DEPARTMENT OF MICROBIOLOGY
SETH G.S MEDICAL COLLEGE AND K.E.M HOSPITAL
SERVICES

LABORATORY WORKING HOURS

The working hours, for the various divisions and specimen acceptance timings are provided in the tables below.

Routine working hours – All divisions	Weekdays	9.00 a.m. to 4.00 p.m.
	Saturdays & Bank Holidays	9.00 a.m. to 12.30 p.m.
Emergency laboratory Services	Weekdays	4.00 p.m. to next day 9.00 a.m.
	Saturdays / Bank Holidays	12.30 p.m. to Sunday / Next working day 9.00 a.m.
	Sundays / O.P.D Holidays	9.00 a.m. to Monday / Next working day 9.00 a.m.

SPECIMEN ACCEPTANCE TIMINGS

	Division	Timing
OPD patients	All divisions	9.00 a.m. – 11.00 a.m.
Indoor patients	All divisions	9.00 a.m. – 11.00 a.m.
Body fluids / Aspirated pus/ Tissue / Ocular specimens / Stool for cholera	Clinical Bacteriology, Mycology and Mycobacteriology	During the entire working period
Urine, Stool and Sputum	Clinical Bacteriology	9.00 a.m. – 11.00 a.m.
Direct walk in clients	Virology and Immunology / ICTC	9.00 a.m to 4.00 p.m

TESTS / SERVICES OFFERED:

Division / Location	**Tests offered**	**Specimen type * and number where applicable**	**Contact Person with intercom number**
Clinical Bacteriology 7th floor, MSB	Microscopy& Culture for aerobic bacteria and anaerobic bacteria Antimicrobial susceptibility test on clinically relevant aerobic bacteria **MIC – Vitek2** Environmental sampling and sterility assurance tests as required	All specimens collected aseptically in sterile containers	Dr Gita Nataraj / Dr Priyanka Prasad / Dr Supriya Paranjpe/ Dr Shivani Shinde 7552 / 7527
Molecular Diagnostics 7th floor, MSB	**HIV** viral load	Whole blood	Dr Preeti Mehta / Dr Nayana Ingole 7272 / 7273
Samples to be sent to Mycobacteriology , 5th floor, MSB	**Xpert MTB/RIF ** assay** # for simultaneous detection of MTB and Rif resistance	Sputum specimen / GL / Extra pulmonary in Falcon tube (procured from dept)	Dr Gita Nataraj/ Dr Swapna Kanade/ Dr Vaishali Surase 7525
Mycology 5th floor, MSB	Microscopy , Culture, Identification for fungi	All specimens collected aseptically in sterile containers	Dr Shashir Wanjare 7824/ 7856
Mycobacteriology 5th floor, MSB	Microscopy , Culture and identification of isolates on request (non-programmatic)	Sputum** – at least 2 specimens of which one is early morning and the other is spot.	Dr Gita Nataraj / Dr Swapna Kanade / Dr Vaishali Surase 7827 / 7525
		Gastric lavage – 3 specimens collected on 3 different days Other specimens – One or more	

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Division / Location	**Tests offered**	**Specimen type * and number where applicable**	**Contact Person with intercom number**
Parasitology 5th floor, MSB	Stool – Routine and Microscopy Stool and other body fluids / tissues –for potential and opportunist parasites	- Stool -BAL -Other respiratory specimens -Hydatid fluid -Other body fluids	Dr Avani Koticha / Dr Alpana Mhashilkar 7832/ 7855
	RDT - malarial antigen	Whole blood (finger prick) / serum	
Serology 5th floor, MSB	ASO Dengue – NS1 antigen (Rapid / ELISA) Dengue – IgG and IgM antibodies (Rapid / ELISA) Leptospirosis – IgM Antibodies RA, Widal RPR / V.D.R.L Chikungunya IgM antibody **Referral of specimens to PCR laboratory at Kasturba Hospital for Leptospirosis , Dengue** **Referral of specimens to PCR laboratory at Kasturba Hospital for Leptospirosis , Dengue and pH1N12009**	Whole blood collected in clean, dry, plain test tube / red top evacuated tubes. PCR – 3-5 ml blood in purple cap evacuated tubes With filled requisition forms PCR pH1N1 – nasal swab / throat swab collected in VTM , triple packed and transported in cold chain	Dr Karmarkar / Dr Rupali Suryawanshi 7984 / 7985
Virology and Immunology 5th floor, MSB –	ICTC @ HIV – antibody detection HCV – antibody detection HBsAg detection RPR CD4 count enumeration	Whole blood collected in clean, dry, plain test tube / Plain vacutainer EDTA vacutainer	Dr Preeti Mehta / Dr Nayana Ingole / Dr Vijaya Torane / Dr Pallavi Surae 7825 / 7822

*Details about the specimen collection will be provided in the sections below.
**, # Specimens should be accompanied by appropriately filled RNTCP laboratory forms
@ Specimens should be accompanied by appropriately filled written informed consent form (Marathi / English) for HIV antibody test

- All sample containers should be adequately labelled.
- All samples should be accompanied by adequately filled requisition form.

TEST INDICATIONS AND LIMITATIONS

Sr.no.	**Specimen / test performed**	**Indications (major)**	**Limitations**
CLINICAL BACTERIOLOGY DIVISION			

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Sr.no.	Specimen / test performed	Indications (major)	Limitations
1	**Blood (conventional)** Aerobic culture & Antimicrobial susceptibility test	CRBSI, Enteric fever, Infection of prosthetic material (implants), Infective endocarditis (IE), Meningitis, Osteomyelitis Pneumonia, PUO, Septicemia	Usually positive only in acute phase. Multiple specimens required in IE. Lesser volumes (<10-20 ml) decrease yield. Blood culture contamination during collection can lead to pseudobacteremia.
2	**Blood (Automated method BACTEC** 9050) Rapid aerobic bacterial culture by automated system	Same as above If patient on antimicrobial, collect just before the next dose is due.	Pre-incubation of automated blood cultures reduces the yield of Pseudomonas, Streptococcus and Candida spp. In case of delay , store at room temperature (20-30°C)
3	**Normally sterile body fluids** – C.S.F, Pleural, Pericardial, Peritoneal (Ascitic), Joint, Smear, Culture and Antimicrobial susceptibility test	Infection at respective sites	Negative microscopy or culture does not rule out disease. Larger volumes improve sensitivity.
4	**Throat swab from suspected diphtheria** case Smear examination by microscopy for Diphtheria Culture on appropriate media	Suspected diphtheria	Microscopy – unreliable A positive culture followed by demonstration of exotoxin production is the gold standard
5	**Sputum -** Smear, Culture and Antimicrobial susceptibility test	Lower Respiratory tract infections, community / hospital acquired	Both sensitivity and specificity are considered </= 50% unless expectorated sputum is purulent.
6	**Respiratory samples** (mini BAL, BAL, endotracheal aspirate) Smear, Culture and Antimicrobial susceptibility test	Lower Respiratory tract infections, community / hospital acquired	Difficult to distinguish colonization from infection even with quantitative cultures. Clinical correlation essential.
7	**Miscellaneous** (Pharyngeal swabs, Skin scraping) Smear, Culture and Antimicrobial susceptibility test	Suspected streptococcal pharyngitis, Localised skin infections	Used to rule in disease. Collect samples in suspected GAS patients from posterior pharyngeal wall and tonsils. The isolate needs to be clinically correlated for its significance as a colonizer / pathogen. Swabs need to be transported to lab immediately. A dried swab is detrimental to growth and can give false negative results.
8	**Ocular specimens** (conjunctival swab, Corneal scrapings, corneal button, eye discharge, vitreous humor, cornea) Smear, Culture and Antimicrobial susceptibility test	Conjunctivitis, corneal transplant, corneal ulcer , other eye infections trachoma,	Negative microscopy or culture does not rule out disease. Bedside inoculation on appropriate media improves yield provided aseptic practices are followed.
9	**Pus** Smear, Culture and Antimicrobial susceptibility test	Localised skin or organ specific	Sensitivity – 70% Specificity - High

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10	**Wound swab** Smear examination by microscopy	Bacterial cellulitis, gas gangrene	Microscopy and culture unreliable. Collect tissue material or purulent discharge whenever possible.
11	**Tissue** (other appropriate specimen) for gas gangrene Smear and Culture (anaerobic)	Gas gangrene, local infection, intra-operative	Gas gangrene is a clinical diagnosis. Microscopy cannot characterize the genus. A negative test does not rule out disease. Swabs collected without appropriate debridement will yield contamination / false negative result.
12	**Specimens from female genital tract** (Vaginal /cervical swab, Urethral discharge, product of conception) and urethral discharge Smear, Culture and Antimicrobial susceptibility test	Vaginitis, cervicitis, urethritis	Specimens from lower genital tract will be contaminated with normal flora and difficult to interpret.
13	**Stool** **Microscopy**	Diarrhoeas, purulent enterocolitis	A negative test for darting motility does not rule out cholera (sensitivity and specificity ~ 60%)
14	**Stool** **Culture & Antimicrobial susceptibility test**	Diarrhoeas, dysentery, purulent enterocolitis	Necessary to process specimens immediately to prevent overgrowth by normal flora.
15	**Urine** **Smear, culture & Antimicrobial susceptibility test**	Recurrent / Complicated UTI Known UTI with treatment failure PUO Asymptomatic bacteriuria in pregnant women	-False positives with clean catch urine specimens is high since the urine sample passes through the distal urethra and can become contaminated with commensal bacteria. -Culture of urine from urine collection bag gives false positive result. -Culture positive urine in a sick patient does not exclude another site of serious infection. -Prior antibiotic therapy may lead to negative urine culture in patients with UTI. -Sterile pyuriamaybe due to causes other than non-fastidious aerobic bacteria.
SEROLOGY DIVISION			
16	**RA Test for rheumatoid factors**	In-vitro detection of Rheumatoid factor in patients serum by latex agglutination method.	-Does not provide definite diagnosis of rheumatoid arthritis and should always be correlated clinically -False positive results are seen in auto immune diseases, acute bacterial and viral diseases - Test can be negative in spme patients with RA.
17	**ASO test**	Detection of antibodies to streptolysin O produced by group A beta hemolytic streptococci by latex agglutination method.	-All positive results should always be correlated clinically -Nonspecific results are seen in lipemic, hemolysed, contaminated and high protein content serum

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Sr.no.	Specimen / test performed	Indications (major)	Limitations
			-False positive results are seen with the use of plasma instead of serum
18	**RPR / VDRL Test**	For detection and quantification of regain antibody in serum/plasma and spinal fluid in syphilitic patients.	-Nonspecific test for syphilis - All positive results should be correlated clinically -All positive samples should be confirmed by TPHA or FTA ABS - False Negative: early primary syphilis; in secondary syphilis because of prozone reaction; and in some cases of late syphilis. -Biological false positive occurs in conditions such as -infectious mononucleosis, viral pneumonia, malaria, lepromatous leprosy, pregnancy, collagen disease, other autoimmune diseases
19	**Widal Test**	Detection of typhoid fever or paratyphoid fever by agglutination method.	-Not a specific (65%) or sensitive test (65%) -All reactive titres should be correlated clinically - TAB vaccinated patients may show high titres
20	**LeptoIgM rapid**	Qualitative detection of IgM class of Leptospira specific antibodies in human serum/ plasma/whole blood by rapid immunochromatography method.	- Less specific than ELISA -All positive results should always be correlated clinically -Intensity of test line depends on the stage of the disease and titre of the antibody -Samples collected during early stage of disease (0-7days) may yield negative results
21	**LeptoIgM ELISA**	Qualitative detection of IgM class of antibodies against Leptospira by ELISA method.	Same as above
22	**Dengue NS1 - Rapid**	Qualitative detection of non-structural protein 1 (NS1) of dengue virus in serum/plasma by rapid immunochromatography method	Samples collected during late stage of disease (after 7 - 9 days of fever) may yield negative results
23	**Dengue NS1 - ELISA**	Same as above	Same as above
24	**Dengue IgG/IgM Rapid**	Qualitative detection of IgG or IgM class of antibodies against dengue virus in human serum/ plasma by rapid immunochromatography method	- Not as specific or sensitive as ELISA -All positive results should always be correlated clinically -Intensity of test line depends on the stage of the disease and titre of the antibody -Samples collected during early stage of disease (0-7days) may yield negative results
25	**Dengue IgM ELISA**	Same as above	Same as above
26	**Chikungunya Antibody - ELISA**	Qualitative detection of IgM class of antibodies against Chikungunya virus by ELISA method.	All positive results should be correlated clinically

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MYCOBACTERIOLOGY DIVISION			
27	**Microscopy**	Clinical suspicion of PTB / EPTB	Sensitivity low (10^5 orgs/ml)
28	**Culture**	All EPTB cases and suspected MDRTB cases	Solid culture – 4 weeks for report
29	**XpertMTB/RIF assay**	MDRTB suspects, pediatric TB, all HIV positive TB suspects and extrapulmonary TB	Detects rifampicin resistance only. Cannot predict for other anti-TB drugs other than INH.
PARASITOLOGY DIVISION			
30	**Stool / other specimens - Microscopy**	Suspected parasitic infection in immunocompetent / immunocompromised patients	For detecting trophozoites, fresh stool specimen essential to be examined within the hour of collection. A negative result on a single stool specimen does not rule out parasitic presence.
31	**Blood – RDT malarial antigen**	Clinically suspected malaria cases	- Detection limit is usually 200 parasites / µl. May not detect low level parasitemia. -Use of RDT does not eliminate the need for malaria microscopy. -The currently approved RDT detects 2 different malaria antigens; one is specific for P. falciparum and the other is found in all 4 human species of malaria. Thus, microscopy is needed to determine the species of malaria other than P.falciparum.
MYCOLOGY DIVISION			
32	**Any specimen – Microscopy(KOH)**	Suspected superficial or deep fungal infection	-The sensitivity of a KOH prep is relatively low (20-75%) -The test may require overnight incubation for complete disintegration of thicker specimens like hair, nail, or skin
33	**Microscopy – India ink**	Suspected cryptococcal infection	-The diagnosis of *C. neoformans* by India ink staining should be considered a presumptive result -Culture, biochemical and serological testing is recommended for final identification. Some strains of *C. neoformans*, as well as other cryptococci may not produce discernible capsule
33	**Culture**	Suspected superficial or deep fungal infection	-Longer time required for growth of different fungi -Contamination by saprophytic fungi

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VIROLOGY AND IMMUNOLOGY DIVISION			
35	**HIV Antibody tests (Rapid)**	-Patients who present with symptoms suggestive of HIV infection. Examples pneumonia, TB or persistent diarrhoea. -Patients with conditions that could be associated with HIV such as STI/RTI. -Prevention of parent (mother) to child transmission - pregnant women who register at ANCs. These also include pregnant women who directly come in labour without any antenatal check-up	-False Negative result : in window period & terminal stage of HIV disease -False positive result: autoimmune disease, multiple blood transfusion, pregnancy etc.
36	**HBsAg ELISA**	• Signs/symptoms suggestive of hepatitis • H/o exposure	-False Negative : during incubation period -False positive: due to presence of other antigens or elevated levels of Rheumatoid factor
37	**Anti HCV ELISA**	• Signs/symptom suggestive of hepatitis • H/o exposure	-False Negative: in window period -False positive: elevated levels of Rheumatoid factor - Cannot differentiate recent from past infection
38	**RPR test**	• Direct walk in patients with high risk behavior • Patients referred by the STI counselor	-See page 20 above
39	**CD4 count**	• HIV positive patients referred from the ART centre	-Nonspecific marker which can be affected by many other conditions
MOLECULAR DIAGNOSTICS			
40	**HIV viral load**	HIV seropositive patients to be initiated / on ART	The detection limit (sensitivity) varies between kits . The current test has a detection limit of 47 rna copies / ml/
REFERRAL OF SPECIMENS			
41	**Lepto PCR**	Suspected leptospirosis, 1st week, antibody negative	A negative test does not rule out disease. A positive test to be correlated clinically and with other microbiological tests. Best results when specimens tested the same day of collection.

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			Follow triple packaging while transporting. Transport in cold chain.
42	**Dengue PCR**	Suspected Dengue, 1st week, NS1 Ag and IgM Ab negative	Same as above. Does not speciate.
43	**Nasal / Throat swabs / other respiratory secretions for pH1N1**	Collect prior to administration of antivirals. Time - 1st week of Influenza Like Illness (ILI) in admitted patients . URT specimens are positive usually only for the first 4 days. LRT specimens are positive for longer periods.	In case of a negative test, collect additional specimens if there is a strong suspicion. False negative results can occur due to improper or poor clinical specimen collection or from poor handling of a specimen after collection and before testing.

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