and a total of 30 instruments placed.

The NDoH funded 9 GX16 and 14 GX4 instruments for the project. FIND (The Foundation for Innovative New Diagnostics) donated 6 GX4 analysers and the Infinity or GX48 was supported by PEPFAR Right to Care funds. All instruments were placed by World TB day March 24th 2011. This placement represented about 10% of national coverage. The basis for the calculations was an assumption that 2 smears at diagnosis would be replaced by 1 Xpert® MTB/RIF assay. All instruments were interfaced to the NHLS Laboratory Information System (LIS) allowing for troubleshooting and data collection.

Since then, 274 GeneXpert instruments of varying sizes (GX4: 88; GX16:184; GX48: 2) have been placed in 207 sites – both urban and rural settings, by the National Priority Programmes of the NHLS and the NDoH, the progress of which is described in point 6 below.

2. Assays performed to date

In summary, a total of 1,747,252 specimens have been processed to date (31 July 2013). In July 167,313 specimens were processed. The total % of *Mycobacterium tuberculosis* complex (MTBC) detected in this cohort was 11.14% (18,640). As a reflection of Xpert MTB/RIF's superior sensitivity over microscopy, the average national TB positivity rate among suspects was found to be 8% using

microscopy but up to 16-18% in the first year and 13-14% in the second and third year, after introduction of Xpert® MTB/RIF assay. To date Kwa-Zulu Natal (KZN) has performed the greatest number of tests which is probably as a result of the number of instruments placed (refer to tables 1 & 2). Average Rifampicin resistance detection rates have remained around 7% since project inception (Refer to tables 3 & 4).

Table 1: GeneXpert MTB Results by province (cumulative)

Province	Year	MTB Detected	MTB Not Detected	Test Unsuccessful	Total	% MTB Detected
	/	1			**1**	
	2011	3294	15413	555	**19 262**	**17.10**
	2012	16092	85782	2893	**104 767**	**15.36**
Eastern Cape	2013	22856	152511	5406	**180 773**	**12.64**
	2011	2844	14830	33	**17 707**	**16.06**
	2012	11667	77094	280	**89 041**	**13.10**
Free State	2013	8078	74217	667	**82 962**	**9.74**
	2011	3112	18936	424	**22 472**	**13.85**
	2012	11057	72748	2285	**86 090**	**12.84**
Gauteng	2013	14126	100194	4601	**118 921**	**11.88**
	/	11	33	1	**45**	
	2011	13172	47523	1729	**62 424**	**21.10**
	2012	25766	144201	6233	**176 200**	**14.62**
Kwa-Zulu Natal	2013	21034	140334	8445	**169 813**	**12.39**
	2011	2088	17870	173	**20 131**	**10.37**
	2012	4239	31469	700	**36 408**	**11.64**
Limpopo	2013	7234	89970	3594	**100 798**	**7.18**
	2011	2643	12769	1107	**16 519**	**16.00**
	2012	4055	22020	1122	**27 197**	**14.91**
Mpumalanga	2013	4060	22761	1028	**27 849**	**14.58**
	2011	3476	14887	657	**19 020**	**18.28**
	2012	5174	29003	1977	**36 154**	**14.31**
North West	2013	6224	45723	2659	**54 606**	**11.40**
	2011	2864	16117	735	**19 716**	**14.53**
	2012	4432	23654	1192	**29 278**	**15.14**
Northern Cape	2013	4344	27311	1474	**33 129**	**13.11**
	2011	2204	10093	31	**12 328**	**17.88**
	2012	13202	68427	596	**82 225**	**16.06**
Western Cape	2013	15783	83901	1732	**101 416**	**15.56**
Total		**235 132**	**1 459 791**	**52 329**	**1 747 252**	**13.46**

NATIONAL HEALTH LABORATORY SERVICE

Table 2: GeneXpert MTB Results by province (01-31 July 2013)

Province	MTB Detected	MTB Not Detected	Test Unsuccessful	Grand Total	% MTB Detected
Eastern Cape	3 951	28 985	927	33 863	11.67
Free State	1 407	15 756	185	17 348	8.11
Gauteng	2 853	19 249	899	23 001	12.40
Kwa-Zulu Natal	3 677	25 292	1 375	30 344	12.12
Limpopo	1 346	21 674	600	23 620	5.70
Mpumalanga	947	5 075	201	6 223	15.22
North West	1 201	9 210	448	10 859	11.06
Northern Cape	658	4 466	205	5 329	12.35
Western Cape	2 600	13 906	220	16 726	15.54
Grand Total	**18 640**	**143 613**	**5 060**	**167 313**	**11.14**

Table 3: Provincial GeneXpert RIF Results in MTB detected cases (01-31 July 2013)

Province	Inconclusive	Resistant	Sensitive	No Result	Grand Total	% Rif Resistant
Eastern Cape	117	240	3 591	3	3 951	6.07
Free State	25	83	1 295	4	1 407	5.90
Gauteng	74	182	2 593	4	2 853	6.38
Kwa-Zulu Natal	90	327	3 251	9	3 677	8.89
Limpopo	43	53	1 211	39	1 346	3.94
Mpumalanga	27	100	816	4	947	10.56
North West	31	55	1 114	1	1 201	4.58
Northern Cape	9	42	607		658	6.38
Western Cape	54	141	2 404	1	2 600	5.42
Grand Total	**470**	**1 223**	**16 882**	**65**	**18 640**	**6.56**

NATIONAL HEALTH LABORATORY SERVICE

Table 4: Provincial GeneXpert RIF Results in MTB detected cases (cumulative)

Province	Year	Inconclusive	Resistant	Sensitive	No Results	Total	% RIF Resistant
	/			1		**1**	
	2011	33	251	2957	53	**3 294**	**7.62**
	2012	213	1098	14647	134	**16 092**	**6.82**
Eastern Cape	2013	537	1566	20635	118	**22 856**	**6.85**
	2011	28	154	2661	1	**2 844**	**5.41**
	2012	163	739	10739	26	**11 667**	**6.33**
Free State	2013	175	458	7433	12	**8 078**	**5.67**
	2011	27	177	2907	1	**3 112**	**5.69**
	2012	137	765	10085	70	**11 057**	**6.92**
Gauteng	2013	308	929	12863	26	**14 126**	**6.58**
	/		2	9		**11**	
	2011	111	966	12033	62	**13 172**	**7.33**
	2012	465	2287	22628	386	**25 766**	**8.88**
Kwa-Zulu Natal	2013	451	1809	18660	114	**21 034**	**8.60**
	2011	27	159	1877	25	**2 088**	**7.61**
	2012	57	287	3820	75	**4 239**	**6.77**
Limpopo	2013	165	403	6588	78	**7 234**	**5.57**
	2011	31	211	2395	6	**2 643**	**7.98**
	2012	57	409	3513	76	**4 055**	**10.09**
Mpumalanga	2013	81	463	3497	19	**4 060**	**11.40**
	2011	40	304	3128	4	**3 476**	**8.75**
	2012	66	390	4704	14	**5 174**	**7.54**
North West	2013	124	357	5714	29	**6 224**	**5.74**
	2011	28	197	2637	2	**2 864**	**6.88**
	2012	64	273	4085	10	**4 432**	**6.16**
Northern Cape	2013	76	231	3752	285	**4 344**	**5.32**
	2011	15	106	2082	1	**2 204**	**4.81**
	2012	150	657	12393	2	**13 202**	**4.98**
Western Cape	2013	296	798	14688	1	**15 783**	**5.06**
Total		**3 925**	**16 446**	**213 131**	**1 630**	**235 132**	6.99

3. Rif Condordance

Rifampicin concordance is good for both LPA and culture. The data is skewed by reporting the GeneXpert immediately, but still have to wait for MGIT and LPA results.

Table 5: Rif Concordance by LPA or DST

Province	Rif Resistant Cases	GeneXpert Confirmation & Rif Concordance								
		MGIT				LPA				
		Confirmed		Rif Concordance		Confirmed		Rif Concordance		Inderterminate
		#	%	#	%	#	%	#	%	
Eastern Cape	2,500	40	1.6%	13	32.5%	222	9%	209	94.1%	1
Free State	1,105	51	4.6%	24	47.1%	230	21%	187	81.3%	45
Gauteng	1,729	68	3.9%	48	70.6%	328	19%	303	92.4%	9
Kwazulu-Natal	4,104	824	20.1%	781	94.8%	806	20%	719	89.2%	26
Limpopo	686	32	4.7%	27	84.4%	94	14%	68	72.3%	0
Mpumalanga	905	157	17.3%	152	96.8%	235	26%	210	89.4%	2
North West	714	33	4.6%	26	78.8%	133	19%	123	92.5%	8
Northern Cape	561	53	9.4%	38	71.7%	98	17%	81	82.7%	10
Western Cape	1,359	21	1.5%	2	0.0%	976	72%	927	95.0%	4
National	13,663	1,279	9.4%	1,111	86.9%	3,122	23%	2,827	90.6%	105

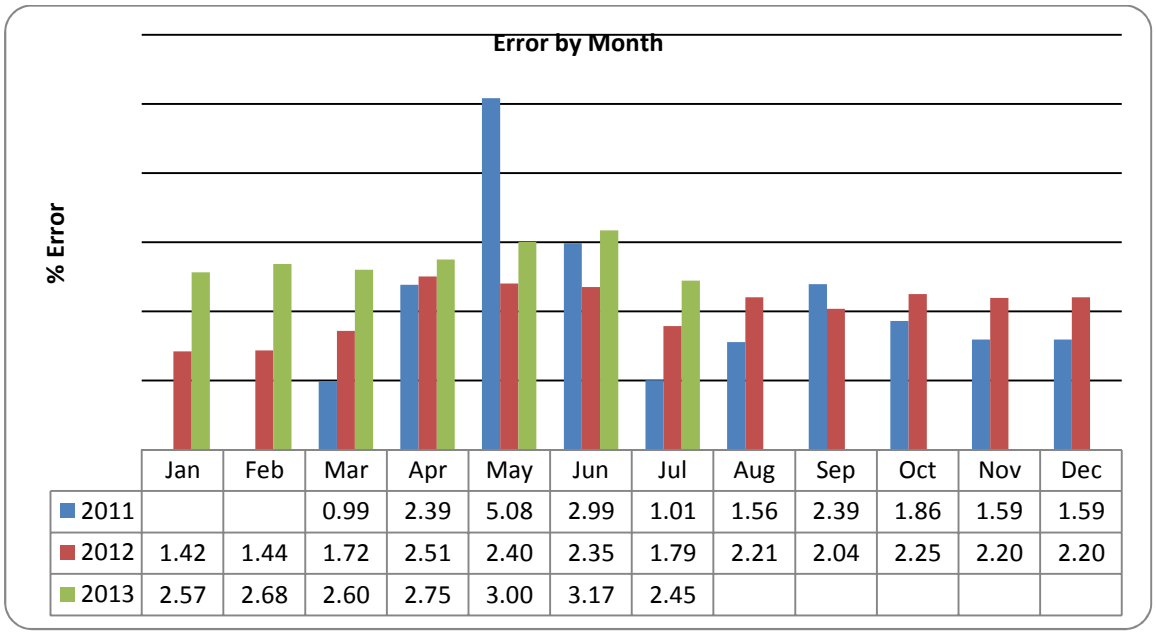
4. Errors

Average error rate has ranged consistently below 3%, however 3/9 provinces reported error rates above 3% in the month of July. 18 laboratories reported an increase in the number of errors due to hardware failures of the modules. Modules were replaced in 15 of the labs. Details of the invalid results, which likely represent sample issues remains below 1%. These are being monitored regularly and corrective action implemented where necessary.

Table 6: Number of Unsuccessful Tests and Reasons (1-31 July 2013)

Province	ERR	INV	NORES	MTB Result	Grand Total	% Error
Eastern Cape	740	114	73	32 964	33 891	2.18
Free State	149	32	4	17 170	17 355	0.86
Gauteng	743	133	23	22 111	23 010	3.23
Kwa-Zulu Natal	1 160	124	91	28 983	30 358	3.82
Limpopo	494	85	21	23 023	23 623	2.09
Mpumalanga	171	20	10	6 029	6 230	2.74
North West	398	37	13	10 413	10 861	3.66
Northern Cape	50	154	1	5 124	5 329	0.94
Western Cape	188	23	9	16 647	16 867	1.11
Grand Total	**4 093**	**722**	**245**	**162 464**	**167 524**	**2.44**

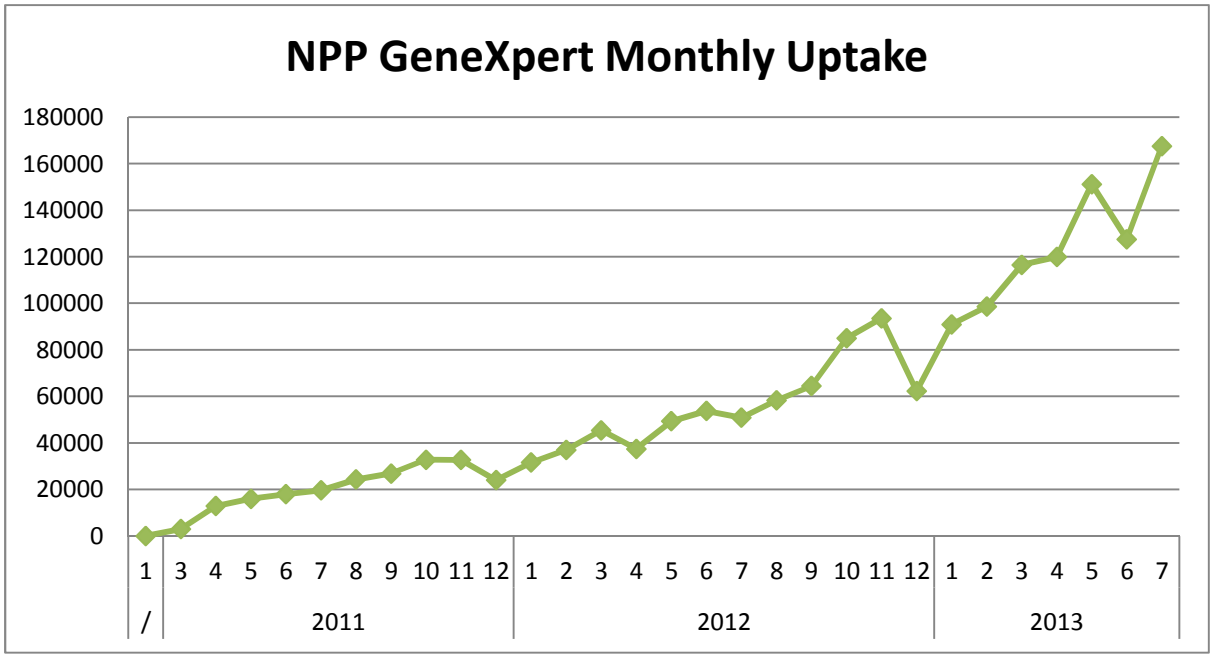
Figure 1: GeneXpert Error by Month



	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
2011			0.99	2.39	5.08	2.99	1.01	1.56	2.39	1.86	1.59	1.59
2012	1.42	1.44	1.72	2.51	2.40	2.35	1.79	2.21	2.04	2.25	2.20	2.20
2013	2.57	2.68	2.60	2.75	3.00	3.17	2.45					

5. Monthly uptake since implementation started

Figure 2: GeneXpert Monthly Uptake



Monthly uptake increased steadily since program inception. The main reason for interruptions is due to the variation in work practices which is expected during the December period. In addition, there

was a global shortage in the supply of Xpert MTB/RIF® cartridges in the months of July, October and November 2012. This was resolved in December 2012. Another shortage was experienced in March. The stock supply was stabilized in April. In addition Cepheid re-introduced the supply of 50 kit cartridges to high volume sites.

6. Further project phases as defined in the NTCM model

Phase I completed and reported on in the section above.

Phase IIa involves full capacitation of existing labs: Completed

Phase IIb: Full capacitation of high burden districts. Completed

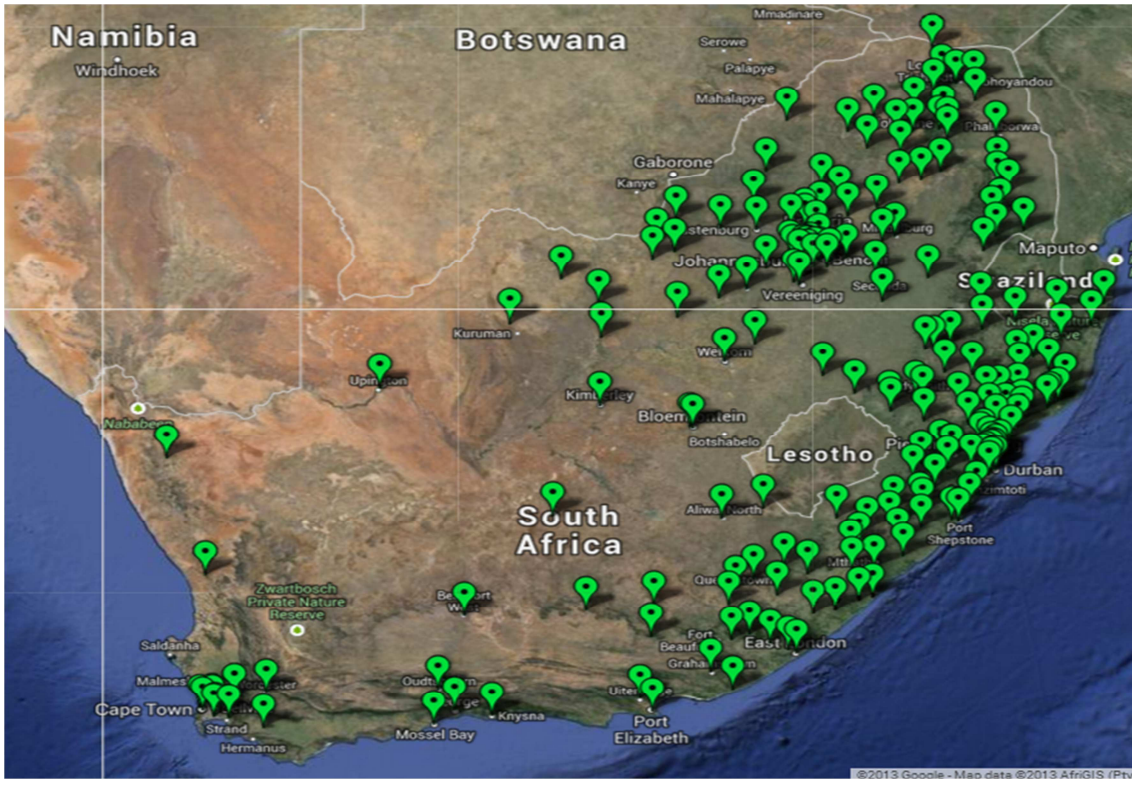
Phase IIIa and b: Gates funded study (Gauteng, EC and Free State). Phase 3a Completed

Phase IIIc: ensuring all districts have a minimum of 1 instrument per district: Completed

Phase IIId: Completion of all current microscopy and clinic sites: Completed

7. Phased Implementation Progress

Figure 3: Current GeneXpert Placement (207 testing centers, 274 **analysers, Gx4:** 88; Gx16-8: 1**; Gx16:** 183; GX48:1; Gx80-48: 1) *20 clinic placements



8. Training: Laboratory and Clinical

A total of 853 laboratory staff and 3,247 health care workers have been trained since December 2011. This will be an ongoing process to support NDoH training on clinical algorithm. Laboratory staff received both clinical and technical training.

9. Challenges identified during the course of the project to date

- Delay in training health care workers, especially doctors whose availability is limited, on clinical algorithm: is being addressed
- Rollout of EGK to avoid duplications
- Multiple specimens submitted for initial diagnosis using the GeneXpert in the Free State: being addressed with the provincial coordinator.

10. Literature Update For GeneXpert

There has been an expansion of the literature with respect to the assay performance. The highlights are summarized in the table below:

Table: Recent publications (GeneXpert for pulmonary TB and extrapulmonary TB)

Manuscript	Sample population and specimen type (n=…)	Results	
		Sensitivity	Specificity
Christopher, 2013, Eur Respir J	96 pleural fluids	Xpert on pleural fluid detected 4 out of 25 patients with confirmed TB resulting in a sensitivity of 16.0% (4/25; 95% confidence interval (CI 5-36%).	The specificity was 100%
Dowdy, 2013, PloS One	Compared the population-level impact of two WHO-endorsed strategies for improving the diagnosis of tuberculosis (TB): same-day microscopy and Xpert MTB/RIF	Same-day microscopy averted an estimated 11.0% of TB incidence over ten years Scaling up Xpert MTB/RIF to all centralized laboratories to achieve 75% population coverage had similar impact on incidence (9.3% reduction) and greater effect on mortality (23.8% reduction By the end of year ten, combining same-day microscopy and Xpert MTB/RIF could reduce annual TB mortality by 44% relative to the	

		current standard of care	
Bates et al, 2013 Trop Med Int Health.	Sputum samples from 94 patients from Zambia, with a primary obstetric or gynaecological condition	The Xpert had a sensitivity of 80.8% compared against MGIT culture was more sensitive than sputum smear microscopy (21/26 (80.8%) vs. 13/26 (50.0%)) and detected an additional eight culture-confirmed cases.	
Park et al, 2013 J Clin Microbiol.	320 consecutive respiratory specimens compared using acid-fast bacilli (AFB) staining, mycobacterial culture with both solid and liquid media, as well as the COBAS and Xpert assays	Compared to culture: • COBAS = 71.4% • Xpert = 67.9% The sensitivities of the COBAS assay for smear-positive and -negative specimens were 87% and 54%, while those of the Xpert were 67% and 69%, respectively	COBAS showed 100% specificity and 100% positive predictive value (PPV) regardless of smear results, while the Xpert assay showed 100% specificity and 100% PPV for smear-positive specimens but 98% specificity and 60% PPV for smear-negative specimens.
Kerkhof et al, 2013, PlosOne	N= 523 Paired sputum specimens were examined using liquid culture, fluorescence microscopy, and the Xpert MTB/RIF. Absolute neutrophil counts (ANC) were measured in blood samples	Multivariable analyses demonstrated that increasing sputum mycobacterial load was positively associated with blood ANC ≥2.6×10(9)/L and with neutrophilia. Increased blood neutrophil counts were independently associated with pulmonary TB and sputum mycobacterial burden in this HIV-infected patient group.	
Shah, 2013 BMC Infect Dis.	Conducted a detailed reference laboratory cost analysis of new rapid molecular assays (Xpert and MTBDRplus) for tuberculosis testing and drug-resistance testing in South Africa	From a laboratory perspective, Xpert MTB/RIF cost $14.93/sample and the MTBDRplus line probe assay cost $23.46/sample, compared to $16.88/sample using conventional automated liquid culture-based methods. Laboratory costs of Xpert and MTBDRplus were most influenced by cost of consumables (60-80%).	
Somoskovi A et al, 2013 J Clin Microbiol. (Abstract only)	Report on false negative results of the assay in the presence of rpoB Leu533Pro that is associated with low level of phenotypic rifampin resistance.		

11. Update on GeneXpert Research projects:

11.1. *GeneXpert Verification and EQA program using Dried Culture spots (DCS)*

- Preparation of verification DCS for the 3rd quarter of Gx implementation are complete and in the process of being sent to sites.
- EQA Panels for the 2nd round of EQA are being prepared for September delivery to NHLS sites.
- TBGxMonitor™ (www.tbgxmonitor.com) is about to undergo an upgrade to include EQA reporting functionality for national and international EQA programs along with a minor functionality upgrade.

11.2. *Diagnosis of Extrapulmonary TB (EPTB) using the GeneXpert MTB/RIF*

A study to develop protocols for diagnosing EPTB from alternative specimen types has been completed. Finding showed good sensitivity for the Xpert on purulent specimens but poor sensitivity for clear watery fluids (pre-submission letter sent to PLoSMedicine). Based on these findings, a new study is underway:

- To determine whether a modified GeneXpert protocol which will not involve addition of SR buffer, can be used to increase the diagnostic sensitivity of the Xpert MTB/Rif assay for clear watery fluid types among aspirates and fluids.

11.3. *Connectivity solutions for the GeneXpert*

- Connectivity: Collaboration with Cepheid ongoing
    i. Remote connectivity – System deployed on more than 100 sites by Cepheid and the NHLS. More than 740,000 results reported to date. The dashboard has been updated to cope with the South African rollout capacity. A deadline has been set for the 15th of September to have NHLS IT have upgraded all existing installations to be included on the dashboard.
    ii. The Cepheid remote calibration should also be fully functional by the 15th of September.

12. Update on other projects

Grand Challenges Canada project: Multiple POC HIV/TB integration feasibility project

GCC is a three year project to investigate the feasibility of integrating multiple POC testing for HIV and TB (using the Xpert MTB/RIF test) integration of services in an active ARV

treatment clinic. This will involve a randomized controlled trial at 3 clinic sites to compare standard of care and Point of care (ongoing).

- *Sub-studies within GCC*
    - Investigating alternative media for blood specimen collection/storage and transport. This study will compare viral load testing on Dried Blood Spots (DBS, gold standard) to new technologies/alternatives such as Hemaform plates, Primestore tubes and a thicker DBS cards.
    - Validation of a rapid HIV/Syphilis DUO test. A laboratory validation of a new rapid strip for simultaneous detection of HIV and syphilis infection.
    - Validation of a new POC chemistry system. A laboratory validation of the a new POC blood analysis system, the Epoc (Alere) will begin in Oct.

13. Grants Submitted

None

14. Funding

Table 9: Total and Percentage Contribution to date by Donor

Donor	% Contribution
NDoH	24.04
Bill & Melinda Gates Foundation	7.20
TB Reach	1.42
MSF	0.90
FIND	0.45
USAID	2.45
CDC NHLS 2010/11	14.78
CDC NDoH	0.72
CDC NHLS 2011/12	1.39
Dr. Niebauer	0.20
Gobal Fund NDOH	40.91
Global Fund RTC	2.78
CDC NDoH	2.77
Subtotal	**100**

CDC has contributed 19, 65% towards the program to date.

15. Recent Campaigns

None in July