NATIONAL HEALTH LABORATORY SERVICE

GeneXpert MTB/RIF

Progress Report

June 2013

NATIONAL HEALTH LABORATORY SERVICE

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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.

The remainder of the roll-out is being performed in a phased manner 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 1,579,649 specimens have been processed to date (30 June 2013). In June 127,110 specimens were processed. The total % of *Mycobacterium tuberculosis* complex (MTBC) detected in this cohort was 10.87% (13,822). 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
Eastern Cape	2011	3294	15413	555	19 262	17.10
Eastern Cape	2012	16092	85782	2893	104 767	15.36
Eastern Cape	2013	18902	123490	4476	146 868	12.87
Free State	2011	2844	14830	33	17 707	16.06
Free State	2012	11667	77094	280	89 041	13.10
Free State	2013	6666	58444	485	65 595	10.16
Gauteng	2011	3102	18890	424	22 416	13.84
Gauteng	2012	11057	72748	2285	86 090	12.84
Gauteng	2013	11270	80911	3702	95 883	11.75
Kwa-Zulu Natal	2011	13172	47523	1729	62 424	21.10
Kwa-Zulu Natal	2012	25766	144201	6233	176 200	14.62
Kwa-Zulu Natal	2013	17349	115018	7070	139 437	12.44
Limpopo	2011	2088	17870	173	20 131	10.37
Limpopo	2012	4239	31469	700	36 408	11.64
Limpopo	2013	5884	68242	2990	77 116	7.63
Mpumalanga	2011	2643	12769	1107	16 519	16.00
Mpumalanga	2012	4055	22020	1122	27 197	14.91
Mpumalanga	2013	3112	17685	824	21 621	14.39
North West	2011	3476	14887	657	19 020	18.28
North West	2012	5174	29003	1977	36 154	14.31
North West	2013	5015	36484	2211	43 710	11.47
Northern Cape	2011	2864	16117	735	19 716	14.53
Northern Cape	2012	4432	23654	1192	29 278	15.14
Northern Cape	2013	3686	22844	1269	27 799	13.26
Western Cape	2011	2204	10093	31	12 328	17.88
Western Cape	2012	13202	68427	596	82 225	16.06
Western Cape	2013	13183	69995	1513	84 691	15.57
Total		**216 450**	**1 315 936**	**47 263**	**1 579 649**	**13.70**

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Table 2: GeneXpert MTB Results by province (01-30 June 2013)

Province	MTB Detected	MTB Not Detected	Test Unsuccessful	Grand Total	% MTB Detected
Eastern Cape	3 085	23 397	1 073	27 555	11.20
Free State	973	10 520	88	11 581	8.40
Gauteng	2 045	15 565	844	18 454	11.08
Kwa-Zulu Natal	2 535	17 976	1 207	21 718	11.67
Limpopo	962	14 406	623	15 991	6.02
Mpumalanga	578	3 455	165	4 198	13.77
North West	788	6 307	568	7 663	10.28
Northern Cape	479	3 222	191	3 892	12.31
Western Cape	2 377	13 436	245	16 058	14.80
Grand Total	**13 822**	**108 284**	**5 004**	**127 110**	**10.87**

Table 3: Provincial GeneXpert RIF Results in MTB detected cases (01-30 June 2013)

Row Labels	Inconclusive	Resistant	Sensitive	No Results	Grand Total	% Rif Resistance
Eastern Cape	98	210	2 759	18	3 085	6.81
Free State	16	61	895	1	973	6.27
Gauteng	64	139	1 840	2	2 045	6.80
Kwa-Zulu Natal	76	231	2 213	15	2 535	9.11
Limpopo	42	45	873	2	962	4.68
Mpumalanga	12	64	502		578	11.07
North West	14	37	737		788	4.70
Northern Cape	9	28	442		479	5.85
Western Cape	59	116	2 202		2 377	4.88
Grand Total	**390**	**931**	**12 463**	**38**	**13 822**	**6.74**

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

Province	Year	Inconclusive	Resistant	Sensitive	No RIF Result	Total	% RIF Resistant
Eastern Cape	2011	33	251	2957	53	**3 294**	**7.62**
Eastern Cape	2012	213	1098	14647	134	**16 092**	**6.82**
Eastern Cape	2013	419	1326	17041	116	**18 902**	**7.02**
Free State	2011	28	154	2661	1	**2 844**	**5.41**
Free State	2012	163	739	10739	26	**11 667**	**6.33**
Free State	2013	150	375	6133	8	**6 666**	**5.63**
Gauteng	2011	27	177	2897	1	**3 102**	**5.71**
Gauteng	2012	137	765	10085	70	**11 057**	**6.92**
Gauteng	2013	234	746	10268	22	**11 270**	**6.62**
Kwa-Zulu Natal	2011	111	966	12033	62	**13 172**	**7.33**
Kwa-Zulu Natal	2012	465	2287	22628	386	**25 766**	**8.88**

Kwa-Zulu Natal	2013	361	1482	15401	105	**17 349**	**8.54**
Limpopo	2011	27	159	1877	25	**2 088**	**7.61**
Limpopo	2012	57	287	3820	75	**4 239**	**6.77**
Limpopo	2013	122	349	5374	39	**5 884**	**5.93**
Mpumalanga	2011	31	211	2395	6	**2 643**	**7.98**
Mpumalanga	2012	57	409	3513	76	**4 055**	**10.09**
Mpumalanga	2013	54	363	2680	15	**3 112**	**11.66**
North West	2011	40	304	3128	4	**3 476**	**8.75**
North West	2012	66	390	4704	14	**5 174**	**7.54**
North West	2013	93	302	4592	28	**5 015**	**6.02**
Northern Cape	2011	28	197	2637	2	**2 864**	**6.88**
Northern Cape	2012	64	273	4085	10	**4 432**	**6.16**
Northern Cape	2013	67	189	3145	285	**3 686**	**5.13**
Western Cape	2011	15	106	2082	1	**2 204**	**4.81**
Western Cape	2012	150	657	12393	2	**13 202**	**4.98**
Western Cape	2013	242	657	12284		**13 183**	**4.98**
Total		**3 454**	**15 221**	**196 209**	**1 566**	**216 450**	**7.03**

3. Rif Condordance

Rifampicin concordance is good for both LPA and culture. There is significant regional variation in Rifampicin mono-resistance. The national average is 12% for DST and 17% for LPA. This could be attributed to a number of factors such as geographical variation, laboratory variation, interpretation of LPA, reliability of gold standard or even strain variation.

Testing and clinical algorithms show variation across provinces, requiring standardisation as this leads to significant confusion in all aspects of the testing cycle, as well as in some cases, being more onerous to the TB patients themselves.

Table 5: Rif Concordance by LPA or DST

		GeneXpert Confirmation & Rif Concordance									
Province	Rif Resistant Cases	DST					LPA				
		Confirmed		Rif Concordance		Pre-analytical	Confirmed		Rif Concordance		Inderterminate
		#	%	#	%		#	%	#	%	
Eastern Cape	1153	47	4.1%	10	21.3%	0	46	4%	45	97.8%	1
Free State	724	15	2.1%	7	46.7%	11	79	11%	64	81.0%	14
Gauteng	895	21	2.3%	16	76.2%	21	90	10%	84	93.3%	2
Kwazulu-Natal	2726	686	25.2%	652	95.0%	0	631	23%	509	80.7%	28
Limpopo	380	28	7.4%	27	96.4%	1	44	12%	39	88.6%	0
Mpumalanga	514	81	15.8%	78	96.3%	1	131	25%	111	84.7%	2
North West	435	8	1.8%	7	87.5%	2	50	11%	47	94.0%	6
Northern Cape	343	24	7.0%	17	70.8%	8	55	16%	47	85.5%	8
Western Cape	782	1	0.1%	0	0.0%	3	235	30%	234	99.6%	0
National	7 952	911	11.5%	814	89.4%	47	1 361	17%	1 180	86.7%	61

4. Errors

Average error rate has ranged consistently below 3%, however 5/9 provinces reported error rates above 3% in the month of June. A couple of laboratories experienced an increase in the number of errors related to cartridges on lot numbers 11305A, 11114A, 111709A and 11108A. Error log reports were downloaded on disc and submitted to Cepheid for investigation. The lost cartridges have been replaced in other labs and others still pending analysis.

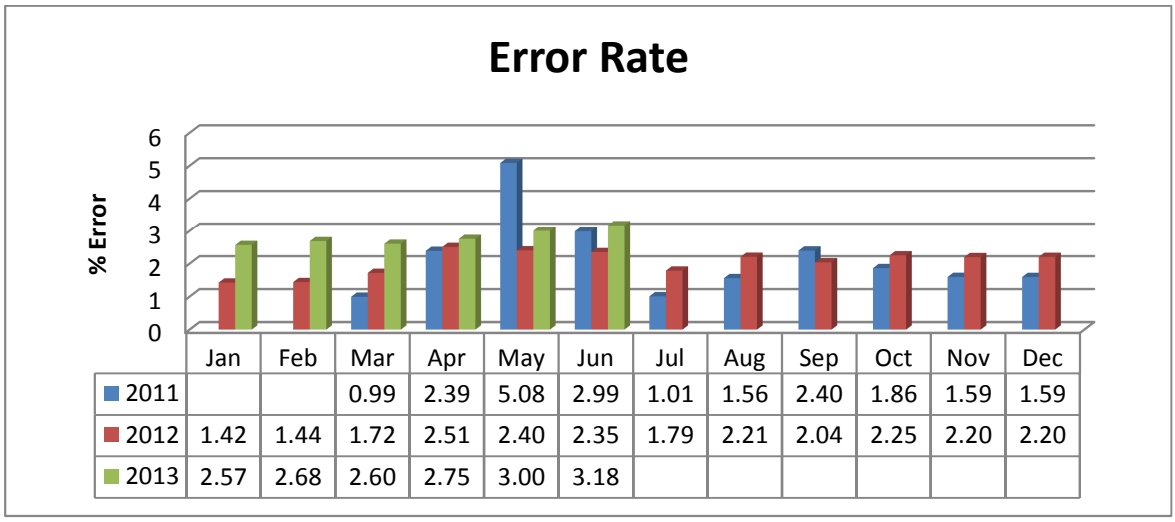
In addition 8 laboratories reported an increase in the number of errors due to hardware failures of the modules. Modules were replaced in all the labs. Details of the invalid results, which likely represent sample issues remains below 1%. These are being monitored regularly and corrective action implemented where necessary.

Table 6: Number of Unsuccessful Tests and Reasons

Province	Year	ERR	INV	NORES	No Raw Result	MTB Result	Total	% Error
Eastern Cape	2011	501	47	6	1	18707	**19 262**	**2.60**
Eastern Cape	2012	2552	198	126	17	101874	**104 767**	**2.44**
Eastern Cape	2013	3375	551	546	4	142392	**146 868**	**2.30**
Free State	2011	27			6	17674	**17 707**	**0.15**
Free State	2012	229	21	26	4	88761	**89 041**	**0.26**
Free State	2013	388	68	27	2	65110	**65 595**	**0.59**
Gauteng	2011	371	47	6		21992	**22 416**	**1.66**
Gauteng	2012	2043	187	55		83805	**86 090**	**2.37**
Gauteng	2013	3207	336	157	2	92181	**95 883**	**3.34**
Kwa-Zulu Natal	2011	1147	541	39	2	60695	**62 424**	**1.84**
Kwa-Zulu Natal	2012	5132	665	436		169967	**176 200**	**2.91**
Kwa-Zulu Natal	2013	5776	603	685	6	132367	**139 437**	**4.14**
Limpopo	2011	134	28	10	1	19958	**20 131**	**0.67**
Limpopo	2012	587	102	11		35708	**36 408**	**1.61**
Limpopo	2013	2551	348	91		74126	**77 116**	**3.31**
Mpumalanga	2011	1027	73	6	1	15412	**16 519**	**6.22**
Mpumalanga	2012	1040	65	17		26075	**27 197**	**3.82**
Mpumalanga	2013	723	71	29	1	20797	**21 621**	**3.34**
North West	2011	616	41			18363	**19 020**	**3.24**
North West	2012	1752	142	83		34177	**36 154**	**4.85**
North West	2013	2001	146	64		41499	**43 710**	**4.58**

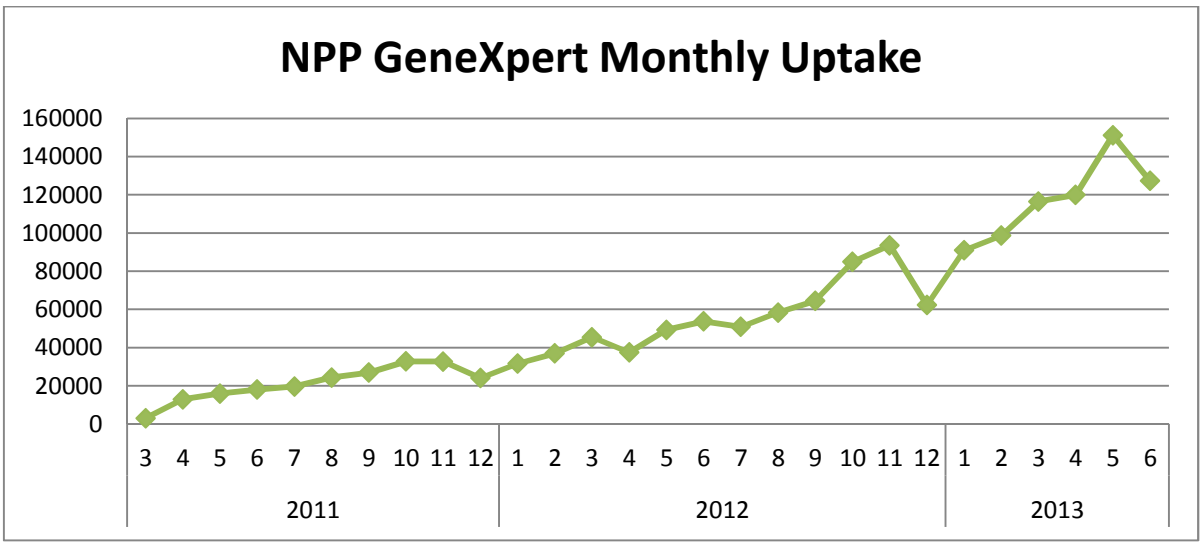
Northern Cape	2011	557	129	14	35	18981	**19 716**	**2.83**
Northern Cape	2012	87	123	9	973	28086	**29 278**	**0.30**
Northern Cape	2013	480	336	30	423	26530	**27 799**	**1.73**
Western Cape	2011	26	5			12297	**12 328**	**0.21**
Western Cape	2012	540	38	18		81629	**82 225**	**0.66**
Western Cape	2013	1350	126	37		83178	**84 691**	**1.59**
Total		**38 220**	**5 037**	**2 528**	**1 478**	**1 532 386**	**1 579 649**	**2.42**

Figure 1: GeneXpert Error by Month



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: In Progress

Phase IIId: Completion of all current microscopy and clinic sites: In Progress

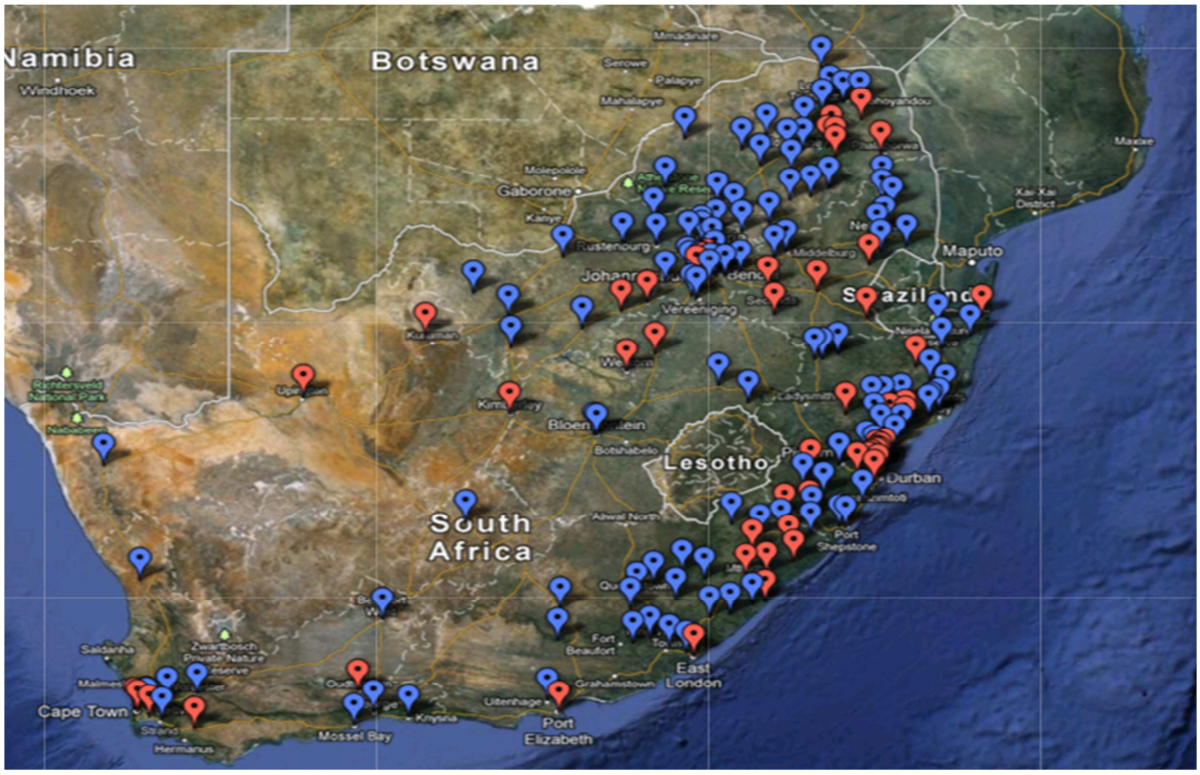
7. Phased Implementation Progress

Table 7: Phased Implementation Progress

Phase	GX4	GX16	GX48	TOTAL	Placed	% Completion
Phase 1/2a	7	30	1	38	38	100
Phase 2b	22	23	1	46	46	100
Phase 3a	3	10	0	13	13	100
Phase 3b	2	11	0	13	13	100
Phase 3c	6	28	0	34	34	100
Phase 3d	41	83	0	124	98	79
TOTAL	**81**	**185**	**2**	**268**	**242**	**90**

To date implementation is 90% complete.

Figure 3: Current GeneXpert Placement (175 testing centers, 242 analysers, Gx4: 77; Gx16-8: 1; Gx16: 162; GX48:1; Gx80-48: 1) ***20 clinic placements**



8. Training: Laboratory and Clinical

A total of 822 laboratory staff and 2,783 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
- Laboratories using GXP for monitoring treatment (and not just diagnosis): is being addressed through training

10. Literature Update For GeneXpert

There has been an expansion of the literature with respect to the assay performance. The highlights are summarized in table 11 below:

Table 8: Recent publications (GeneXpert for pulmonary TB and extrapulmonary TB)

Manuscript	Sample population and specimen type (n=…)	Results	
		Sensitivity	Specificity
Hanrahan, PLoS one 2013	N=641 tuberculosis suspects at a primary care clinic in Johannesburg	Among 116 individuals diagnosed with TB, 66 (57%) were Xpert negative, of which 44 (67%) were empirical or radiological diagnoses and 22 (33%) were Xpert negative/culture-positive. The median time to tuberculosis treatment: • 0 days (IQR: 0-0) for Xpert positives, • 14 days (IQR: 5-35) for those diagnosed empirically, • 14 days (IQR: 7-29) for radiological diagnoses, • 144 days (IQR: 28-180) for culture positives	
Antonenka et al, BMC Infect Dia, 2013	121 pre-characterized respiratory specimens tested for the presence of MTB complex by the three assays: • Xpert MTB/RIF • ProbeTec ET DTB (DTB) (Becton-Dickinson) • COBAS TaqMan MTB (CTM-MTB) (Roche).	Overall sensitivity for detection of MTB complex in culture positive samples: • 74.6% Xpert MTB/RIF • 73.8% CTM-MTB • 79.1% DTB	Specificity was best for CTM-MTB (100%) and lowest for Xpert MTB/RIF (96.2%)

11. Update on GeneXpert Research projects:

- ~1700 Dried Culture Spots (DCS) for verification of GeneXperts for quarter 3 of implementation are in preparation.
    - **i.** A manuscript has been submitted to JCM on the performance monitoring of the DCS product
- DCS for EQA program: EQA panels have been prepared for =144 NHLS sites (64 Gx4; 133 Gx16; 2 Gx48). These have been shipped out. Result submission has been

extended to the end of July as some sites did not receive the correct number of panels. To date, 104 reports have been issued.

- DCS for ACTG sites: EQA panels have been shipped to n=23 international ACTG sites (27Gx4; 2Gx16; 1Gx80). This includes all three EQA rounds for the whole year. To date 22 ACTG sites have already submitted their EQA results.
- Awaiting CDC approval of TB EQA 5 matrix evaluation manuscript.

- TBGxMonitor™ (www.tbgxmonitor.com) automated GeneXpert Verification and EQA reporting platform has been upgraded to include full EQA report processing. Both Verification and EQA components have been completed. The next major upgrade Phase 3 has been completed and is currently live. The new site automatically releases all reports in real-time for both Verification and EQA. Currently the site has processed over 100 EQA reports for NHLS laboratories in the first GeneXpert EQA round for 2013 which is still ongoing. This site is also currently serving the NHLS / Wits EQA provided to the ACTG Group with laboratories in 15 countries.
- Alternative specimen preparation protocols:
    i. Protocols being developed for TB diagnosis in children. A manuscript is underway on peadiatric TB diagnosis using the GeneXpert.
    ii. The evaluation of 1175 EPTB specimens has been performed and data analysed. The table below summarises the overall sensitivity and specificity of Xpert performance across all specimens. This has further been analysed by specimen type, sample volume, sample viscosity, appearance, requirement of centrifugation etc. the manuscript is underway. This data set was also submitted to the WHO as part of the metanalysis

Xpert assay perfromance *Total sample number*	MGIT culture n=1175	Xpert	Sensitivity (Culture Reference)	Specificity (Culture Reference)
M.tb positive, n(%)	277 (23.5%)	260 (22%)		
M.tb negative, n(%)	774 (65.9%)	909 (77.4%)	n=1045	
Contaminated/ error, n(%)	124 (10.5%)	6 (0.5%)	59% (53, 65)	92% (90, 94)
MTB sensitive, n		231		
MTB Rif resistant, n		25		
MTB Rif indeterminate, n		4		

- Connectivity: Collaboration with Cepheid ongoing
    i. Remote connectivity – System deployed on more than 100 sites by Cepheid and the NHLS. More than 340,000 results reported to date. The current pilot system cannot handle the additional testing capacity which will be addressed in the full product version. Discussions are currently under way to include the remainder of the NHLS sites on the system, purge the data and begin monitoring again to assist in the evaluation of the ongoing rollout.
    ii. The first point of care site (Botshabelo Clinic, North West Province) has gone live on the Cepheid Dashboard with an additional 2 sites to be connected. These sites are using Metacom-sponsored routers (3G) connection for reporting.

12. HIV/TB Integration

- Grand Challenges Canada project: Multiple POC HIV/TB integration feasibility project
    - Phase I complete
    - Phase II: Evaluation of nurse operated POC versus routine lab completed at HJH Themba Lethu clinic (n=326) complete.
        - Manuscript in progress.
    - RCT: ~n=452 patients (POC arm =226; SOC =226) recruited into the study.
        - An interim analysis of turnaround times indicates that:
        - 75% of specimens are collected from the clinic and received at the laboratory within one day
        - 17% of lab results have a same day turnaround time, with 68% completed in one day
        - Once laboratory results are printed, 17% are stamped in the clinic within the same day and 55% a day later
    - Sub-study 1: to investigate feasibility and patient acceptance of multiple finger sticks for POC testing: Completed. Awaiting re-submission.
    - Sub-study 2: to investigate various blood specimen storage and transport options: This study will compare viral load testing on Dried Blood Spots (DBS) to new

technologies/alternatives such as Hemaform plates, Primestore tubes and a thicker DBS cards.

- Patient recruitment has begun at Themba Lethu Clinic, n=15 to date.

- Sub-study 2: to investigate volumes of blood collected from a finger stick for point of care testing:
  - This is in collaboration with Northwestern University
  - Patient recruitment at Themba Lethu Clinic is complete, n=100
  - Data analysis is underway.

- Connectivity:
  - Conworx (POCcelerator) and LDS (AegisPOC) to be trialed in 2 sites during RCT. AegisPOC was installed at the first connectivity on 15 September, 2012. The Conworx solution was installed on the 14th of December, 2012. An antenna was installed and sufficiently boosted the signal. Both systems are currently running. The connectivity down time experienced at Tigane was resolved.
  - A preliminary evaluation and comparison of the systems is about to commence as part of the study outputs. The proposed evaluation includes the option to switch the control site (paper-based) to one of the live systems in order to document and measure the impact on workflow before and after the installation of the system. This proposal is being discussed.

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 June