

An Automated Culture System for Use in Preclinical Testing of Host-Directed Therapies for Tuberculosis

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Materials

Name	Company	Catalog Number	Comments
IX51 Fluorescent Microscope	Olympus, Japan	N/A	AFB detection and imaging
2 mL microtube, flat bottom, screw cap, sterile	Sarstedt, North Carolina, USA	72.694.006	Mtb infection of macrophages
5 mL syringe, Luer lock	BD Biosciences, San Jose, CA, USA	SZR-150-031K	Mtb infection of macrophages/CFU
50 mL tube, sterile	Sarstedt, North Carolina, USA	62.547.254	Mtb infection of macrophages
all-trans-Retinoic Acid (ATRA) ≥98% (HPLC)	Sigma Aldrich, Missouri, USA	R2625	Host directed therapy candidate
BacT/ALERT 3D Microbial Detection System	Biomerieux (Hampshire, UK)	247001	Broth-based colorimetric detection system
BACT/ALERT MP BACT/ALERT MP Nutrient Supplement	Biomerieux (Hampshire, UK)	414997	Broth-based colorimetric detection assay
BACT/ALERT MP culture bottles	Biomerieux (Hampshire, UK)	419744	Broth-based colorimetric detection assay
BD BBL Middlebrook ADC Enrichment, 20 mL	BD Biosciences, San Jose, CA, USA	M0553	Mycobacterium liquid culture
BD BBL Middlebrook OADC Enrichment, 20mL	BD Biosciences, San Jose, CA, USA	M0678	Colony Forming Units
Cell scraper, 25 cm	Sarstedt, North Carolina, USA	83.1830	Harvest of Imacrophage lysates
Corning Syringe Filter, 0.2 µm	Corning Incorporated, Germany	431219	Sterilization of lysis buffer
Cover glass (borosilicate), 24 x 50 mm, #1.5 thickness	VWR International Limited	631 - 0147	
Cycloheximide, from microbial	Sigma Aldrich, Missouri, USA	C7698	Colony Forming Units
Dako Fluorescent Mounting Medium	Agilent Technologies Ireland Limited	S3023	Antifade mounting medium
Dulbecco's Phosphate Buffered Saline	Sigma Aldrich, Missouri, USA	D8537	Mtb infection of macrophages
Fetal Bovine Serum, Gibco	Thermo Fisher, Massachusetts, USA	10270106	Macrophage cell culture
Glycerol, Difco	BD Biosciences, San Jose, CA, USA	228220	Colony Forming Units
Hoescht 33342 (bisBenzimide H 33342 trihydrochloride)	Sigma Aldrich, Missouri, USA	B2261	Nuclear stain

IFN γ , recombinant human	R&D Systems Inc, Minnesota, USA	285-IF	Host directed therapy candidate
Labtek 2-well chamber slide, sterile, Nunc	Thermo Fisher, Massachusetts, USA	TKT-210-150R	Mtb infection of macrophages
L-Asparagine, anhydrous	Sigma Aldrich, Missouri, USA	A4159	Colony Forming Units
Linezolid	Sigma Aldrich, Missouri, USA	PZ0014	Antibiotic
Microlance Hypodermic Needle 25 G	BD Biosciences, San Jose, CA, USA	300400	Mtb infection of macrophages/CFU
Middlebrook 7H10 Agar Base	BD Biosciences, San Jose, CA, USA	M0303	Colony Forming Units
Middlebrook 7H9 Broth Base	BD Biosciences, San Jose, CA, USA	M0178	Mycobacterium liquid culture
Modified Auramine O Stain and Decolourizer	Scientific Device Laboratory, IL, USA	345-250	AFB stain
Paraformaldehyde	Sigma Aldrich, Missouri, USA	158127	Mtb infection of macrophages
Petri dishes, 92 x 16mm (20/bag)	Sarstedt, North Carolina, USA	82.1473.001	Colony Forming Units
Phorbol 12-myristate 13-acetate (PMA)	Sigma Aldrich, Missouri, USA	P8139	Macrophage cell culture
Polysorbate 80, Difco	BD Biosciences, San Jose, CA, USA	231181	Mycobacterium liquid culture
RPMI-1640, Gibco	Thermo Fisher, Massachusetts, USA	52400025	Macrophage cell culture
Sterile Cell Spreader, L-Shaped	Fisherbrand, Thermo Fisher, MA, USA	RB-44103	Colony Forming Units
T25 TC flask, angled neck, filter cap, sterile, Nunc	Thermo Fisher, Massachusetts, USA	156367	Mycobacterium liquid culture
THP-1 cell line	ATCC, Virginia, USA	ATCC TIB-202	Macrophage cell culture