Performance of QMAC DST™, rapid drug susceptibility testing system for *Mycobacterium tuberculosis* integrated with MGIT960 culture

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INTRODUCTION

- Due to the lack of a rapid drug susceptibility test (DST) to determine the appropriate drug prescription for the tuberculosis patients, we applied a microfluidic agarose channel system that can reduce turnaround time of the anti-microbial susceptibility test through a time-lapse imaging of bacterial growth.
- Recently, the QMAC DST™ have been developed as rapid DST system based on automated microscopic technology tracking growth of *Mycobacterium tuberculosis* (MTB).
- Drug susceptibility test(DST) for many kinds of anti-tuberculosis drugs can be completed within 7days.
- Previously, we have developed the QMAC DST™ system to use cultured colonies from solid media that requires longer culture time of 4weeks compared to 2 weeks of liquid culture system.
- In this study, we developed optimized test process for integrating with the MGIT 960 to reduce overall turnaround time of DST and aimed to evaluate the performance of QMAC DST™ integrated MGIT 960.

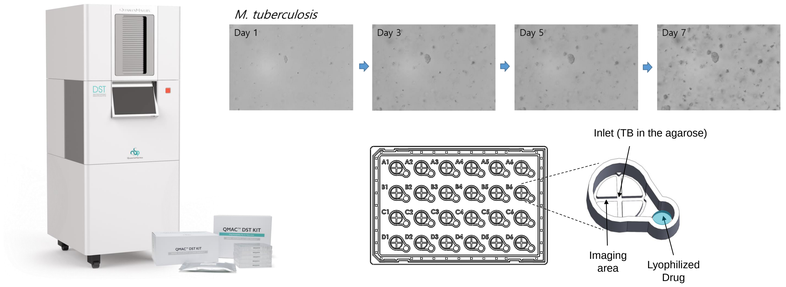

Figure 1. The fully automated QMAC DST™ system performs DST based MTB time lapse imaging for reliable DST results and schematic of the CAT(cross agarose channel for tuberculosis) chip containing 13 anti-TB lyophilized drugs.

METHODS

- Liquid cultured MTB suspensions were dispersed with glass beads and were mixed with 0.5% agarose at 37°C by vortexing.
- Agarose mixture with MTB cell suspension were loaded into each well of QMAC DST™ chips where every anti-tuberculosis drug is lyophilized at critical concentrations.
- Middlebrook 7H9 broth with 10% OADC is added to each well. Prepared QMAC DST™ chips were completely sealed incubated at 37°C for 7 days.
- MTB growth are automatically captured with a 10× lens on an inverted microscope every other day for imaging analysis.

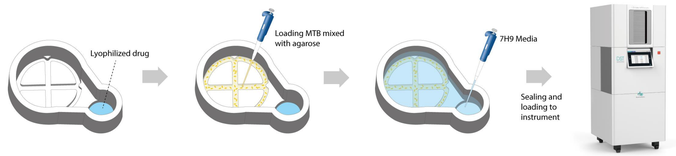

Figure 2. Inoculation and test procedure: i) Loading MTB cells mixed with agarose, ii) introducing 7H9 media to make lyophilized drug dissolved and diffused into the matrix, iii) time-lapse imaging in the instrument.

- Seventeen clinical sputum and forty spiked liquid cultures were used for preliminary study of QMAC DST™ on 11 anti-tuberculosis drugs. After process verification for QMAC DST™ integrated with MGIT960, 124 clinical sputum and 30 spiked liquid cultures were tested on 13 drugs.

Table 1. Antibiotics panel list : 4 first line drugs and 9 second line drugs, multi-concentrations for first line drugs

1st line drug	Streptomycin, Ethambutol, Isoniazid, Rifampicin
2nd line drug	Ofloxacin, Amikacin, Moxifloxacin, Capreomycin, Levofloxacin, Para-aminosalicylic acid, Kanamycin, Rifabutin, Ethionamide

RESULTS

- Previously, there were 96.2% overall agreement rates between the QMAC DST using cultured colonies solid media between using conventional method.
- In preliminary study for system verification, the QMAC DST™ integrated MGIT 960 showed overally 93.7% agreement rates compared with the DST results of the conventional method.
- In 154 clinical test, the QMAC DST™ integrated MGIT 960 showed overally 97.96% agreement rates compared with the DST results of the conventional method.
- All drugs were compared with the DST results using the Löwenstein-Jensen medium to classify the agreement/discrepancy, and the agreement rates of S and R strains were calculated for each drug.

Table 2. Comparison of susceptibility/resistant agreement between M-kit and QMAC DST™ for 13 drug tested

Conventional Method (M-kit)				QMAC DST System							
Drug	Concentration	S	R	Concentration	Susceptible Result		Resistant Result		Total Result		
					#Agreement	% Agreement	#Agreement	% Agreement	#Agreement	% Agreement	
Isoniazid	0.2	110	44	0.1	107	97.27	44	100.00	151	98.05	
Rifampicin	40	122	32	1	122	100.00	29	90.63	151	98.05	
Ethambutol	2	127	27	5	125	98.43	23	85.19	148	96.10	
Streptomycin	10	123	31	2	116	94.31	31	100.00	147	95.45	
Amikacin	30	144	10	1	144	100.00	10	100.00	154	100.00	
Ofloxacin	2	142	12	2	142	100.00	12	100.00	154	100.00	
Moxifloxacin	2	142	12	0.5	142	100.00	11	91.67	153	99.35	
Levofloxacin	2	142	11	1.5	142	100.00	10	90.91	152	99.35	
Kanamycin	30	143	10	2.5	143	100.00	10	100.00	153	100.00	
Rifabutin	20	125	29	0.5	124	99.20	29	100.00	153	99.35	
Para-amino salicylic acid	1	142	12	4	138	97.18	12	100.00	150	97.40	
Capreomycin	40	143	11	2.5	143	100.00	9	81.82	152	98.70	
Ethionamide	40	141	13	5	126	89.36	13	100.00	139	90.26	
Total		1900	254		1867	**98.26**	243	**95.67**	2110	**97.96**	

CONCLUSIONS

QMAC DST showed successful integration with MGIT 960 as demonstrating high agreement results compared to the conventional DST method. It allows reducing overall turnaround time from liquid culture to QMAC DST within 3 weeks compared to 8 weeks of conventional solid culture and conventional DST methods. Further studies are on going to yield maximized diagnostic accuracy compared to standard DST methods and molecular characteristics.

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