ADVANCED DIAGNOSTIC LABORATORY
ADVANCED MEDICAL AND DENTAL INSTITUTE
UNIVERSITI SAINS MALAYSIA

ADL USER MANUAL
(AMDI/ADL/UM)

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ADVANCED DIAGNOSTIC LABORATORY (ADL)

CHAPTER 1

ADVANCED DIAGNOSTIC LABORATORY (ADL)

1.0 INTRODUCTION

Advanced Diagnostic Laboratory (ADL) is one of the services located at Clinical Trial Complex, Advanced Medical and Dental Institute (AMDI), Bertam, Kepala Batas, Pulau Pinang.

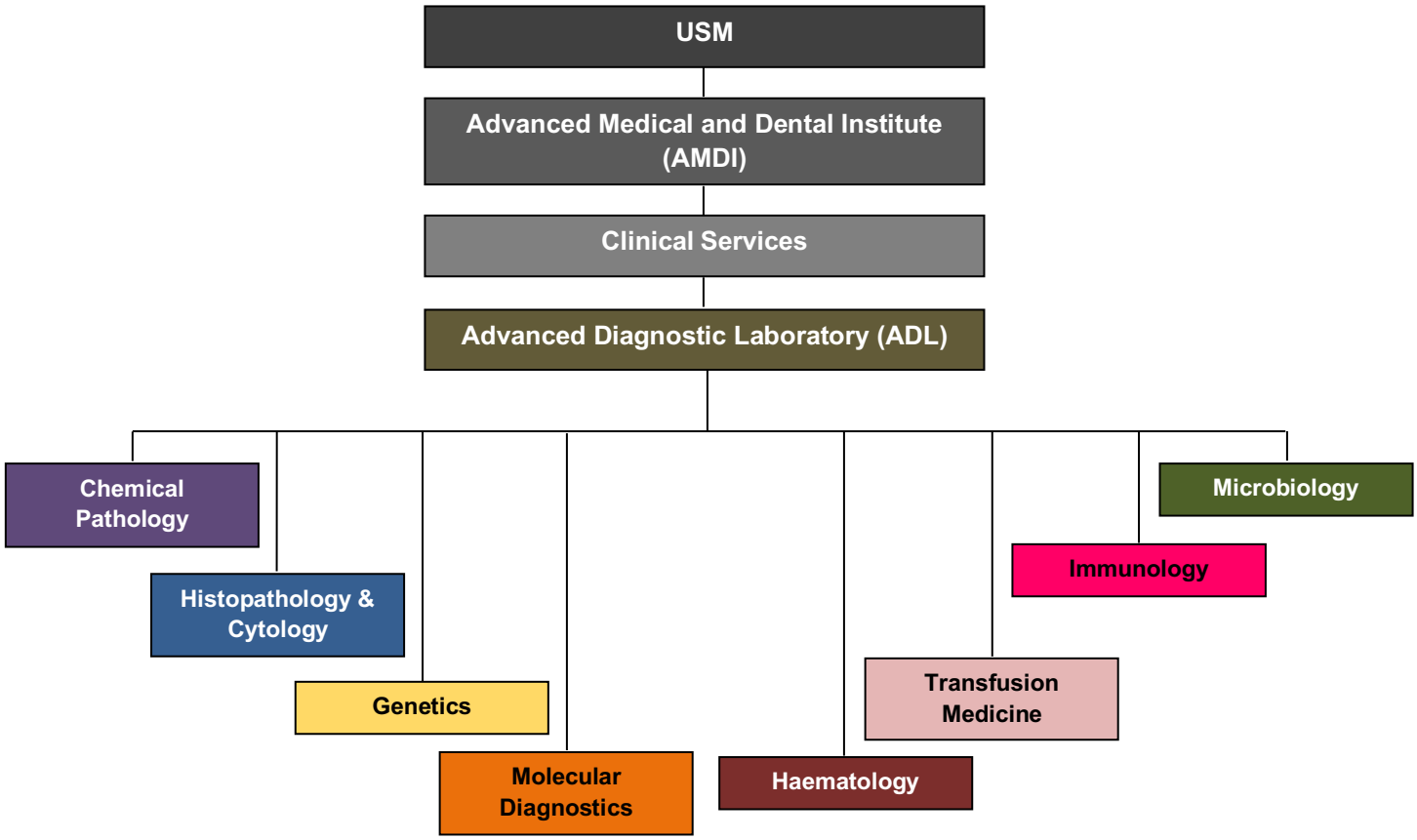
Services are offered to government and private hospitals / clinics / laboratories. This manual emphasizes the importance of proper pre-analytical procedures regarding the collection of specimens, correct usage of specimen containers and proper transportation to ensure efficient, effective, precise and accurate tests and delivery of results within the acceptable time interval.

2.0 OBJECTIVES

ADL provides medical laboratory services in Haematology, Chemical Pathology, Genetics, Histopathology and Cytology, Microbiology, Transfusion Medicine, Molecular Diagnostics and Immunology. Our customers include AMDI wards / clinics, government hospitals / clinics / laboratories and private hospitals / clinics / laboratories. To achieve quality services in line with international practice, ADL is committed to:

- providing professional services, ensuring accurate and precise results in accordance with international standard methodologies,
- implementing a quality management system, which is compiled to MS ISO 15189 so as to ensure management plans and technical operations are established and controlled,
- ensuring all personnel who are involved in examination activities are familiar with the Laboratory Quality Management System (LQMS), and implement the possible process at all the time, and
- ensuring continual improvement of its LQMS.

3.0 ORGANIZATION CHART AND VARIOUS UNITS



The laboratory services include:

a) Chemical Pathology
b) Histopathology and Cytology*
c) Genetics
d) Molecular Diagnostics*
e) Haematology
f) Transfusion Medicine
g) Immunology*
h) Microbiology

***Note: Cytology, Molecular Diagnostic and Immunology services are not yet MS ISO 15189 accredited.**

4.0 WHO TO CONTACTS

Advanced Diagnostic Laboratory (ADL),
Clinical Complex,
Advanced Medical and Dental Institute (IPPT),
Universiti Sains Malaysia,
13200 Kepala Batas,
Pulau Pinang.

AMDI Operator: 04-562 2888
Direct line: 04-562 XXXX*

*Extension:

Receiving counter	- 2711
Haematology	- 2693
Chemical Pathology	- 2686
Genetics	- 2692
Histopathology and Cytology	- 2685
Microbiology	- 2696
Transfusion Medicine	- 2700
Molecular Diagnostics**	- 2293
Immunology**	- 2331

****Note: Molecular Diagnostics Unit and Immunology Unit are located at Animal Research Complex (ARC), IPPT.**

5.0 BUSINESS HOURS

Schedule for receiving specimen:

All specimens must reach the ADL receiving counter.

DAY	SHIFT	
	MORNING	AFTERNOON
*Monday - Thursday	8.10am - 1.00 pm	2.00 pm - 5.10 pm
*Friday	8.10am - 12.15 pm	2.45 pm - 5.10 pm

After office hours / public holiday all the request shall be sent directly to the specific unit (Chemical Pathology, Haematology, Transfusion Medicine and Microbiology)

***Note:**

1. **For specimen involving Genetics Unit & Immunology Unit, clients should make appointments and samples should reach ADL receiving counter before 3.00 pm except for urgent samples.**
2. **For specimen involves Immunology Unit, clients should make a contact to the Immunology Laboratory to make appointment. Preferably patient's sample should be arrived before 10.00 am unless agreed otherwise.**

6.0 SAMPLE SUBMISSION REQUIREMENTS

ADL only receive samples from requesting parties.
All samples should be sent directly to the ADL receiving counter.
All samples should be treated as biohazards and to be dispatched to ADL as soon as possible.
Please refer specific requirements for samples requiring special transportation procedures.

The request form should be completely and correctly filled manually or equivalent. Samples and request forms should be sent to ADL receiving counter. Details of patient must include the following information:

- Name
- Identification Card No.
- Age / Date of birth
- Gender
- Location / contact details of the patient
- Clinical summary
- Date and time of sample collection
- Type of sample and test requested
- Name and signature of the doctor requesting the test and stamp

7.0 URGENT REQUESTS

Clients need to inform ADL via phone before sending urgent samples (except for Immunology Unit and Histopathology and Cytology Unit). Urgent samples must reach ADL counter as soon as possible and should be labelled '**URGENT**' or equivalent, with date and time of request clearly stated.

ADL will inform the results via phone upon request followed by printed report / result (except for Immunology Unit and Histopathology and Cytology Unit).

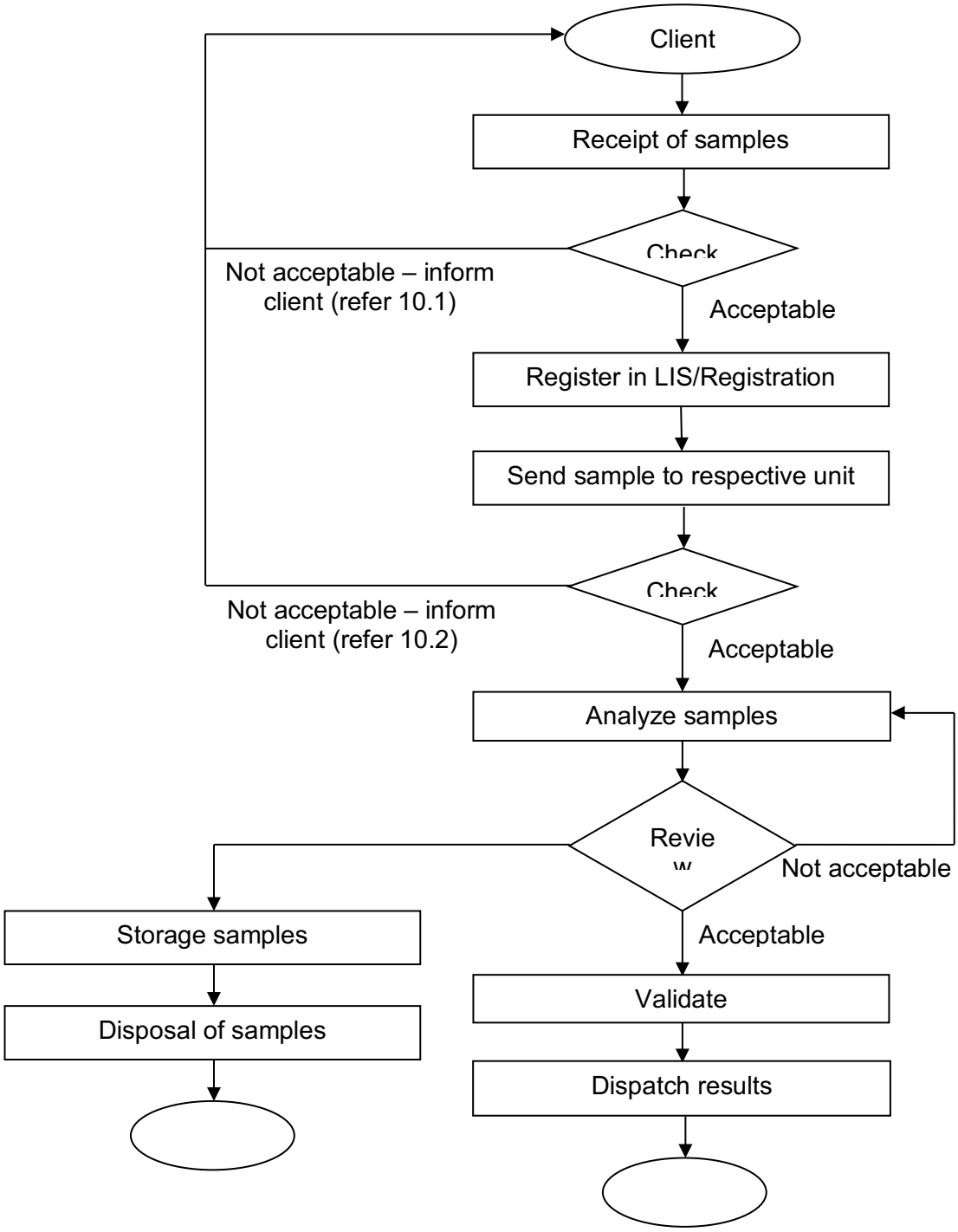
Requests which fail to adhere to the above instructions will not be entertained as '**URGENT**' or equivalent.

For list of urgent request, please refer to each specific unit.

8.0 PANIC VALUE

Panic value is defined as any result outside the normal ranges to a degree that may pose immediate health risk to the individuals or require immediate action on the part of the treating doctors. It is the responsibility of the laboratory to immediately NOTIFY the doctor of the panic values.

9.0 ADL WORKFLOW



10.0 REJECTION CRITERIA

10.1 First level rejection criteria - samples shall not be accepted or processed if any of the following criteria exist:

a) No name on the request form or electronic form (LIS) / specimen.
b) Information on request form or electronic form (LIS) is not tally with specimen.
c) Incomplete identification card / registration number.
d) No name, signature and stamp of authorized requester / clinician.
e) Test is not indicated.
f) Unsatisfactory specimen, for example spillages, breakages and etc.

10.2 Second level rejection criteria – sample not suitable to process if the following criteria exist:

a) Inappropriate sample container / request form.
b) Compromised sample such as haemolysed, clotted, insufficient, aged or other causes of unsuitable sample for analysis.
c) Test is not available.
d) Inappropriate sample transportation.
e) No clinical history information (whenever applicable).

11.0 VERBAL REQUESTS

ADL accepts verbal requests for additional tests by calling or informing the respective units by the requesting medical practitioner followed by request form or electronic equivalent within a given time. Acceptance criteria for verbal requests are stated in the user manual for each unit.

12.0 UNDERSTANDING OF EXAMINATION PROCESSES (TEST METHODOLOGY)

The users are expected to understand the general principles of examination processes for the test results generated in ADL. A detail description of examination processes will be provided upon request.

13.0 SPECIMEN COLLECTION & LABELLING

13.1 Specimen is to be collected in the ward / clinics / others. Please refer to specific unit / test for proper specimen collection procedure (where indicated).

13.2 Clear instruction is to be given to the patient if the specimen needs to be collected by the patient.

13.3 All specimens from the patients are to be put in biohazard bag and attached with the request forms.

13.4 All specimens shall be treated as biohazard and to be dispatched to the laboratory as soon as possible. (Please refer to the specific test that requires special transportation procedures in each unit).

13.5 For multiple specimen collection, please follow “order of draw” rules.

13.6 Each specimen shall have a label firmly attached to the specimen container and bearing the following information:

- Patient's name
- Patient's registration number (PID)
- Name of Ward or Clinic
- Type of specimen (including specific anatomic site)
- Date and time of specimen collection

14.0 CONSENT

Patient consent is required where referral is needed (e.g. consent to disclose clinical information and family history to relevant healthcare professionals).

15.0 KNOWN FACTORS THAT AFFECT THE PERFORMANCE OF THE EXAMINATION / INTERPRETATION OF THE RESULT

a. Clotted sample
b. Lysed sample
c. Lipemic sample
d. Aged sample
e. Icteric sample
f. Contaminated sample
g. Insufficient sample
h. Wrong container
i. Inappropriate sample transportation

16.0 CONFIDENTIALITY

All tests done by ADL, AMDI are confidential between ADL and customers. Personnel are not allowed to disclose any information that can destitute the safety and confidential of test work.

17.0 CUSTOMER COMPLAINTS AND FEEDBACK

Any complaints, compliments or feedback can be directly addressed to ADL through verbal or written. Verbal complaint can be done through phone or directly to the staff while written complaint via complaint form, formal letter or email (adl@usm.my). Customer Complaint Form (*ADL/QP6/F-1*) can be downloaded from ADL website.

18.0 FURTHER INQUIRIES

Further Inquiries regarding this user manual could be directed to:

Quality Manager
Phone: 04-562 2656
Email: adl@usm.my

CHAPTER 2
CHEMICAL PATHOLOGY UNIT

CHAPTER 2

CHEMICAL PATHOLOGY UNIT

1.0 INTRODUCTION

Chemical Pathology Unit offers diagnostic investigations in chemical pathology.

2.0 LIST OF AVAILABLE TESTS AND SAMPLE COLLECTION

All samples to be sent to Chemical Pathology Unit should use specific containers according to tests requested. Required volume and container type for each test provided is listed in the table below:

PROFILE	TEST	CONTAINER	VOLUME	SPECIMEN	REMARKS
*Blood Gas	Arterial / Venous Blood gas	Heparinized syringe	1 - 2 mL	Blood	Refer to 8.0 Special Procedures
Renal Function Test (RFT)	Sodium	Plain tube / Lithium Heparin	3 - 5 mL	Blood	-
	Potassium				
	Chloride				
	Urea				
	Creatinine				
	Uric Acid				
	Calcium				
	Inorganic Phosphate				
Liver Function Test (LFT)	Total Protein	Plain tube / Lithium Heparin	3 - 5 mL	Blood	-
	Albumin				
	Globulin				
	Albumin / Globulin ratio				
	Alanine Aminotransferase (ALT)				
	Aspartate Aminotransferase (AST)				
	Alkaline Phosphatase (ALP)				
	Total Bilirubin				
	Direct Bilirubin				
Lipid Profile (LP)	Total Cholesterol	Plain tube / EDTA tube	3 - 5 mL	Blood	8 - 10 hours fasting
	Triglyceride				
	High Density Lipoprotein (HDL)				
	Low Density Lipoprotein (LDL)				

PROFILE	TEST	CONTAINER	VOLUME	SPECIMEN	REMARKS
Cardiac Profile	Creatinine Kinase (CK)	Plain tube / Lithium Heparin	3 - 5 mL	Blood	-
	Lactate Dehydrogenase (LDH)				
	Aspartate Aminotransferase (AST)				
Diabetic Profile	Glucose	Sodium Fluoride tube	2 mL	Blood	Refer to 8.0 Special Procedures (MGTT)
	*HbA1c	EDTA tube	2.5 ml	Blood	-
-	Magnesium	Plain tube / Lithium Heparin	3 mL	Blood	-
-	*Osmolality	Plain tube	3 ml	Blood	-
		Universal bottle	5 ml (spot)	Urine	
-	Amylase	Plain tube	3 ml	Blood	-
-	*C-Reactive Protein (CRP)	Plain tube	3 ml	Blood	-
Tumor Markers	CA 125	Plain tube	3 - 5 mL	Blood	-
	*CA 15-3				
	Carcino Embryonic Antigen (CEA)				
	Total Prostate Specific Antigen (TPSA)				
	*Alpha Feto Protein (AFP)				
	*Beta Human Chorionic Gonadotropin (BHCG)				
Thyroid Function Test	Thyroid Stimulating Hormone (TSH)	Plain tube	3 - 5 mL	Blood	-
	Free Thyroxine (FT_4)				
	Free Triiodothyronine (FT_3)				
Anaemia Test	Vitamin B12	Plain tube	3 - 5 mL	Blood	-
	Ferritin				
	Folate				
*Urine Analysis	Urine microscopy	Urine container	10 ml	Urine	Magnification x 40
	pH				-
	Specific Gravity				
	Haemoglobin				
	Nitrite				
	Glucose				
	Protein				
	Bilirubin				

PROFILE	TEST	CONTAINER	VOLUME	SPECIMEN	REMARKS
*Urine Analysis	Urobilinogen	Urine container	10 ml	Urine	-
	Ketone				
*Urine Drug	Cannabis	Urine container	10 ml	Urine	-
	Morphine				
	Amphetamine				
*Urine Pregnancy Test (UPT)	Pregnancy Test	Urine container	10 ml	Urine	-

Note: All specimen collection with EDTA, Sodium Fluoride and Lithium Heparin tube should be mixed gently and thoroughly for 1 minute by a rotary wrist movement.

***Note: Marked tests are not MS ISO 15189 accredited.**

3.0 URGENT TEST

- Blood Urea Serum Electrolyte (BUSE)
- Calcium
- Creatinine
- Total Bilirubin (Paediatric case)
- Beta hCG
- Amylase

4.0 OUTSOURCE TESTS

Transport of intended outsource samples to referral lab (HUSM, Kubang Kerian) will be done once per week upon availability of sample / request. Test availability, method used, procedure and test schedule, please refer to the referral laboratory.

5.0 SAMPLE COLLECTION AND TRANSPORTATION

Procedure for Packaging and Transportation of Samples from External Laboratories, Hospitals and Institutions to Chemical Pathology Unit, Advanced Diagnostic Laboratory (ADL), IPPT, USM.

1) Request forms from either the requesting party or Chemical Pathology Unit, ADL, IPPT, USM are acceptable.
2) The request form shall be duly filled with the following information:
    - Name
    - Identification Card No. / RN
    - Age / Date of Birth
    - Gender
    - Location
    - Clinical summary
    - Date and time of sample collection
    - Type of sample and test requested
    - Name and signature of the doctor requesting the test and stamp

3) Sample in gel / plain tube shall be centrifuged and / or separated into labelled secondary tube prior to send. For whole blood and urine, it has to be sent in primary tube.
4) All samples shall be transported in ice-box with reusable ice-packs. Ideally the ice-packs has been freeze for minimum of 8 hours prior to use.
5) Samples shall reach Chemical Pathology Laboratory, ADL Unit, IPPT, USM during office hours **ONLY (refer to User Manual AMDI/ADL/UM 5.0 Business hours)**.

6.0 ACCEPTANCE CRITERIA FOR URGENT REQUESTS

6.1 Request shall be made via phone by authorized personnel.
6.2 Samples must be sent immediately to the laboratory.

7.0 ACCEPTANCE CRITERIA FOR REQUEST OF REPEAT / ADDITIONAL TEST

7.1 Clients need to call lab for request repeat / additional test.
7.2 Request must be made on the same day sample is sent to the laboratory and within stability period.
7.3 Request can only be accepted if sample is sufficient and within the stability period for the requested analyte.
7.4 Requested test is suitable to sample collected.

8.0 SPECIAL PROCEDURES

TEST	COLLECTION PROCEDURE	REMARKS
Blood Gas	1. Use disposable heparinised syringe. 2. Draw 1 mL of blood. Invert the syringe and remove air bubbles inside the syringe. 3. Use a lock stopper instead of needle to avoid exposure to air and to avoid blood sample leakage. Mix well by rotating the syringe to prevent clotting. 4. Put the sample in ice bath	**The specimen must be kept embedded in crushed ice and sent immediately to the laboratory for analysis (at least within ½ hour).**
Modified Glucose Tolerance Test (MGTT)	1. Patients must fast for at least 8 hours. 2. Collect blood sample. 3. Give patient 75 g glucose in 300 ml water and drink within 5 minutes. 4. For children, the glucose dose is 1.75 g/kg body weight to a maximum of 75 g. (Exception: Pregnant women receive 50 g glucose to screen for gestational diabetes) 5. Collect blood sample after 2 hours.	**Each blood specimen collected must be labelled correctly.**

9.0 REPORTING RESULTS

Turn Around Time (TAT)

CASE		TAT
Biochemistry	*Urgent	Within 1 hour from the time sample is received
	*Non-urgent	Within 4 hours working day from the time sample is received
Immunoassay	Urgent	Within 3 hours from the time sample is received
	*Non-urgent	Within 6 working days from the time sample is received

***Note: 1. The TAT for urgent & non-urgent biochemistry tests is applicable only during office hours (from 9.00 am- 5.00 pm)**

2. Non-urgent immunoassay tests will be performed once a week.

10.0 PANIC VALUES

a) Values for Adults

ANALYTES		RESULTS		
		UNITS	LOWER CRITICAL LIMIT	UPPER CRITICAL LIMIT
Sodium		mmol/L	125	155
Potassium		mmol/L	2.8	6.0 (no haemolyse sample)
Magnesium		mmol/L	0.41	2.00
Glucose		mmol/L	2.8	20.0
Calcium		mmol/L	1.50	3.00
Inorganic Phosphate		mmol/L	0.32	2.87
Amylase		U/L	-	1000
Arterial Blood Gas	pH	-	7.20	7.55
	PCO_2	mmHg	-	69.8
	PO_2	mmHg	58.5	-

b) Values for Pediatric

ANALYTES		RESULTS		
		UNITS	LOWER CRITICAL LIMIT	UPPER CRITICAL LIMIT
Sodium		mmol/L	125	155
Potassium		mmol/L	2.8	6.0 (no haemolyse sample)
Magnesium		mmol/L	0.50	1.80
Total Bilirubin		µmol/L	-	Neonates (< 28 days) 513 µmol/L Children 428 µmol/L
Calcium		mmol/L	1.70	3.10
Inorganic Phosphate		mmol/L	0.40	2.80
Urea		mmol/L	-	19.0
Arterial Blood Gas	pH	-	-	7.60
	PCO_2	mmHg	19.5	68.2
	PO_2	mmHg	43.88	121.51

Reference:

The cut off values above are adapted from: Quick Guide for Improving Notification of Critical Laboratory Results in MOH Hospitals*. A project for Improving Patient Safety.*

11.0 REFERENCE RANGES

PROFILE	TEST	REFERENCE RANGES			SI UNIT
		CRITERIA		RANGES	
*Blood Gas	Arterial Blood gas (ABG)	pH		7.35 - 7.45	-
		PCO2		35 - 45	mmHg
		PO2		75 - 100	
		HCO3		24 - 32	mmol/L
		TCO_2		24 - 30	
Renal Function Test (RFT)	Sodium	-		136 - 146	mmol/L
	Potassium	-		3.5 - 5.1	
	Chloride	-		101 - 109	
	Urea	-		2.8 - 7.2	
	Creatinine	Male		59 - 104	µmol/L
		Female		45 - 84	
	Uric Acid	Male		208 - 428	
		Female		154 - 357	
	Calcium	Adult		2.20 - 2.65	mmol/L
		Child	0 - 10 days	1.90 - 2.60	
			2 - 12 years	2.20 - 2.70	
	Inorganic Phosphate	Adult		0.81 - 1.45	
		Child		1.29 - 2.26	
Liver Function Test (LFT)	Total Protein	Adult		66 - 83	g/L
		Child (1 - 18 years)		57 - 80	
	Albumin	Adult		35 - 52	
		New-born		28 - 44	
	Globulin	-		23 - 35	g/L
	A/G Ratio	-		1.10 - 2.10	-
	Alanine Transaminase (ALT)	Male		<50	U/L
		Female		<35	
	Aspartate Aminotransferase (AST)	Male		<50	
		Female		<35	

PROFILE	TEST	REFERENCE RANGES				SI UNIT
		CRITERIA		RANGES		
Liver Function Test (LFT)	Alkaline Phosphate (ALP)	Adult (> 17 years)		30 - 120		U/L
		Child	Age	Male	Female	
			1 - 30 d	75 - 316	48 - 406	
			30 d - 1 y	82 - 383	124 - 341	
			1 - 3 y	104 - 345	108 - 317	
			4 - 6 y	93 - 309	96 - 297	
			7 - 9 y	86 - 315	69 - 325	
			10 - 12 y	42 - 362	51 - 332	
			13 - 15 y	74 - 390	50 - 162	
			16 - 18 y	52 - 171	47 - 119	
	Total Bilirubin	Adult		5 - 21		µmol/L
		Child	0 - d	24 - 149		
			1 - 2 d	58 - 197		
			3 - 5 d	26 - 205		
	Direct Bilirubin	Adult and child		< 3.4		
	Indirect Bilirubin	-		< 17.6		
Lipid Profile (LP)	Total Cholesterol	Desirable		< 5.2		mmol/L
		Borderline High		5.2 - 6.2		
		High		≥ 6.2		
	Triglyceride	Normal		< 1.70		
		Borderline High		1.70 - 2.25		
		High		2.26 - 5.64		
		Very high		≥ 5.65		
	High Density Lipoprotein (HDL)	Major risk factor for coronary heart disease		< 1.03		
		"Negative" risk factor for coronary heart disease		≥ 1.55		
	Low Density Lipoprotein (LDL)	Optimal		< 2.59		
		Near optimal		2.59 - 3.34		
		Borderline High		3.36 - 4.11		
		High		4.14 - 4.89		
		Very high		≥ 4.91		
Cardiac Profile (CE)	Creatinine Kinase (CK)	Male		≤ 171		U/L
		Female		≤ 145		
	Lactate Dehydrogenase (LDH)	Male		< 248		
		Female		< 247		
		Child	0 - 4 d	290 - 775		
			4 - 10 d	545 - 2000		
			10 d - 24 m	180 - 430		
			24 m - 12 y	110 - 295		
	Aspartate Aminotransferase (AST)	Male		< 35		
		Female		< 31		

PROFILE	TEST	REFERENCE RANGES			SI UNIT
		CRITERIA		RANGES	
Diabetic Profile	Glucose	Fasting	Adult	4.1 - 5.9	mmol/L
			Child	3.3 - 5.6	
		Random		≤ 7.8	
	MGTT	Fasting	Normal	< 6.1	mmol/L
			DM	> 7.0	
			IGT	6.1 - 7.0	
		2 hours	Normal	< 7.8	
			DM	> 11.1	
			IGT	7.8 - 11.1	
	*HbA1c	Normal		<5.6% (38)	mmol/mol
		Pre-diabetes		5.6 - 6.2% (38 - 44)	
		Diabetes		≥6.3% (45)	
Others	Amylase	-		34 - 117	U/L
	Magnesium	Male		0.73 - 1.06	mmol/L
		Female		0.77 - 1.03	
	*C-Reactive Protein (CRP)	Adult		< 5	mg/L
	*Osmolality	Serum		276 - 297	mOsm/kg H_2O
		Urine		300 – 900	
Tumor Markers	CA 125	-		0.0 - 35.0	U/mL
	*CA 15-3	-		≤ 31.3	
	Carcino Embryonic Antigen (CEA)	-		0.00 - 5.00	ng/mL
	Total Prostate Specific Antigen (TPSA)	-		0 - 4.00	ng/mL
	*Alpha Feto Protein (AFP)	-		0.89 - 8.78	ng/mL
	*Beta Human Chorionic Gonadotropin (BHCG)	Female	Non-pregnant	≤ 5.00	mIU/mL
			May be indicative of early pregnancy	5.00 - 25.00	
			Pregnant	≥ 25.00	
		Male		≤ 5.00	
Thyroid Function Test	Thyroid Stimulating Hormone (TSH)	-		0.35 - 4.94	uIU/mL
	Free Thyroxine (FT_4)	-		9.009 - 19.048	pmol/L
	Free Triiodothyronine (FT_3)	-		2.89 - 4.88	
Anaemia Test	Vitamin B12	-		187 - 883	pg/mL
	Ferritin	Male		21.81 - 274.66	ng/mL
		Female		4.63 - 204.00	
	Folate	-		3.1 - 20.5	ng/mL

PROFILE	TEST	REFERENCE RANGES			SI UNIT
		CRITERIA		RANGES	
*Urine FEME	*Urine microscopy	Erythrocytes		0 - 1	Per high power field (/hpf)
		Leucocytes		1 - 4	
		Epithelial cells	Squamous	5 - 15	
			Renal	Not Detectable	
		Casts	Hyaline	Occasional	
			Epithelial, Erythrocyte, granulated, leucocyte	Not Detectable	
*Urine Analysis	pH	Normal		4.6 - 8.0	-
	Specific Gravity	Normal		1.001 - 1.035	-
	Erythrocytes	Normal		< 3	Ery/µL
	Leukocytes	Normal		NEG	Leu/µL
	Nitrite	Normal		NEG	-
	Glucose	Normal		< 30	mg/dL
	Protein	Normal		< 15	
	Bilirubin	Normal		< 0.02	
	Urobilinogen	Normal		< 1.0	EU/dL
	Ketone	Normal		NEG	-
*Urine Drug	Cannabis	Normal		NEG	-
	Morphine	Normal		NEG	-
	Amphetamine	Normal		NEG	-
*Urine Pregnancy Test (UPT)	Pregnancy Test	Not pregnant		NEG	-
		Pregnant		POS	-

***Note: Tests that are not MS ISO 15189 accredited.**

References:

a) Biochemistry: Beckman Coulter Reagent Guide Manual
b) HbA1c: HbA1c test pamphlet
c) Immunoassay: Abbott Architect Reagent Pamphlet
d) Urine:
   i. Siemens Clinitek Advantus test pamphlet
   ii. Roche Diagnostics Reference Ranges for Adults and Children (*Pre-Analytical Consideration*) 2002/03
   iii. Men B-Hcg: http://en.wikipedia.org/wiki/Human_chorionic_gonadotropin
e) Malaysian CPG – Management of Type II DM
f) Mediven ProDetect Drug of Abuse Test (MOP) test pamphlet.
g) Mediven ProDetect Drug of Abuse Test (THC) test pamphlet.
h) Mediven ProDetect Drug of Abuse Test (AMP) test pamphlet.
i) Mayo Clinic Laboratories, Endocrinology Catalog Additional Information: mml-diabetes-metabolic-nutrition.
j) Total Protein and A/G Ratio: University of Rochester Medical Center.

CHAPTER 3
HISTOPATHOLOGY AND CYTOLOGY UNIT

CHAPTER 3

HISTOPATHOLOGY AND CYTOLOGY UNIT

1.0 INTRODUCTION

Histopathology and Cytology Unit is divided into two: Histopathology and Cytology.

Histopathology subunit provides services for routine diagnostic histopathology services including special staining tests for histochemistry and immunohistochemistry. This unit also provides appointment-based Histopathology services for Frozen Section samples.

Cytology subunit provides services for gynaecologic and non-gynaecologic specimens. This unit also provides appointment-based cytology services for Fine Needle Aspiration Cytology (FNAC) samples.

2.0 LIST OF AVAILABLE TESTS

2.1 **Routine Test:**

SUBUNIT	ROUTINE TEST
Histopathology	1) Routine Diagnostic 2) Histochemistry 3) Immunohistochemistry
Cytology	1) Non- Gynaecologic Cytologic a) Bile duct brushings b) Bronchiole brushings and washings c) Cerebrospinal fluid d) Esophageal brushing e) Gastric brushing f) Pericardial fluid g) Peritoneal fluid h) Pleural fluid i) Sputum cytology j) Urinary tract specimen 2) Gynaecologic Cytology a) Cervical smear

2.2 **Appointment:**

SUBUNIT	APPOINTMENT
Histopathology	1) Frozen Section ▪ Appointments must be made by Surgeon / Medical Officer by informing the Pathologist in-charge at least one day before the surgery. Only the Pathologist in-charge is able to accept the requests. ▪ Appointment date and time must be informed to the Pathologist in-charge by the surgeon / MO. ▪ Frozen section is only elligible during working hours with inclusion of afternoon breaktime.
Cytology	1) Fine Needle Aspiration Cytology (FNAC) ▪ Appointment should be made at least 24 hours before the procedure. ▪ FNAC appointment can be made anytime during office hour just for case by case (urgent or special case only).

***Note: The cytology tests are not MS ISO 15189 accredited.**

3.0 SAMPLE COLLECTION AND REQUIREMENT

3.1 **Histopathology**

TYPES OF SAMPLE	COLLECTION AND REQUIREMENT	FIXATIVE
Tissues	▪ All tissues should be immersed immediately in fixative in a properly closed container and labeled with patient identification. ▪ Recommended ratio of specimen size to fixative volume is 1:10. ▪ All tissues should be sent to the laboratory as soon as possible.	10% neutral buffered formalin
Resection specimen	▪ Fresh tissue must be sent directly from the operation theatre within 1 hour (without delay) during office hours only. ▪ If operation ends an hour before 5.10 pm, the tissue specimen must be immediately placed into 10% neutral buffered formalin.	WITHOUT fixative
Frozen Section	▪ All specimens must be sent fresh without any fixative/liquid and kept in a biohazard bag. ▪ All type of tissue specimen is illegible except for hard tissues such as bones and teeth as well as specimen containing any calcification. ▪ The specimen must be sent to lab with a completed filled request form by operating theatre attendant.	WITHOUT fixative

3.2 **Cytology:**

TYPES OF SAMPLE	COLLECTION AND REQUIREMENT	FIXATIVE
Body fluid	Specimen is collected into sterile containers (preferably 20 ml of sample volume) and labeled with patient identification. Specimen should be sent immediately to the laboratory at the same day of collection. If the specimen cannot be delivered immediately, it should be kept refrigerated at 2 - 8 °C (DO NOT FREEZE).	-
Brushing	Thin layer of smear is attached onto a clean slide, which has been labeled with patient's identification.	The slide is preserved immediately with 95% alcohol or spray fixative.
Cervical smear	Smear is attached onto a slide which has been labeled with patient's identification. The labeling should be written at frosted-end with using 2B pencils.	The slide is preserved immediately in a container containing 95% alcohol or fixed with spray fixative.
FNAC	Sample is taken by the Pathologist / Clinical Specialist / Medical Officer directly from patients. Smearing is done to several slides which has been labeled with patient's identification.	Some of the slides are preserved immediately with 95% alcohol and the rest are air dried.

4.0 SAMPLE SUBMISSION

4.1 All specimen sent to ADL should be attached with 2 copies of request form, which must be filled clearly with patient's information, clinical history and time/date of sample collection. The request forms will be provided by ADL upon request. Request forms from other institution are also accepted.

4.2 Both the specimen and request form sent must carry the same patient's identification.

4.3 Fresh specimen from the operation theatre can be sent directly without delay to the laboratory by 4.00pm on the same day, so that specimen can be examined fresh and necessary grossing can done.

4.4 Specimen collected after 4.00pm can be dispatched by the next working day morning after immersion in fixative.

4.5 Routine specimen fixed with proper fixative can be sent anytime (during office hours) directly to ADL counter.

5.0 ACCEPTANCE CRITERIA FOR VERBAL REQUESTS

5.1 Requests should be made by the requesting doctors.

5.2 Requests can only be accepted if the sample is sufficient and relevant to the cases.

6.0 REPORTING RESULTS

Turnaround Time (TAT)

SAMPLE	TAT
Histopathology	Results for simple cases (the cases which the diagnosis is based on Haematoxylin and Eosin only) will be released within 7 working days from the day of receiving. (Note: Result for urgent cases will be released within 5 working days.)
	Results for complex cases (the cases which the diagnosis require additional test such as Immunohistochemistry test, Histochemistry test or other related) will be released within 14 working days from the day of receiving. (Note: Result for urgent cases will be released within 10 working days)
	Result for second opinion will be released within 28 working days from the day of receiving.
Cytology	Results for gynaecologic test will be released within 10 working days from the day of receiving.
	Results for non-gynaecologic and FNAC test will be released within 5 working days from the day of receiving.
	Results for non-gynaecologic and FNAC test (the test which the diagnosis require additional tests such as cell block preparation, Immunohistochemistry test, Histochemistry test or other related) will be released within 10 working days from the day of receiving.

CHAPTER 4
GENETICS UNIT

CHAPTER 4

GENETICS UNIT

1.0 INTRODUCTION

Genetics Unit offers diagnostic investigations in genetics.

Genetics testing is divided into Cytogenetics and Molecular Cytogenetics. Cytogenetics is the study of the chromosome which includes analysis of G-banded chromosomes and other banding techniques. Molecular cytogenetics involves the combination of molecular biology and cytogenetics.

2.0 LIST OF AVAILABLE TESTS & INDICATIONS

2.1 Cytogenetics:

- Blood cytogenetics
- Bone marrow cytogenetics*

***Note: The bone marrow cytogenetics is not MS ISO 15189 accredited.**

2.2 Molecular Cytogenetics (FISH):

- DiGeorge Syndrome microdeletion probe
- Prader-Willi / Angelman Syndrome microdeletion probe
- Williams Syndrome microdeletion probe

2.3 Molecular Genetics (Molecular Diagnostics Unit):

- Sex-determining Region Y (SRY) gene (performed with cytogenetics test for Ambiguous genitalia case or sex related syndrome / abnormality).

Indications for cytogenetics test (karyotyping)	
Significant family history of	Chromosome rearrangements.
	Mental retardation of possible chromosomal origin.
	A relative with history of pregnancy losses, malformed fetus or stillbirth.
Patients with	Primary or secondary amenorrhea or premature menopause.
	Sperm abnormalities – azoospermia or oligospermia.
	Clinically significant abnormal growth – short stature, excessive growth, microcephaly, macrocephaly.
	Ambiguous genitalia.
	Abnormal clinical phenotype or dysmorphism.
	Congenital abnormalities.
	Mental retardation or developmental delay.
	Suspected deletion / microdeletion / duplication syndrome.
	X-linked recessive disorder in a female.
	Clinical features of a chromosome instability syndrome, including isolated haematologic findings monitoring after bone marrow transplantation.

Couples with	Recurrent pregnancy losses (3 or more); stillbirths, or neonatal deaths where it is not possible to study the affected conceptus.
	Child with chromosome abnormality or unusual variant.
	Unexplained infertility.
Indications for testing of SRY Gene test (PCR) – performed by Molecular Diagnostics Unit	
Referral reasons	Determination of new-born sex (ambiguous genitalia).
	Assessing the presence or absence of SRY in patients with abnormal sexual differentiation.

3.0 SAMPLE COLLECTION

3.1 All samples sent to the Genetics Unit should be requested via appointment. Appointment can be made by contacting **04-562 2692**. For non-urgent cases, samples can be sent on Monday and Tuesday. Samples for urgent cases to be sent on any working days and lab personnel have to be informed. **Urgent samples are:**

- **Patau Syndrome case (trisomy 13)**
- **Edwards Syndrome case (trisomy 18)**
- **Ambiguous Genitalia case**

3.2 Request form and anticoagulant lithium heparin tube (for internal only) will be provided by ADL (if requested by customer).

3.3 The ADL Chromosomal Studies request form shall **be complete and correctly filled** with necessary information:

3.3.1 Name (Patient)
3.3.2 Identification number (IC or passport number)
3.3.3 Sex
3.3.4 Date of birth (DOB)
3.3.5 Date and time of specimen taken
3.3.6 Hospital and ward or clinic
3.3.7 Father's and mother's name and age
3.3.8 Family history and pedigree
3.3.9 Patient investigation of unrecognized/suspected syndrome or sex abnormality
3.3.10 Name, signature and stamp of requesting **Specialist or Medical Officer (after consultation with the Specialist)**

3.4 Blood Cytogenetics, FISH and SRY gene:

- 3 ml of blood is collected in lithium heparin tube. **The blood should be mixed well to prevent clotting (mix by inversion 8 - 10 times).**
- For sub-standard samples, culturing may be attempted if repeat sampling would not be appropriate or possible as in case of death of the patient e.g.
  i. Sample clotted
  ii. Sample delayed for more than 48 hours
  iii. Non-viable sample (lysed, frozen, fixed)

- The blood should be transported on ice (**make sure not to freeze the blood**) and accompanied by a **complete and correctly filled request form and proof of payment**.

3.5 Bone Marrow Cytogenetics:

- At least 2.5 - 3ml sample is needed from the **first aspirate collected under sterile conditions**. The sample should be mixed well to prevent clotting.
- The sample should be transported on ice and accompanied by a complete and correctly filled genetic request form and proof of payment.

Note: Preparation of the test is very sensitive to the contamination. Make sure all the procedures are followed. Failure to follow the procedures will affect the result. We have the right to reject the samples.

4.0 SAMPLE TRANSPORTATION

All samples should be sent to the Genetics Unit as soon as possible (before 3 pm) on the day of appointment. All blood specimens should be sent to the laboratory within **24 hours** of collection. If there is an unavoidable delay between the collection of the sample and dispatch, the blood can be stored in a refrigerator at 4 °C. It is the responsibility of the requesting staff to ensure that samples arrive at the laboratory in suitable condition. Samples should be transported on ice without freezing.

5.0 ACCEPTANCE CRITERIA FOR ADDITIONAL TEST

5.1 Request should be made by the requesting / treating doctor.
5.3 Additional test criteria (have to informed lab and followed by formal request).
5.4 Additional test such as FISH can be requested later within 1 week of sample received but limited to available FISH probe and sample.
5.5 Additional test will be charged separately.

6.0 REPORTING RESULTS

Turnaround Time (TAT) – Blood cytogenetics

CASE	TAT
Urgent cases for cytogenetic	Within 14 days from the date sample is received
Non-urgent cases for cytogenetic	Within 28 days from the date sample is received
Cases that need second opinion and further additional investigation	Within 40 days from the date sample is received

*Turnaround Time (TAT) – Bone marrow cytogenetics: Within 2 months

Final results will be issued when chromosome analysis is completed and validated by specialists. Reports include a karyotype (latest ISCN guideline) where appropriate and clinically relevant interpretative comment. Results of non-urgent cases will be sent by post while results for urgent cases will be faxed to the requesting hospital / clinic / ward and followed by formal report (post).

CHAPTER 5

MOLECULAR DIAGNOSTICS UNIT

CHAPTER 5

MOLECULAR DIAGNOSTICS UNIT

1.0 INTRODUCTION

Molecular Diagnostics Unit* offers diagnostic investigations in molecular diagnostics by applying molecular biology to medical testing. Technique that is widely used in molecular diagnostics is Polymerase Chain Reaction (PCR). Some tests carried out by Molecular Diagnostic Unit are additional / further tests to support other units in ADL.

***Note: The tests in Molecular Diagnostics are not MS ISO 15189 accredited.**

2.0 LIST OF AVAILABLE TESTS

2.1 Polymerase Chain Reaction (PCR):

- Southeast Asian Ovalocytosis (SAO)
- Sex-determining Region Y (SRY) gene*

2.2 DNA Extraction:

- Blood samples
- Tissue samples: chorionic villus sampling, fresh tissue sample

2.3 Microdeletion Y Screening*

2.4 Multiplex PCR for Molecular Alpha Thalassemia**

Note:

***Normally samples are sent from Genetics Unit for ambiguous genitalia or other sex related syndrome / abnormality.**

****Normally samples are sent form Haematology Unit.**

3.0 SAMPLE COLLECTION

3.1 All samples sent to the Molecular Diagnostics Unit should be requested by contacting **04-562 2293**. Samples can be sent from Monday to Thursday accompanied by a complete and correctly filled Molecular Diagnostic Request Form.

3.2 The request should be accompanied with an Informed Consent for DNA test (except for DNA extraction request).

3.3 PCR SRY gene:

- 2.5 ml of blood is collected in EDTA tube. **However, if the sample is send together with chromosomal study to Genetics Unit, the blood should be collected in lithium heparin tube***. The blood should be mixed well to prevent clotting.

  ***Note: Please refer to Chapter 4 for more details.**

- The blood should be transported in ice (make sure not to freeze the blood) and accompanied by a complete and correctly filled Molecular Diagnostic Request Form.

3.4 PCR SAO:

- 2.5 ml of blood is collected in EDTA tube. The blood should be mixed well to prevent clotting.
- For prenatal screening, chorionic villus sampling (CVS) can be sent to Molecular Diagnostic Unit together with **parents' blood sample**.
- The blood should be transported in ice (make sure not to freeze the blood) and accompanied by a complete and correctly filled Molecular Diagnostics Request Form.
- **The request must be accompanied with a copy of recent FBP (< 3 months) result.**
- **All paediatrics (≤ 12 years old) samples must be accompanied with parents' samples.**

3.5 Blood DNA Extraction:

- 2.5 ml of blood is collected in EDTA tube. The blood should be mixed well to prevent clotting.
- The blood should be transported in ice (make sure not to freeze the blood) and accompanied by a complete and correctly filled Molecular Diagnostics Request Form.

3.6 Microdeletion Y Screening:

- 2.5 ml of blood is collected in EDTA tube. The blood should be mixed well to prevent clotting.
- The blood should be transported in ice (make sure not to freeze the blood) and accompanied by a complete and correctly filled Molecular Diagnostics Request Form.

3.7 Multiplex PCR Molecular Alpha Thalassaemia:

- 2.5 ml of blood is collected in EDTA tube. The blood should be mixed well to prevent clotting.
- The blood should be transported in ice (make sure not to freeze the blood) and accompanied by a complete and correctly Molecular Diagnostics Request Form.
- **The request must be accompanied with a copy of recent FBP (< 3 months) result and Hb Analysis result.**
- **All paediatrics (≤ 12 years old) samples must be accompanied with parents' samples.**

4.0 SAMPLE TRANSPORTATION

All samples should be sent to the Molecular Diagnostic Unit within 48 hours of collection. After delivering the sample/s please inform the laboratory staff by contacting **04-562 2293** to make sure that the lab is aware there is/are sample/s. If there is an unavoidable delay between the collection of the sample and dispatch, the sample can be stored in a refrigerator at 4 °C. It is the responsibility of the requesting staff to ensure that samples arrive at the laboratory in suitable condition. Samples should be transported in ice without freezing.

5.0 ACCEPTANCE CRITERIA FOR VERBAL REQUESTS

5.1 Request should be made on same day sample is sent to the lab.

5.2 Request should be made by the requesting / treating doctor.

6.0 REPORTING RESULTS

Turnaround Time (TAT)

TEST	TAT
PCR SRY	Within 14 working days from the date sample is received
PCR SAO	Within 14 working days from the date sample is received
DNA extraction	Within 7 working days from the date sample is received
Microdeletion Y Screening	Within 14 working days from the date sample is received
Multiplex PCR for Molecular Alpha Thalassemia	Within 14 working days from the date sample is received

CHAPTER 6
HAEMATOLOGY UNIT

CHAPTER 6

HAEMATOLOGY UNIT

1.0 INTRODUCTION

Haematology Unit offers diagnostic investigations in haematology.

2.0 LIST OF AVAILABLE TESTS

2.1 **Full Blood Count (FBC)**

- Haemoglobin (Hb)
- Total White Blood Cell (TWBC)
- Total Red Blood Cell (TRBC)
- Platelet
- Packed Cell Volume (PCV)
- MCV, MCH, MCHC

2.2 **Full Blood Picture (FBP)**

- Haemoglobin (Hb)
- Total White Blood Cell (TWBC)
- Total Red Blood Cell (TRBC)
- Platelet
- Packed Cell Volume (PCV)
- MCV, MCH, MCHC
- Differential White Cell (DC)
- Reticulocytes
- Peripheral Blood Film (PBF)

2.3 Routine Coagulation Test (PT / APTT/INR)*
2.4 DIC Screening
2.5 D-Dimer (qualitative)
2.6 Erythrocytes Sedimentation Rate (ESR)*
2.7 G6PD Screening (qualitative)*
2.8 Haemoglobinopathy Analysis (High Performance Liquid Chromatography)*
2.9 Haemoglobin Electrophoresis*

***Note: The marked tests are not MS ISO 15189 accredited.**

3.0 SAMPLE COLLECTION AND TRANSPORTATION

3.1 Sample collection

3.1.1 A correct amount of blood volume as stated on the tube is collected into the appropriate anticoagulant for the test requested. To ensure the volume of blood collected is correct, fill the tube to the indicator line.

3.1.2 The blood should be mixed gently and thoroughly for 1 minute by a rotary wrist movement and stored in ice before send to laboratory.

3.1.3 All specimens should be sent to the laboratory immediately after collection. Delayed transportation of sample may affect the result.

3.1.4 Sample for urgent cases need to be informed by contacting **04-562 2693**.

3.1.5 Every specimen should be accompanied by details of clinical history and diagnosis in the electronic request or a completely filled request form.

3.1.6 All samples to be sent to Haematology Unit should use specific containers according to test requested.

NO	TEST		REQUIRED VOLUME AND CONTAINER TYPE
1.	Full Blood Count (FBC)		Blood in EDTA tube
2.	Full Blood Picture (FBP)		Blood in EDTA tube
3.	Routine Coagulation Test (PT / APTT / INR)		Blood in Tri-sodium Citrate tube
4.	DIC Screening		Blood in Tri-sodium Citrate tube
5.	D-Dimer		Blood in Tri-sodium Citrate tube
6.	ESR		Blood in EDTA tube
7.	G6PD Screening		Blood in EDTA tube / filter paper
8.	Haemoglobinopathy Analysis		Blood in EDTA tube
9.	Haemoglobin Electrophoresis		Blood in EDTA tube
10.	Thalassemia Screening	FBP	Blood in EDTA tube
		Hb Quantitative	
11.	Thalassemia Confirmation	Hb Quantitative	2 tubes of blood in EDTA tube
		Hb Electrophoresis	
		Polymerase Chain Reaction, if necessary*	

Notes: 1. All specimen collection with EDTA, Tri-sodium Citrate and Lithium Heparin tube should be mixed gently and thoroughly for 1 minute by a rotary wrist movement.

2. Sample in Tri-sodium Citrate tube should be transported to the lab in ice.

3*. Please refer to Chapter 5: Molecular Diagnostics Unit for more detail of sample collection and transportation.

4.0 COAGULATION AND THROMBOPHILIA TEST

4.1 Guidelines for coagulation test

NO	INDICATIONS	TEST
1.	Warfarin therapy control	PT (Prothrombin Time)
2.	Heparin therapy control	APTT (Activated Partial Thrombin time)
3.	DIC screening	PT, APTT, Fibrinogen, Di Dimer and platelet.
4.	Liver biopsy	PT (Prothrombin Time)
5.	Pre-operative cases	PT, APTT

4.2 Guidelines for specimen collection

4.2.1 Good specimen collection e.g. clean venepuncture with minimal stasis, not from indwelling catheters or arterial lines, mix well by inverting 5-6 times gently.

4.2.2 Recommended to use a 21-gauge needle or butterfly. 19 gauge may be used in adults with good veins: 23 gauge may be required for infants.

4.2.3 Do not use heparin-contaminated venous lines. If unavoidable, flush the lines with crystalloid and discard first few millilitres of blood.

4.2.4 Correct ratio 1 part of sodium citrate to 9 parts of blood is essential. Collect blood to the indicator line on the tube to ensure correct amount of blood collected.

4.2.5 Keep the blood (in collection tube) in ice.

4.2.6 Send blood to the laboratory immediately. The sample ideally tested within 1 hour of blood collection.

4.2.7 Note down exact time blood is collected.

4.2.8 If haematocrit is >0.55, contact laboratory for a tube with adjusted volume of anticoagulant.

4.3 Coagulation screening test

Where a patient is suspected of having a bleeding disorder, PT, APTT, Fibrinogen and FBP should be performed as a preliminary screen. Bleeding time should be done after consulting Haematologist. If results are abnormal, or in any case of doubt, the attending clinician should consult Haematologist, full coagulation studies will then be arranged if indicated.

5.0 ACCEPTANCE CRITERIA FOR VERBAL REQUESTS

5.1 Request should be made less than 4 hours after sample is received.

5.2 Request can only be accepted if the sample is sufficient.

5.3 Requested test is suitable to sample.

5.4 Request should be made by requesting doctor.

6.0 REPORTING RESULTS

Turnaround Time (TAT)

CASE	TAT
Urgent cases for FBC & FBC-DC	Within 40 minutes from the time sample is received
Non-urgent cases for FBC & FBC-DC	Within 160 minutes from the time sample is received
Urgent cases for FBP	Within 2 working days from the time sample is received
Non-urgent cases for FBP	Within 5 working days from the time sample is received

7.0 NORMAL RANGES

7.1 Normal range of full blood count for children (up to 12 years old)

VARIABLES	BIRTH	DAY 1 - DAY 3	DAY 4 - DAY 7	DAY 8 - DAY 14	DAY 15 - 1 MONTH	2 MONTHS	3 - 6 MONTHS	6 MONTHS - 2 YEARS	2 - 6 YEARS	6 - 12 YEARS
WBC ($x10^9$/l)	10 - 26	7 - 23	6 - 22	6 - 22	5 - 19	5 - 15	6 - 18	6 - 16	5 - 15	5 - 13
RBC ($x10^{12}$/l)	5.0 - 7.0	4.0 - 6.6	3.9 - 6.3	3.6 - 6.2	3.0 - 5.4	3.1 - 4.3	4.1 - 5.3	3.9 - 5.1	4.0 - 5.2	4.0 - 5.2
Hb (g/dl)	14.0 - 22.0	15.0 - 21.0	17.1 - 17.9	16.1 - 16.9	11.5 - 16.5	9.4 - 13.0	11.1 - 14.1	11.1 - 14.1	11.0 - 14.0	11.5 - 15.5
HCT (%)	45 - 70	45 - 67	42 - 66	31 - 71	33 - 53	28 - 42	30 - 40	30 - 38	34 - 40	35 - 45
MCV (fl)	100 - 120	92 - 118	88 - 126	86 - 124	92 - 116	87 - 103	68 - 84	72 - 84	75 - 87	77 - 95
MCH (g/dl)	31 - 37	31 - 37	31 - 37	31 - 37	30 - 36	27 - 33	24 - 30	25 - 29	24 - 30	25 - 33
MCHC (g/dl)	300 - 360	290 - 370	280 - 380	280 - 380	290 - 370	285 - 355	300 - 360	320 - 360	310 - 370	310 - 370
Platelets ($x10^9$/l)	100 - 450	210 - 500	160 - 500	170 - 500	200 - 500	210 - 650	200 - 550	200 - 550	200 - 490	170 - 450
RDW SD	-	-	-	-	-	-	-	-	-	-
RDW CV	-	-	-	-	-	-	-	-	-	-
Neutrophils ($x10^9$/l)	4 - 14	3 - 5	3 - 6	3 - 7	3 - 9	1 - 5	1 - 6	1 - 7	1.5 - 8	2 - 8
Lymphocytes ($x10^9$/l)	3 - 8	2 - 8	3 - 9	3 - 9	3 - 16	4 - 10	4 - 12	3.5 - 11	6 - 9	1 - 5
Monocytes ($x10^9$/l)	0.5 - 2.0	0.5 - 1.0	0.1 - 1.7	0.1 - 1.7	0.3 - 1.0	0.4 - 1.2	0.2 - 1.2	0.2 - 1.0	0.2 - 1.0	0.2 - 1.0
Eosinophils ($x10^9$/l)	0.1 - 1.0	0.1 - 2.0	0.1 - 0.8	0.1 - 0.9	0.2 - 1.0	0.1 - 1.0	0.1 - 1.0	0.1 - 1.0	0.1 - 1.0	0.1 - 1.0
Basophils ($x10^9$/l)	-	-	-	-	-	-	-	-	-	-
Reticulocytes ($x10^9$/l)	120 - 400	50 - 350	50 - 100	50 - 100	20 - 60	30 - 50	40 - 100	30 - 100	30 - 100	30 - 100

Reference: Dacie and Lewis Practical Haematology, S.M.Lewis, B J Bain and I Bates 12th Edition 2016

7.2 Normal range of full blood count for adult

VARIABLES	MALE		FEMALE
	<60	≥60	
WBC ($x10^9$/l)	4.078 - 11.370		
RBC ($x10^{12}$/l)	4.53 - 5.95	3.86 - 5.62	3.87 - 5.21
Hb (g/dl)	13.5 - 17.4	11.8 - 16.9	11.6 - 15.1
HCT (%)	40.1 - 50.6	35.7 - 48.9	35.1 - 44.9
MCV (fl)	80.6 - 95.5		
MCH (g/dl)	26.9 - 32.3		
MCHC (g/dl)	31.9 - 35.3		
Platelets ($x10^9$/l)	142 - 350		171 - 399
RDW SD	37.5 - 48.1		
RDW CV	12 - 14.8		
Neutrophils ($x10^9$/l)	3.929 - 7.147		
Lymphocytes ($x10^9$/l)	1.847 - 4.807		
Monocytes ($x10^9$/l)	0.385 - 1.141		
Eosinophils ($x10^9$/l)	0 - 0.827		
Basophils ($x10^9$/l)	0 - 0.095		
Reticulocytes ($x10^9$/l)	0.4 - 1.6		

Note: < 60 = Below 60 years old, **≥** 60 = 60 years old and over

Reference: Ambayya A *et al.*, (2014) Haematological Reference Intervals in a Multiethnic Population. *PLoS ONE*

7.3 Coagulation Tests

VARIABLES	ANALYZER REAGENT	STA COMPACT DIAGNOSTIC STAGO
PT (sec)	STA-Neoplastine CI Plus 5	12.2 - 14.2
INR		0.97 - 1.18
APTT (sec)	STA-PTT Automate 5	31.7 - 44.0
Fibrinogen level (g/l)	STA-Liquid Fib	2.32 - 4.44
D-dimer (ug/ml)	STAGO D-Dimer Test	<0.5 / (-)

7.4 Special Haematology Tests

- G6PD screening test
  - Normal: fluorescent seen

- Haemoglobin Analysis

Hb Electrophoresis

Hb A (%)	96.0 - 98.5
Hb A_2 (%)	1.5 - 4.0 (electrophoresis technique) 2.43 -4.29 (acid elution technique)
Hb F (%)	<1.0 (electrophoresis technique) 0.11 - 0.99 (modified Betke technique)
HPLC (%)	1.75 - 3.25

8.0 PANIC VALUES

Panic value is defined as any result outside the normal ranges to a degree that may pose immediate health risk to patients or require immediate action on behalf of the treating doctors. Laboratory is responsible to immediately NOTIFY doctors of the panic values.

8.1 Haematology Tests

8.1.1 For Adults

TEST	LOWER CRITICAL LIMIT	UPPER CRITICAL LIMIT	UNIT
Hb	6	19	g/dl (adult)
Hct	20	60	% (adult)
Platelet	20	1000	X 10^9 /L
WBC	1	50	X 10^9 /L

8.1.2 For Paediatrics

TEST	LOWER CRITICAL LIMIT	UPPER CRITICAL LIMIT	UNIT
Hb	7	20	g/dl (adult)
Hct	20	40	% (adult)
Platelet	50	1000	X 109 /L
TWBC	2	50	X 109 /L

8.1.3 For Neonate

TEST	LOWER CRITICAL LIMIT	UPPER CRITICAL LIMIT	UNIT
Hb	8	22	g/dl (adult)
Hct	25	70	% (adult)
Platelet	50	1000	X 109 /L
TWBC	2	50	X 109 /L

8.2 Coagulation Tests

VARIABLES	PANIC VALUES
PT (sec)	>40
APTT (sec)	>100
INR	>5

References:

a) Stanford University Medical Center, Hospital and Clinic school of medicine critical values
b) www.medicine.uiowa.edu/path_handbook/Appendix/Common/UN_CRIT_LAB_VAL.html critical value for hematology
c) depts.washington.edu/labweb/PatientCare/Clinical/Critical.htm critical value for hematology
d) DLMP Critical Values / Critical Results List [CL 041647.004]
e) Quick Guide for Notification of Critical Laboratory Results in MOH Hospitals, Institute for Health Systems Research, 2010
f) Macking, et al. Guideline on the laboratory aspects of assays used in haemostasis and thrombosis, International Journal of Laboratory Hematology, August 2012

CHAPTER 7
TRANSFUSION MEDICINE UNIT

CHAPTER 7

TRANSFUSION MEDICINE UNIT

1.0 INTRODUCTION

The procurement of blood, the provision of safe and quality blood products and pre-transfusion testing are the main components of the blood bank. Thus this User Manual serve as guidelines and reference for the clinicians, laboratory personnel and other paramedical staffs to improve the quality of blood transfusion services.

2.0 LIST OF TESTS, PRODUCTS AND SERVICES

2.1 List of tests

TEST	ROUTINE	STAT	SPECIAL
Group & Crossmatch (GXM)	✓	✓	
Group, Screen and Hold (GSH)	✓	✓	
ABO and Rh Grouping	✓	✓	
Rhesus Phenotyping	✓		
Antenatal Study	✓	✓	
Neonatal Study	✓	✓	
Direct Anti Globulin Test	✓	✓	
Antibody Screening	✓		
Antibody Identification	✓		
Antigen Phenotyping	✓		
Investigation of Transfusion Reaction	✓		
Isohemagglutinin titre			✓
Antibody titre			✓
Adsorption and Elution Test			✓
Saliva secretor status			✓

Short turn-around time (STAT) or urgent request can be made through phone call.
An appointment need to be made for request of special test.

2.2 List of blood products

	PRODUCTS	DETAILS
ROUTINE	Blood Components	The blood components available are: a) Whole Blood b) Packed Red Blood Cells c) Random Platelet d) Fresh Frozen Plasma e) Cryoprecipitate f) Cryosupernatant
	Apheresis	Blood components such as platelet, plasma or red cells are obtained through apheresis technique using automated cell separation equipment.
SPECIAL	Leuco-reduced packed cells	The leucocyte is reduced to less than 5 x 10^6 per unit by using a filter. Leucocyte reduced blood is indicated for the following purposes: a. To prevent febrile non-hemolytic transfusion reactions. b. To minimize the risk of transfusion-related CMV transmission. c. To minimize HLA alloimmunization.
	Irradiated blood component	The red cell or platelet is irradiated to inactivate lymphocytes to prevent transfusion-associated graft-versus-host disease (TA-GVHD).

2.3 List of service

SERVICES	DETAILS
Blood donation	In-house blood donation is opened to staffs and public from 8.30 am till 4.30 pm on working days. Types of donation include whole blood donation and apheresis donation. Apheresis donation is by appointment.
Blood donation campaign	Blood donation campaign is held whenever is necessary inside or outside of TMU.
Blood Irradiator	It is used to provide irradiated blood components. This procedure is performed from 8.30 am till 4.30 pm on working days.
Stem cell collection	Stem cells are collected via leucapheresis using automated cell separation equipment. The collection is by appointment

3.0 LIST OF FORMS

Transfusion Medicine Unit provides 5 types of form as below

FORM	PURPOSE
Borang Permohonan Transfusi Darah	To request GXM and GSH
Immunohematology Test Request Form	To request single immunohematology test other than blood components
Slip Pengambilan Darah /Komponen Darah	To collect blood components from TMU
Transfusion Reaction Request Form	To request investigation of transfusion reaction
Borang Pemulangan Darah Tidak Digunakan	Used for returning of unused/ remnant blood component to the laboratory

4.0 ORDERING BLOOD FOR TRANSFUSION

The clinician managing the patient shall be responsible for prescribing blood for the patient. The clinician should contact and discuss with the transfusion medicine specialist where necessary.

5.0 CONSENT FOR TRANSFUSION

5.1 The patient must give written informed consent prior to transfusion.

5.2 The clinician in charge of the patient shall explain to the patient the indication, benefits, risks and alternatives to transfusion therapy, and ensure that the patient understands the issues discussed.

5.3 If for any reason, the patient is unable to personally give consent, a responsible family member of the patient shall be asked to do so. If no such family member is available, or in emergencies when the need for transfusion leaves no time for consent, the decision shall be made by two fully registered medical practitioners. This decision shall be clearly documented.

6.0 SPECIMEN COLLECTION

6.1 Patient identification

The phlebotomist shall ensure that the patient is correctly identified by:

a) Asking the patient to state their full name and IC number (or use of at least 2 identifiers) in open-ended questions.

b) Check the answers given against the information stated on the patient's identification wristband and/or case notes.

6.2 If it is not possible to identify the patient in the above manner, the phlebotomist shall identify the patient by asking the relative or career to name the patient and then check the answer given against the information stated on the patient's identification wristband and case notes.

7.0 BLOOD SAMPLING

7.1 The procedure shall be carried out as one process by one person at the bedside.

7.2 Only one patient shall be attended to at any one time till completion.

7.3 The phlebotomist shall clearly and accurately label the blood sample at the patient's bedside immediately after blood taking.

7.4 Use of pre-printed label is not encouraged. If this cannot be avoided, the ward / clinic should implement a procedure to ensure that patients are correctly identified using the printed labels.

7.5 Information on the label shall include, at the minimum, the patient's full name, hospital registration number / identity card number, the date and time of collection and the initial of the phlebotomist.

8.0 BLOOD SAMPLE AND CONTAINER FOR RED CELLS TRANSFUSION

Collect the required amount of blood into the appropriate sample tube as below. Specimen collection with anticoagulant tube should be mixed gently and thoroughly for 1 minute by a rotary wrist movement.

8.1 Infant up to 4 months old

a) The sample to be taken from the infant shall be in EDTA pediatric tube (volume is as per stated on the tube container)

b) Blood sample in EDTA tube shall be also taken from the mother (volume is as per stated on the tube container).

c) Both samples shall be sent to the TMU together under a single request.

d) No further sample is required for repeat transfusion of the same set of the paedipack. However, infant's sample is required for subsequent transfusion if another set of paedipack is going to be issued. For this, crossmatching will be performed using the infant's sample.

8.2 Older than 4 months old

a) The sample to be taken shall be in EDTA tube (volume is as per stated on the tube container).

b) If a patient requires repeated red cell transfusion, each request for red cells shall be accompanied by a new request form and a new blood sample in EDTA tube (volume is as per stated on the tube container).

8.3 Elective cases

a) For elective cases, samples should be sent to the TMU during office hours, at least 24 hours before the blood is required.

b) Except for rare blood groups and/or Rh(D) negative where the TMU should be informed at least 5 working days in advance.

9.0 BLOOD SAMPLE AND CONTAINER FOR COMPONENTS (OTHER THAN RED CELLS) TRANSFUSION

9.1 A new request for blood component other than red cells shall be accompanied by blood sample taken in EDTA tube.

9.2 For a patient who has at least two previous blood grouping records at the TMU, a new blood sample need NOT accompany the request for blood component. However, a copy of the previous request form clearly stating the blood grouping results shall be attached to the new request form.

9.3 If previous request form is not available, a fresh blood sample shall be sent to the laboratory.

10.0 BORANG PERMOHONAN TRANSFUSI DARAH

All requests for blood/blood components must be made using '*Borang Permohonan Transfusi Darah'.*

a) The request form should be completely filled and contains relevant patient information
b) Write the name of staff that takes and labels the sample.
c) The quantity and the approximate time when the blood and blood component would be required must be stated. Requests for blood to be made available 'as soon as possible' should be avoided as this would not assist the blood bank personnel in determining priorities.
d) The clinician shall ensure that each request form is completed, sign and clearly state his name in block letters or stamped.
e) A different request form must be used for different blood components.
f) Crossmatched blood will be kept for a further 48 hours.

11.0 IMMUNOHAEMATOLOGY TEST REQUEST

11.2 All single request for another immunohematology test (other than a request for transfusion) must be made using 'Immunohematology Request Form'.

11.3 A copy of request form should be completely filled with patient information and clearly tick/ write the test request.

11.4 The sample to be taken from the infant shall be in EDTA pediatric tube (volume is as per stated on the tube container)

11.5 The sample to be taken for a patient older than 4 months shall be in EDTA tube (volume is as per stated on the tube container).

12.0 GROUP, SCREEN AND HOLD CASES (GSH)

12.1 Tick the appropriate box for a GSH request.

12.2 GSH is a procedure that consists of ABO and RhD grouping, and antibody screening for the patient's sample. The patient's serum or plasma is subsequently retained for a minimum of 48 hours.

12.3 It is recommended only for cases where there is a higher chance of requiring a blood transfusion during admission.

12.4 For elective clinical procedures, GSH shall be requested in accordance with the established Maximum Surgical Blood Ordering Schedule (MSBOS).

12.5 Should the patient require transfusion following GSH, blood will be made available on time.

12.6 In case of unexpected antibody was detected, the medical officer in charge will be informed. The laboratory will attempt to supply compatible blood for the patient if required.

12.7 Preferably, the request for GSH is reached at the transfusion laboratory at least 24 hours prior to the operation.

13.0 GROUP & CROSS-MATCH (GXM)

13.1 GXM consists of checking ABO & RhD grouping and antibody screening for the patient's sample and crossmatching patient and donor unit for compatibility.

13.2 GXM shall be requested for cases with high certainty for transfusion at that time.

13.3 Requests for non-urgent cases such as thalassemia, leukemia, and anemia should be sent preferably before 3.30 pm during normal office hours. This is to ensure that the Medical Laboratory Technologist (MLT) on-call can attend promptly and properly to the urgent requests.

13.4 The approximate time when the blood is needed should be stated on the request form.

13.5 Crossmatched blood will be kept for a maximum of 48 hours only. The blood will be automatically canceled on the third day of the request, unless the ward staff or medical officer informs for further reservation.

14.0 TRANSFUSION IN CASES OF LIFE-THREATENING BLEEDING

The choice of blood for transfusion in cases of life-threatening bleeding is dependent on the urgency for transfusion and the time available. The options available are:

14.1 Uncrossmatched Group O RhD positive packed red cells (Safe O)

a) In Transfusion Medicine Unit, commonly Group O RhD positive packed cells are used as Safe O. Safe O can be used for resuscitation in a dire emergency while waiting for group-specific or crossmatched blood to be available.

b) Any decision to use Safe O shall only be made after the clinician has carefully assessed the urgency of the patient's need for blood. The requesting doctor shall clearly state the reasons for the transfusion in the patient's records and in the request form.

c) Tick 'serta merta tanpa ujian keserasian darah' box in the request form.

d) A pre-transfusion blood sample must be collected from the patient before starting transfusion with 'Safe O'. Send this sample together with the request form.

14.2 Uncrossmatched group specific packed cells

a) If the blood group of the patient is known, uncrossmatched group specific blood may be given.

14.3 Emergency crossmatch

a) If the blood is required urgently (within 30 minutes), tick 'Segera' box on the request form

b) The laboratory will issue units of blood that are found to be compatible at immediate spin after 5 minutes incubation at room temperature.

For Safe O, uncrossmatch and emergency crossmatch request,

a) The laboratory will proceed to completion of the compatibility testing and antibody screening of the units of blood issued, at 37 °C and in the AHG phase.
b) If there is any incompatibility detected, it will be immediately informed to the clinician concerned for appropriate action.
c) It should be accompanied by a phone call to the TMU to facilitate the process.

15.0 NEONATAL TRANSFUSION

15.1 Blood used for neonatal transfusions shall be compatible with the mother's blood. For that reason, samples from mother and neonate should be obtained.

15.2 The volume of blood to be transfused is calculated based on the neonate's body weight.

Volume required (mL) = body weight (kg) x Hb rise required g/dL x transfusion factor (0.4)

16.0 CONTAINER, TRANSPORT AND SAMPLE MANAGEMENT

TEST	CONTAINER	TRANSPORT	STORAGE
All	EDTA	Place in biohazard plastic. Sent without ice as soon as possible	In case of delayed transportation, keep the sample in the refrigerator within 2°C - 6°C.

17.0 ISSUE, TRANSPORT AND STORAGE OF BLOOD

17.1 Issue of blood and blood components

a) The ward personnel collecting the blood shall bring '*Slip Pengambilan Darah / Komponen Darah'* and an appropriate container.
b) The laboratory will provide **Bed Head Ticket (BHT) sticker** to accompany the blood product.


IPPT USM BERTAM

B

Lab No 100913

R/N PI[redacted]

POS Collected 2018-06-12 Expires 2018-07-24

Bag No: 415015327111

PC350

12:28:07 / 2018-06-25

This unit is designated for transfusion to:

ABDUL RAHMAN BIN HASSAN

I/D [redacted] Age 70Yrs Race MALAY Sex M Ward IN PATIENT

Transfusion Details

Date	Time Start	Stopped	Vol.transf.	Done By	Reaction Yes / No
Details					

IMPORTANT:- FILL IN & RETURN UPPER SECTION TO BLOOD BANK

Lower Section: Detach and attach to patient file

Lab No 100913 PC350 R/N PID10025527 Bag No 415015327111 Collected 2018-06-12 Expires 2018-07-24

ABDUL RAHMAN BIN HASSAN

I/D 471117025203 Age 70Yrs Race MALAY Sex M Ward IN PATIENT ABO/Rh B POS

Transfusion Details

Date	Time Start	Stopped	Vol.transf.	Done By	Reaction Yes / No
Details					

c) The laboratory personnel and ward personnel shall verify that the particulars of patient match those of the blood to be issued.
d) The laboratory personnel who issued the blood, shall record their name in the request form.
e) The person collecting the blood shall record their name, time of collecting and initial in the request form.
f) For all wards and private hospitals, which do not have a dedicated blood refrigerator, all cross-matched blood will be kept in the blood bank. Blood must be collected from the blood bank only when it is required for transfusion.
g) For wards and private hospitals, which have a dedicated blood refrigerator, blood can be collected for transfusion during normal office hour. However, it is very important that the blood must be stored at 2 $^{\circ}$C to 6 $^{\circ}$C in the dedicated blood refrigerator without any undue delay. To maintain the quality of the blood, there must be a quality control program for the dedicated blood refrigerator. The blood bank will assist in the setting up of a quality control program if requested.

17.2 Transport and Storage

a) The ward personnel shall transport the issued blood to the ward or returned blood to the TMU without delay.
b) Transportation shall be carried out in an appropriate temperature
c) Issued blood shall be transfused without undue delay. However, in the event where the delay is inevitable, the ward shall maintain the blood at the appropriate temperatures and condition until they are used or returned to the blood bank immediately.

	WHOLE BLOOD/ RED CELLS	PLATELET	PLASMA COMPONENTS (FFP/ CRYO)
SUPPLY	After crossmatch	Group-specific	Group-specific After thawed
CONTAINER	Blood box with ice	Blood box without ice	Blood box with ice
USE	As soon as possible	Transfuse immediately	Transfuse immediately
STORAGE	2°C to 6°C	Room temperature 20°C to 24°C on the continuous agitator	Should not be stored in the ward
AFTER USE	Fill up BHT and return together with an empty bag to laboratory	Fill up BHT and return together with an empty bag to laboratory	Fill up BHT and return together with an empty bag to laboratory

a) Whole blood and Packed Cells must not have direct contact with ice packs. There should be a cloth / cardboard in between them to avoid hemolysis.
b) Containers for the transportation of platelets must not contain any ice packs. However, for long-distance transportation of platelets in hot and humid conditions or in a non-air-conditioned transport, an ice pack may be put at the bottom of the container in order to sustain an ambient temperature for the viability of the platelets. The platelet must never come into contact with the ice pack.

18.0 IDENTIFICATION CHECK PRIOR TO TRANSFUSION

18.1 Each hospital shall establish a procedure for carrying out identification checks, to prevent any error occurring at this final stage before transfusion commenced.

18.2 A check shall be conducted to ensure that the patient's information matches those on the:

a) Blood bag label
b) Bag Head Ticket / blood compatibility label
c) Patient's identification
d) Patient's blood request form
e) Case notes

18.2 The blood shall also be checked to ensure that it has not expired and that it conforms to the following in appearance:

a) No change in color
b) Absence of clots
c) No foamy appearance
d) No leakage

18.3 In the event of any discrepancy in the identification check of the intended recipient, blood compatibility label, request form and blood component, the laboratory shall be immediately informed. The implicated blood shall be immediately returned to the blood bank for appropriate measures to be taken.

19.0 DISCONTINUED TRANSFUSION

19.1 Any blood remaining from a discontinued transfusion shall not be used.

19.2 Remnants of blood shall be returned to the TMU immediately together with completed form of '*Borang Pemulangan Darah Yang Tidak Digunakan'*.

20.0 RETURN OF UNUSED BLOOD / BLOOD COMPONENT

20.1 The ward shall return all un-transfused blood immediately to laboratory accompanying with completed form of '*Borang Pemulangan Darah Yang Tidak Digunakan'.*

20.2 The ward shall inform the laboratory if any of the un-transfused blood returned to the laboratory has not complied with the storage or transportation temperature.

21.0 RETURN OF USED BLOOD BAG

21.1 Upon completion of blood transfusion, the ward staff must ensure that the BHT tag / compatibility label attached to each bag of blood component is filled completely and returned to laboratory together with the used blood bag.

22.0 TRANSFUSION REACTION

22.1 If an adverse transfusion reaction is detected or suspected, the transfusion shall be stopped immediately. A doctor shall immediately assess and stabilize the patient. Further management depends on the type and severity of the reaction.

22.2 All transfusion reaction must be investigated. The request must be made using '*Transfusion Reaction Request Form'*.

22.3 To facilitate investigation of an adverse transfusion reaction, the following shall be carried out:

a) Preserved the blood bag and tubing set with all attached labels
b) Take blood samples in two EDTA tubes
c) Take blood sample in one plain tube
d) Prepare blood culture sample
e) Prepare urine sample in a plastic container
f) Filled BHT tag / blood compatibility label

22.4 Label specimens as "Post-transfusion" with proper identification

22.5 Send all investigations sample and request form to TMU immediately.

22.6 The ward shall keep the transfused blood bag and its transfusion set under appropriate conditions to ensure the integrity and to avoid microbial contamination.

22.7 For urticarial cases please send blood bag and '*Transfusion Reaction Request Form'* only.

23.0 ACCEPTANCE CRITERIA FOR VERBAL REQUESTS

22.1 The verbal request can be made to add-on blood components, additional testing or to inform cancellation of the test.

22.2 Acceptance of verbal request depend on

a. Request made less than 24 hours after the sample is received.
b. The request can only be accepted if the sample is sufficient.
c. The requested test is suitable to sample.
d. Request made by / on behalf of the medical officer

24.0 REJECTION CRITERIA

a. Incomplete relevant information on the request form
b. Wrong form
c. No name and signature of requesting doctors.
d. Information in the request form and sample container is different.
e. Wrong sample container
f. Mislabeling / incomplete labeling of sample
g. Unsatisfactory specimen when received, for example, spillages, breakages and etc.
h. The sample was taken and labeled by a different person
i. Blood hemolyzed
j. Blood clotted
k. Aged sample more than 48 hours
l. Insufficient sample

25.0 REPORTING RESULTS

Turnaround Time (TAT)

CASE	TAT
Emergency crossmatch	30 minutes
GSH	2 hours
GXM	3 hours
GSH convert to GXM	1 hour
ABO Rh (D)	2 hours
DCT	2 hours
Another immunohematology test	5 working days
Blood irradiation	2 hours
Leucofiltered	2 hours

CHAPTER 8

IMMUNOLOGY UNIT

CHAPTER 8

IMMUNOLOGY UNIT

1.0 INTRODUCTION

Immunology Unit offers diagnostic investigations in immunology.

***Note: The tests in Immunology are not MS ISO 15189 accredited.**

2.0 LIST OF AVAILABLE TESTS AND SAMPLE COLLECTION

All samples to be sent to Immunology Unit should use the specific containers according to the tests requested. Required volume, type of blood container and requirement of healthy control sample for stated test are listed in the table below:

PROFILE	TEST	CONTAINER	VOLUME	SPECIMEN	REMARKS
Immunoglobulin Level	Immunoglobulin G (IgG)	Plain tube	3.5 ml whole blood	Serum	-
	Immunoglobulin A (IgA)				
	Immunoglobulin M (IgM)				
Subclasses Immunoglobulin G	Subclasses Immunoglobulin G1 (IgG1)	Plain tube	3.5 ml whole blood	Serum	-
	Subclasses Immunoglobulin G2 (IgG2)				
	Subclasses Immunoglobulin G3 (IgG3)				
	Subclasses Immunoglobulin G4 (IgG4)				
Complement Assay	Complement 3 (C3)	Plain Tube	3.5 ml Whole blood	Serum	-
	Complement 4 (C4)				

PROFILE	TEST	CONTAINER	VOLUME	SPECIMEN	REMARKS
Standard Immuno-phenotyping Enumeration	T cell B cell Natural killer cell	EDTA tube	2 ml whole blood	Whole blood	Require healthy 2 ml whole blood control sample (any age matched or adult sample accepted). Strictly sent to our lab within 8 hours after blood withdrawal. Failure to comply may cause the sample to be rejected
		Paediatric EDTA tube	2 x 0.5 ml whole blood	Whole blood	
***Phagocyte Function Test [Phagoburst / dihydrorhodamine (DHR)]**		Lithium Heparin without gel	4 ml whole blood	Whole blood	Require healthy 4 ml whole blood control sample (any age matched or adult sample accepted). Strictly sent to our lab within 8 hours after blood withdrawal. Failure to comply may cause the sample to be rejected
		Paediatric Lithium Heparin	2 x 0.5 ml whole blood	Whole blood	
Allergy Test	Total Immunoglobulin E	Plain tube	2 ml whole blood	Serum	-
	001 - Seafood Allergy Panel (pacific squid, COD fish, crab, shrimp, clam, anchovy)	Plain tube	2 x 3.5 ml whole blood	Serum	-
	002 - Seafood and Food Allergy Panel (pacific squid, COD fish, crab, shrimp, clam, anchovy, egg white, egg yolk, milk, peanut, chicken, soya bean, wheat, beef)				
	003 - Food Allergy Panel (white, egg yolk, milk, peanut, chicken, soya bean, wheat, beef)				
	004 - Animal Allergy Panel (cat, pigeon drop, cockroach)				
	005 - Mites, Grass Pollen and Fungal Allergy Panel				

PROFILE	TEST	CONTAINER	VOLUME	SPECIMEN	REMARKS
	(Dermatophagoides pteronyssinus, Dermatophagoides farinae, Blomia tropicalis, mix grass pollen, mix fungal microorganism)				
	006 - 26 All Allergens (pacific squid, COD fish, crab, shrimp, clam, anchovy, egg white, egg yolk, milk, peanut, chicken, soya bean, wheat, beef, cat, pigeon drop, cockroach, Dermatophagoides pteronyssinus, Dermatophagoides farinae, Blomia tropicalis, mix grass pollen, mix fungal microorganism, latex)				
	007 - Occupational Allergy Panel (latex)				
	Allergen d1 (Dermatophagoides pteronyssinus)	Plain tube	2 x 3.5 ml whole blood	Serum	-
	Allergen d2 (Dermatophagoides farinae)				
	Allergen d201 (Blomia tropicalis)				
	Allergen E1 (Cat Dander)				
	Allergen E215 (Pigeon Feathers)				
	Allergen F1 (Egg White)				
	Allergen F2 (Milk)				
	Allergen F3 (COD Fish)				
	Allergen F4 (Wheat)				

PROFILE	TEST	CONTAINER	VOLUME	SPECIMEN	REMARKS
	Allergen F9 (Rice)				
	Allergen F13 (Peanut)				
	Allergen F14 (Soya Bean)				
	Allergen F23 (Crab)				
	Allergen F24 (Shrimp)				
	Allergen F27 (Beef)				
	Allergen F41 (Salmon)				
	Allergen F58 (Pacific Squid)				
	Allergen F75 (Egg Yolk)				
	Allergen F83 (Chicken Meat)				
	Allergen F93 (Cacao)				
	Allergen F207 (Clam)				
	Allergen F313 (Anchovy)				
	Allergen GX2 (Grass Pollen)				
	Allergen I6 (Cockroach)				
	Allergen K82 (Latex)				
	Allergen MX2 (Mix Fungi)				
Allergy Screening	Phadiatop	Plain tube	2 ml whole blood	Serum	-
	Phadiatop Infant (<5 Years old)				
***Specific Antibody Response**	Pneumovax vaccination	Plain tube	2 ml whole blood	Serum	-Take the pre vaccination sample -Vaccinate the patient -Take the post vaccination sample 4 - 6 weeks after vaccination
	Tetanus toxoid vaccination				
***Lymphocyte Proliferation Test (LPT)**		Lithium Heparin without gel	12 ml Whole blood	Whole blood	Require healthy control sample 12 ml whole blood (any age matched or adult sample accepted). Strictly sent to our lab within 8 hours after blood withdrawal. Failure to comply may cause the sample to be rejected.

PROFILE	TEST	CONTAINER	VOLUME	SPECIMEN	REMARKS
***Extended Immuno-phenotyping**	Switched memory B cells	EDTA tube	2 ml	Whole blood	Require healthy control sample 2 ml whole blood (any age matched or adult sample accepted). Strictly sent to our lab within 8 hours after blood withdrawal. Failure to comply may cause the sample to be rejected.
	CD40 Ligand Assay	Lithium Heparin without gel	2 ml	Whole blood	Require healthy control sample2 ml whole blood (any age matched or adult sample accepted). Strictly sent to our lab within 8 hours after blood withdrawal. Failure to comply may cause the sample to be rejected.
	Bruton's tyrosine kinase (Btk) function Assay	EDTA tube	2 ml	Whole blood	Require healthy control sample2 ml whole blood (any age matched or adult sample accepted). Strictly sent to our lab within 8 hours after blood withdrawal. Failure to comply may cause the sample to be rejected.
Serology test	Anti-streptolysin 'O' test (ASOT) Rheumatoid Factor (RF)	Plain Tube	3 ml	Serum	-

* For Specific Antibody Response and LPT, the tests should be requested via appointment. Appointment can be done by contacting **04-562 2331**.

3.0 VERBAL REQUEST

Immunology Unit does not accept any verbal request due to optimum blood usage as requested per each test.

4.0 REPORTING RESULTS

Turn Around Time (TAT)

PROFILE	TAT
Immunoglobulin Level	Within 18 working days from the date sample is received
Standard Immunophenotyping Enumeration	Within 14 working days from the date sample is received
Phagocyte Function Test	Within 14 working days from the date sample is received
Allergy Test	Within 15 working days from the date sample is received
Specific Antibody Response	Within 4 months from the date sample is received
Lymphocyte Proliferation Test	Within 14 working days from the date sample is received
Extended Immunophenotyping	Within 14 working days from the date sample is received
Complement Assay	Within 18 working days from the date sample is received
Anti-streptolysin 'O' test (ASOT)	Within 14 working day from the date sample is received
Rheumatoid Factor (RF)	Within 14 working day from the date sample is received

5.0 REFERENCE RANGE

5.1 Immunoglobulin Level

AGE	IgG (g/l)	IgA (g/l)	IgM (g/l)
0 - 2 weeks	6.5 - 12.6	< 0.16	0.03 - 0.24
0.5 - 4 months	2.6 - 7.8	0.06 - 0.57	0.10 - 0.55
4 - 6 months	2.2 - 11.3	0.08 - 0.90	0.07 - 0.65
6 - 24 months	2.6 - 15.2	0.16 - 1.1	0.10 - 1.2
2 - 6 years	4.3 - 13.4	0.19 - 2.2	0.21 - 1.8
6 - 16 years	5.2 - 15.6	0.54 - 3.6	0.13 - 2.4
Adults	7.0 - 16.0	0.70 - 4.0	0.40 - 2.3

Reference:

i. Adapted from: E. de Vries, Clin Exp Immuno. 2012; 167(1). Vademecum diagnostisch onderzoek Sanquin. 2008, pp90

AGE GROUPS	IgG1 (g/l)	IgG2 (g/l)	IgG3 (g/l)	IgG4 (g/l)
≤ 1 year	1.51 - 7.92	0.26 - 1.36	0.093 - 0.920	0.004 - 0.464
> 1 year to ≤ 3 years	2.65 - 9.38	0.28 - 2.16	0.087 - 0.864	0.009 - 0.742
> 3 years to ≤ 6 years	3.62 - 12.28	0.57 - 2.90	0.129 - 0.789	0.013 - 1.446
> 6 years to ≤ 12 years	3.77 - 11.31	0.68 - 3.88	0.158 - 0.890	0.012 - 1.699
> 12 years to ≤ 18 years	3.62 - 10.27	0.81 - 4.72	0.138 - 1.058	0.049 - 1.985
> 18 years	4.05 - 10.11	1.69 - 7.86	0.11 - 0.85	0.03 - 2.01

The following reference ranges (2.5th to 97.5th percentile in g/l) were established by testing samples from 405 apparently healthy children from North America and Central Europe as well as 279 apparently healthy adults from Central Europe with N Latex IgG3 and

References:

i. Product information edition –SIEMENS

ii. Stiehm E.R et al; 2004, 5th edition, Elsevier Saunders and Immunology Today June 1992

5.2 Standard Immunophenotyping Enumeration

	%	ABSOLUTE (x 10^6/L)
Total T Cells	67	2161
Total B Cells	25	803
Th Cells (CD4)	33	1060
Th Cells (CD8)	29	921
NK Cells	8	248

Normal range [percentage % and absolute count (X 10^6/L)]

SUB-POPULATION	AGE GROUPS							
	1 day - 11 months*		1 - 6 years*		7-17 years*		> 18 years**	
	%	Absolute	%	Absolute	%	Absolute	%	Absolute
Total T Cells	58 - 67	1700 - 3600	62 - 69	1800 - 3000	66 - 76	1400 - 2000	53 - 80	988 - 3912
Total B Cells	19 - 31	500 - 1500	21 - 28	700 - 1300	12 - 22	300 - 500	5 - 22	130 - 716
Th Cells (CD4)	38 - 50	1700 - 2800	30 - 40	1000 - 1800	33 - 41	700 - 1000	19 - 46	431 - 1976
Ts Cells (CD8)	18 - 25	800 - 1200	25 - 32	800 - 1500	27 - 35	600 - 900	17 - 49	385 - 1805
NK Cells	8 - 17	300 - 700	8 - 15	200 - 600	9 - 16	200 - 600	8 - 37	227 - 1354

References:

i. *Immunology Today June 1992

ii. **Laboratory Normal Range for adults over 18

5.3 Total Immunoglobulin (Ig) E

AGE GROUPS	Ig E (kU/L)
6 weeks	0.6 - 2.3
3 months	1.0 - 4.1
6 months	1.8 - 7.3
9 months	2.6 - 10
12 months	3.2 - 13
2 years	5.7 - 23
3 years	8.0 - 32
4 years	10 - 40
5 years	12 - 48
6 years	14 - 56
7 years	16 - 63
8 years	18 - 71
9 years	20 - 78
> 10 years	22 - 85

References:

i. Bhala RB, Rappaport I, De Filippi I, Schwartz MK. Serum IgE levels in a northest United States causian population. In: Heusghem C, Albert A, Benson E S, eds. Advanced interpretation of clinical laboratory data. New York: Marcel Dekker Inc, 1982:295-305.

ii. Kjellman NI, Johansson SGO, Roth A. Clin Allergy 1976;6:51-9.

iii. Bjorksten B, Weeke B.Allergy 1985;40 (Suppl 4).

5.4 Specific Immunoglobulin (Ig) E

ImmunoCAP Allergens:

RESULT (kUA/l)	SENSATION LEVEL	CLINICAL RELATION
0 - 0.09	Undetectable	Unlikely
0.10 - 0.50	Very Low	Uncommon
0.51 - 2.00	Low	Low
2.01 - 15.00	Moderate	Common
15.01 - 50.00	High	High
> 50.01	Very High	Very High

Allergens Mixes:

RESULT (kUA/l)	INTERPRETATION
Below 0.35	Undetectable levels or very low levels, of allergen specific IgE antibodies
≥ 0.35	Specific IgE antibodies to one or more of the allergens coupled to immunoCAP Allergen mixes

5.5 Phadiatop and Phadiatop Infant

RESULT (kUA/l)	INTERPRETATION	
Below 0.35	NEGATIVE (non-atopic)	Undetectable levels or very low levels, of allergen specific IgE antibodies
≥ 0.35	POSITIVE (atopic)	Specific IgE antibodies to one or more of the allergens coupled to immunoCAP Allergen mixes

Reference:

i. Normal range provided by ImmunoCAP

CHAPTER 9
MICROBIOLOGY UNIT

CHAPTER 7

MICROBIOLOGY UNIT

1.0 INTRODUCTION

The objective of this user manual is to provide basic information to the users of the Microbiology Unit services on appropriate selection, collection and proper management of microbiological specimens so as to obtain clinically relevant laboratory results in the diagnoses of infectious diseases.

Note: Only Blood Culture & Sensitivity and Urine Culture & Sensitivity tests are MS ISO 15189 accredited.

2.0 COMPLETING MICROBIOLOGY REQUEST FORM

Request for a specific test can be done by using Microbiology request form or/and *Care2x System*. Microbiology request form contains two copies, where each copy is different at the back page. Each specimen should be accompanied by fully completed Microbiology request form including the following information:

a. Patient's Name, Age and Sex
b. Patient's registration number (PID)
c. Name of Ward or Clinic
d. Relevant clinical history and provisional diagnosis
e. Doctor's name (stamped) and signature
f. Type of specimen
g. Date and time of specimen collection
h. Test requested (to state clearly the specific test required)
i. Antibiotics/procedure, if any, that the patient is receiving

*This information is important to accurately interpret results and relate the results to patient care

3.0 SPECIMEN LABELING

Each specimen shall have a label firmly attached to the specimen container and bearing the following information:

a. Patient's name
b. Patient's registration number (PID)
c. Name of Ward or Clinic
d. Type of specimen (including specific anatomic site)
e. Date and time of specimen collection

4.0 STAT SPECIMEN (URGENT PRIORITY SPECIMEN)

4.1 STAT specimen is defined as specimen from patient with potentially life threatening illnesses requiring immediate attention.

4.2 All STAT specimens must inform the lab immediately before sending sample. Any other specimen request for urgent processing that not listed below need prior consultation with a Clinical Microbiologist.

4.3 STAT specimens are processed immediately upon arrival at the laboratory. STAT specimens include:

i. Blood for culture
ii. Cerebrospinal fluid (CSF) – from lumbar puncture procedure or direct sampling in operating theatre
iii. Eye specimens – in cases of endophthalmitis
iv. Joint fluids – if septic arthritis is diagnosed
v. Pericardial fluid
vi. Serum specimen from victim of needle-prick injury for anti-HBs
vii. Serum specimen from source of needle-prick injury for anti-HIV
viii. Serum specimen from source of needle-prick injury for HbsAg

5.0 LIST OF TESTS, SPECIMEN COLLECTION, TRANSPORTATION AND TURNAROUND TIME (TAT)

(TAT is defined as the time period from specimen receipt at Microbiology Unit until results are available)

NO.	NAME OF TEST	TYPE OF SPECIMEN	SPECIMEN COLLECTION	TAT
BACTERIOLOGY TESTS				
1.	Culture & Sensitivity	Blood	Bactec® Plus + Aerobic/F: 8 - 10 ml (Adult) Bactec® Plus + Anaerobic/F: 8 - 10 ml (Adult) Bactec® Peds Plus/F: 1 - 3 ml (Paed / Neonates)	Negative culture result: 6 days from the date sample is received Positive culture result: 3 - 8 days from the date sample is received (depends on time of growth detection by the Bactec® system)
	Fungal Culture (on request)		Bactec® Myco/F-Lytic: 1 - 5 ml	Negative culture result: 4 weeks from the date sample is received Positive culture result: 4 - 6 weeks from the date sample is received (depends on time of growth detection by the Bactec® system)
2.	Microscopy	Urine	10 ml urine in a sterile container	2 hours from the date sample is received
	Culture & Sensitivity			3 - 6 days from the date sample is received
3.	Culture & Sensitivity	Sputum	Early morning sputum in sterile container	3 - 6 days from the date sample is received
4.	Ova and cyst (wet mount)	Stool	Fresh stool in sterile container	1 day from the date sample is received
	Culture & Sensitivity		Sterile container	6 days from the date sample is received
5.	Culture & Sensitivity	Cerebrospinal fluid (CSF)	0.5 to 3 ml in a sterile container	3 - 6 days from the date sample is received
6.	Culture & Sensitivity	Pus (wound) swabs	Swab in transport media	3 - 6 days from the date sample is received
7.	Culture & Sensitivity	Aspirates from abscesses or deep wound	Sterile container	3 - 7 days from the date sample is received

NO.	NAME OF TEST	TYPE OF SPECIMEN	SPECIMEN COLLECTION	TAT
8.	Culture & Sensitivity	Tissue & Biopsy	Sterile container	3 - 7 days from the date sample is received
9.	Culture & Sensitivity	Conjunctival (eye) Swab	Swab in transport media	2 - 5 days from the date sample is received
10.	Culture & Sensitivity	Throat swab	Swab in transport media	3 days from the date sample is received
11.	Culture & Sensitivity	Ear Swab	Swab in transport media	2 - 5 days from the date sample is received
12.	Culture & Sensitivity	Intravenous catheter tips	Sterile container	3 days from the date sample is received
13.	Culture & Sensitivity	High vaginal swab	Swab in transport media	2 - 5 days from the date sample is received
14.	Culture & Sensitivity	Endocervical swab	Swab in transport media	2 - 5 days from the date sample is received
15.	Culture & Sensitivity	Urethral swab (male)	Swab in transport media	2 - 5 days from the date sample is received
16.	Culture & Sensitivity	Sterile body fluids (pleural, pericardial, synovial, intraocular fluid, peritoneal)	Sterile container	2 - 5 days from the date sample is received
17.	Culture & Sensitivity	Nasal swab for MRSA Screening	Swab in transport media	2 - 5 days from the date sample is received
18.	Culture & Sensitivity	Endotracheal Tube (ETT) secretions (tracheal aspirate)	Sterile container	2 - 5 days from the date sample is received
19.	Culture & Sensitivity	Bronchiol Alveolar Larvage (BAL) / Bronchial Washing / Bronchial Brushing	Sterile container	2 - 5 days from the date sample is received
MYCOBACTERIOLOGY TEST				
1.	AFB detection	Sputum	5 - 10 ml early morning sputum collected in sterile container	1 day from the date sample is received
2.	AFB detection	Sterile body fluid (other than CSF)	5 - 10 ml in sterile container	1 day from the date sample is received

NO.	NAME OF TEST	TYPE OF SPECIMEN	SPECIMEN COLLECTION	TAT
SEROLOGY TESTS				
1.	Hepatitis B surface antigen (HBsAg)	Blood / Serum	3 ml in plain tube	7 days from the date sample is received
2.	Hepatitis B surface antibody (anti-HBs)	Blood / Serum	3 ml in plain tube	7 days from the date sample is received
3.	Human immunodeficiency virus antigen/ antibody (HIV Ag/Ab)	Blood / Serum	3 ml in plain tube	7 days from the date sample is received
4.	Hepatitis C virus antibody (anti-HCV)	Blood / Serum	3 ml in plain tube	7 days from the date sample is received
5.	RPR anti-nontreponemal screening for syphilis	Blood / Serum	3 ml in plain tube	3 days from the date sample is received
6.	Treponema pallidum particle agglutination (TPPA)	Blood / Serum	3 ml in plain tube	3 days from the date sample is received
7.	Dengue NS1/IgM/IgG	Blood / Serum	3 ml in plain tube	2 hours from the time sample is received
8.	Leptospira IgM/IgG	Blood / Serum	3 ml in plain tube	2 days from the date sample is received
9.	Rotavirus	Stool	Sterile container	2 days from the date sample is received
10.	Blood film for malarial parasites (BFMP)	Blood	2 ml in EDTA tube	6 hours from the time sample is received
11.	Needle Prick Injury Screening (Urgent)	Blood	3 ml in plain tube	1 day from the date sample is received
OUTSOURCE TESTS				
1.	Human immunodeficiency virus antigen / antibody (HIV Ag/Ab) Confirmation	Blood / Serum	3 ml in plain tube	6 weeks from the date sample is received
2.	Hepatitis B surface antigen (HBsAg) Confirmation	Blood / Serum	3 ml in plain tube	6 weeks from the date sample is received
3.	Hepatitis C virus antibody (anti-HCV) Confirmation	Blood / Serum	3 ml in plain tube	6 weeks from the date sample is received
4.	Mycoplasma Serology	Blood / Serum	3 ml in plain tube	1 month from the date sample is received
5.	Toxoplasma IgM/IgG	Blood / Serum	3 ml in plain tube	1 month from the date sample is received
6.	Cytomegalovirus IgM/IgG (IgM/IgG CMV)	Blood / Serum	3 ml in plain tube	1 month from the date sample is received
7.	Herpes Simplex Virus Types 1&2 IgG	Blood / Serum	3 ml in plain tube	1 month from the date sample is received
8.	Epstein-Barr virus (EBV)	Blood / Serum	3 ml in plain tube	1 month from the date sample is received
9.	Swab specimens for viral culture (Respiratory Syncytial Virus (RSV)	Swab	Viral Transport Medium (VTM)	1 month from the date sample is received

6.0 GUIDELINES FOR PROPER SELECTION, COLLECTION, TRANSPORTATION AND MANAGEMENT OF SPECIMENS

6.1 General Guidelines for Proper Specimen Collection

a) Specimens should be collected before antibiotics given, if possible. If antibiotics are given, indicates in the request form.
b) Specimens should be collected from correct anatomic sites using proper sterile techniques to avoid contamination from indigenous flora.
c) Specimens collected should be adequate of volume and be placed in appropriate container. Inadequate amounts of specimen may yield to false-negative results.
d) Identify the specimen source and/or specific site correctly so that proper culture media will be selected during processing in the laboratory.
e) Check expiration date of the container or transport media before taking specimen. Transport medium prevents specimen drying, helps maintain pathogen viability between collection and inoculation and retards the growth of microbial contamination.

6.2 General Guidelines for Proper Specimen Transportation

a) Each specimen should be accompanied by fully completed request form. Place the container and the form into separate compartments of a plastic specimen transport bag.
b) Properly collected specimens should be sent to laboratory promptly within office hours without delay (preferably within 2 hours). This is to ensure the survival and isolation of pathogens and to provide a more accurate diagnosis of the infectious-disease process.
c) Specimens for viral culture should be delivered on wet ice or a cold pack.
d) If delayed transportation is anticipated, store the specimen at appropriate temperature to maintain its viability.

6.3 Guidelines for Bacteriology Tests

3.3.1 Blood Culture & Sensitivity

a) Choose the appropriate culture media to be inoculated and know the optimal blood volume for inoculation.
b) Check blood bottle medium for gross contamination before use. Bottle with gross turbidity should be returned to the laboratory.
c) The operator should wear sterile gloves after proper hand washing.
d) Disinfection the skin area of venepuncture site:
    - Clean the site with 70% alcohol or 2% chlorhexidine or iodine based preparation. If povidone iodine is used, wait for 2 minutes to dry, or if tincture of iodine is used, wait for 30 seconds.
    - Swab the disinfectants on the skin beginning in the centre of the area and moving outward in concentric circles.
e) Do not palpate vein at this point.
f) Remove the outer seal and cap from each blood culture bottle. Disinfect culture bottle with apply 70% alcohol to rubber stopper and wait 1 minute.

g) Inoculate the blood specimen into the blood culture medium by inserting the needle through the rubber stopper.
h) Inoculate a second blood culture bottle without changing the needle. Changing the needle before inoculating the second bottle creates a risk of contamination. Mix the contents of the culture bottle by gentle swirling.
i) Factors directly influencing blood culture results:
    i. Volume of blood collected
    ii. Method of skin disinfection
j) General recommendation for blood Culture & Sensitivity collection are as follow:
    i. Acute sepsis: 2 - 3 sets from separate sites all within 10 minute
    ii. Acute endocarditis: 3 sets from 3 sites over 1 - 2 hours
    iii. Subacute endocarditis: 3 sets from 3 sites taken > 15 minutes apart
    iv. Fever of unknown origin: 2 - 3 sets from separate sites > 1 hour apart
k) Only one blood culture bottle should be inoculated at one time from one venepuncture site.
l) Send specimen to lab immediately. If delay is inevitable keep bottle culture at room temperature for not more than 24 hours.

6.3.2 Urine Culture & Sensitivity

a) Date and time of collection of urine specimens are very critical information and must be stated clearly in the request form and specimen container. If this information is not provided, the urine specimen may be rejected.
b) The specimen should reach the laboratory within 1 hour after collection. In case of delay, store urine at 4°C (refrigerated) for not more than 24 hours.
c) The request form should indicate patient is symptomatic or not and method of urine collection. This information is critical to quantitative culture interpretation, especially of low-count urine specimens.

METHOD OF URINE	SELECTION, COLLECTION AND TRANSPORTATION
Catheters Urine	1. For indwelling urinary catheter, urine must be taken *only* from the sampling port of the catheter after *meticulous* preparation of the port. 2. Never collect urine from the bag. 3. Urinary catheter tips are not acceptable specimens for culture. 4. Urine obtained by a straight catheter from women is usually acceptable for culture if attention to site preparation has been accomplished and aseptic collection is successful. 5. Catheter should clean with an alcohol pad. 6. Use a sterile needle and syringe to puncture and aspirate the urine from the proximal lumen or sampling port of the catheter. 7. Transfer the urine into a sterile specimen container.

METHOD OF URINE	SELECTION, COLLECTION AND TRANSPORTATION
Midstream (Clean-Catch) Urine	1. The specimen should be collected after the patient has been given specific instructions. Do not assume that a patient knows how to do what is expected. 2. Collect 2 – 10 ml midstream urine in a sterile container. Early morning urine is preferable to enable the organism to multiply in the bladder before collection. If not possible a period of 2 hours must elapse after the last urination. 3. For Male patient, cleanse the glans (the head of the penis) with plain soap and water. 4. For Female patient, should similarly cleanse the genital area. 5. Discard the first portion of voided urine and collect the midstream urine directly into a sterile bottle.
Suprapubic Aspirate urine	1. The method is particularly useful in paediatric patients, patients with a spinal cord injury, and patients for whom a definitive culture has not been obtained. 2. This method is required for diagnosing anaerobic urinary tract infections. 3. Decontaminate the skin (with antiseptics as in preparation for surgery) from the umbilicus to the urethra. Anaesthetize the skin at the insertion site. 4. Introduce the needle into the full bladder at the midline between the symphysis pubis and the umbilicus, 1-2 cm above the symphysis pubis. 5. Aspirate about 20 ml of urine from the bladder. 6. Transfer the urine aseptically into a sterile screw-capped container.
Bladder Washout	1. Bladder washout can be used to assist in determining whether a bladder infection or a kidney infection exists. If the kidney is involved, the post bladder rinse specimen should contain large numbers of organisms, whereas in bladder infections, this specimen shows no growth. 2. For collect this type of urine: a) Insert an indwelling catheter into the bladder, then save the last portion of the urine flow for culture. Refrigerate it immediately. b) Introduce a specified volume of a solution of neomycin (0.1 to 0.2%). Allow the solution to remain in the bladder for 30 minutes. c) Wash the bladder with 2 liters of sterile irrigating fluid and drain the bladder. d) Collect three samples at 10-minute intervals. Label the initial and subsequent timed collections. (Later specimens should represent urine from the kidney without contamination from organisms previously located in the bladder).
Nephrostomy Tube Urine	1. To collect this type of urine: a) Clean the catheter with an alcohol pad. b) Use a sterile needle and syringe to puncture and aspirate the urine from the catheter. c) Transfer the urine into a sterile specimen container.

6.3.3 Sputum Culture & Sensitivity

a) Sputum may not be the best specimen for determining the etiologic agent of bacterial pneumonia (Broncho-alveolar lavage or blood specimens may be more accurate).
b) Sputum specimens are rejected if microscopic examination reveals epithelial cells, which indicated contamination from the oropharyngeal flora. In such cases, the specimen should be recollected. Careful attention to the instructions given the patient greatly reduces the number of inappropriate specimens.
c) Send specimen immediately to laboratory. If delay unavoidable store it in refrigerator for not more than 24 hours.
d) Pooled specimens are not recommended for culture.

TYPE OF SPUTUM	SELECTION, COLLECTION AND TRANSPORTATION
Sputum (Expectorate)	To collect this type of sputum: a. Have patient rinse and gargle water to remove superficial flora b. Instruct patient to cough deeply to produce lower respiratory specimen and collect **early morning sputum in a sterile container**. Instruct the patient in the difference between sputum and spit.
Sputum (Induced)	To collect this type of sputum: a. Have the patient rinse the mouth with water after brushing the gums and tongue. b. With the aid of nebulizer, have the patient inhale ~25 ml of 3 - 10% sterile saline. c. Collect the induced sputum in a sterile container.

6.3.4 Stool Culture & Sensitivity / Rotavirus

a) Instruct patients to excrete directly into the cup or collection device.
b) Alternatively, using a swab collect about one third of the container by dipping and rotating in the faeces taking care to include materials containing pus, mucus or blood if present.
c) Places the stool into a sterile container, screw the cap tightly and send it immediately to the laboratory.
d) Do not allow urine to contaminate the specimen and never take a specimen from the water in a toilet.
e) Routine cultures are done for isolation of *Salmonella spp., Shigella spp., Campylobacter spp., Aeromonas hydrophila*, EHEC, EPEC. If other agents are suspected, request specifically to the laboratory.
f) Stool specimens for bacterial culture and sensitivity testing must be transported to the laboratory within 1 - 2 hours after collection for immediate processing. Stool specimens that are received more than 24 hours after collection would be rejected. If delay is unavoidable, store at 4°C not more than 24 hours in respective ward.

6.3.5 Cerebrospinal Fluid (CSF) Culture & Sensitivity

a) Disinfect the skin over the lumber puncture site.
b) Using strictly aseptic techniques perform a lumbar puncture and collect about 0.5 - 3 ml of CSF directly into sterile container.
c) Send the specimen immediately to the laboratory.
d) Do not store in a refrigerator as organisms causing meningitis are usually very sensitive to cold.

6.3.6 Wound Swabs Culture & Sensitivity

a) When the lesion is larger or when there are several lesions, multiple specimens from different sites must be collected.
b) Surface lesion samples are unsuitable for anaerobic studies.
c) For collection & transportation:
    i. Remove as much of the superficial flora as possible by decontaminating the lesion using sterile swab.
    ii. Place the swab in appropriate transport medium.
    iii. Label the specimen and state the names of the *specific anatomic locations* that are the sources of specimens.
    iv. Send the specimen immediately to the laboratory.

6.6.7 Aspirates from Abscesses or Deep Wound Culture & Sensitivity

a) Disinfect the surface with 70% alcohol and then with povidone-iodine solution.
b) Aspirate abscess fluid from the deepest portion of the lesion with a sterile syringe and needle and transfer into sterile container (Avoid contamination by the wound surface).
c) Do not allow air to enter the transport container, some anaerobic organisms are killed by oxygen.
d) Send the specimen immediately to the laboratory. Do not store in a refrigerator.

6.6.8 Tissue & Biopsy Culture & Sensitivity

a) Collect tissue aseptically and transfer into a sterile container.
b) Do not place the tissue or biopsy in a fixative solution (formalin). If necessary, moistened the tissue with 0.85% normal saline to prevent drying.
c) Send immediately to the laboratory.

6.6.9 Eye Swab Culture & Sensitivity

a) Specify and state the specific sources of specimens (e.g., conjunctival eye, aqueous or vitreous sample, etc.) and eye side (e.g., left or right eye).
b) In bilateral conjunctivitis, culture of a specimen from only one eye is necessary.

c) For conjunctival swab, purulent material should collect on a sterile cotton swab from the lower conjunctiva sac without contamination with eyelid. The swab should be placed in Amies transport media and transport immediately to the laboratory.
d) Specimen should be collected before application of antibiotics, irrigating solution or other medicines.

6.6.10 Throat Swab Culture & Sensitivity

a) The sample is taken at the back of throat and tonsillar areas of inflammation and exudates. Success with culture depends on firmly and completely sampling the involved areas.
b) Insert the sterile cotton swab carefully but firmly into mouth and rub the swab over several areas of exudates or over the tonsils and posterior pharynx.
c) Do not touch the cheeks, teeth, or gums with the swab so as to avoid.
d) Place the swab in Amies transport medium and send to the laboratory immediately.

6.6.11 Ear Swab Culture & Sensitivity

a) The external ear canal is cleaned with a sterile swab moistened with sterile saline.
b) Pass a swab gently into the external canal and collect the pus or exudates from the middle or inner ear by using a sterile swab.
c) Place the specimen in appropriate transport medium and send to the laboratory immediately.
a) Label the specimen and state in request form the names of the specific anatomic locations that are the sources of specimens.

6.6.12 Intravenous Catheter Tip Culture & Sensitivity

a) Relevant clinical history that should accompany specimen includes:
    i. Any suspected or proven infection
    ii. Duration of cannulation
    iii. Anatomical location and designated purpose for the cannulation
    iv. Any antimicrobial therapy received by patient while the catheter was in place
    v. Any sign of inflammation at the catheter site (e.g., presence of purulent and lymphangitis, erythema, tenderness, increased warmth, palpable thrombosed vein)
b) It is important that the skin surface is cleaned up from any antimicrobial ointment or blood clot before removal of the catheter.
c) Aseptically cut 1 to 4 inches of the distal tip of the catheter using a pair of sterile scissors. Allow the cut segment to drop directly into a sterile container.
d) Send the specimen immediately to the laboratory to avoid drying of the catheter tip.

6.6.13 High Vaginal Swab (HVS) Culture & Sensitivity

a) Use a speculum without lubricant.
b) Obtain secretion from the mucosa high in the vaginal canal with a sterile swab.
c) Place the specimen in appropriate transport medium and send to the laboratory immediately.

6.6.14 Endocervical Swab Culture & Sensitivity

a) Routine tests are done for detection of *Neisseria gonorrhoea, Candida spp.* and *Trichomonas vaginalis.*
b) Moisten the speculum with sterile saline. Do not use lubricants as it can be toxic to *Neisseriae*.
c) Remove from the cervical of any mucous or vaginal material before collecting endocervical discharge with a swab. Alternatively, insert the swab into the cervical os, allow it to remain in place for a few seconds, and remove it.
d) Add a small quantity of sterile saline to the swab specimen to keep it moist before sending it promptly to the laboratory. Do not refrigerate the specimen.

6.6.15 Urethral Swab (Male) Culture & Sensitivity

a) This specimen is useful for definitive diagnosis of gonorrhoea in male patients.
b) Remove the external skin flora of the urethral meatus as in preparation for obtaining a urine specimen.
c) Collect the express exudates from the urethra and collect by using the sterile swab and place the swab in appropriate transport medium. Material from a site about 2 cm inside the urethra or expressed pus is the specimen of choice.
d) If exudates is unavailable, insert a urethrogenital swab about 2 - 4 cm into the urethral lumen, gently rotates it. Leave the swab in place for at least 2 seconds to facilitate absorption before removing it.
e) Send the specimen to the laboratory immediately. Do not refrigerate specimen.

6.6.16 Sterile Body Fluids (Pleural, Pericardial, Synovial, Intraocular Fluid, Peritoneal) Culture & Sensitivity

a) These body fluids are diagnostic for infection if cultures become positive because they are sterile.
b) For peritoneal fluid specimen, a volume of 30 - 50 ml is needed.
c) Clean the needle puncture site with 70% alcohol or 2% chlorhexidine followed by 1% iodine in 70% alcohol or povidone iodine.
d) Aseptically perform percutaneous aspiration to obtain pleural, pericardial, peritoneal, or synovial fluids.
e) Transfer fluids aspirated aseptically to a sterile container and send to the lab without delay.

6.6.17 Nasal Swab for MRSA Screening

a) Lesions in the nose require samples from the advancing margin of the lesion.
b) Nasal swab culture is routinely done only for MRSA screening.
c) Carefully insert sterile swab at least 1 cm into the nostril and firmly sample the membrane by rotating the swab and leaving it in place for 10 to 15 seconds. Then, withdraw the swab and insert it in a transport container.
d) Use separate swabs for each nostril.
e) Detection of carriage of methicillin-resistant *S. aureus* (MRSA) can be increased by also sampling another body sites, such as the rectum and axilla.

6.6.18 Endotracheal Tube (ETT) Secretions (Tracheal Aspirate) Culture & Sensitivity

a) Collect the specimen through a tracheostomy or endotracheal tube (ETT).
b) Carefully pass the catheter through the site and into the trachea.
c) Aspirate material from the trachea using a syringe or an intermittent suction device and place the specimen in sterile container.
d) Send the specimen to the laboratory quickly. Do not refrigerate the specimens.

6.6.19 Bronchiol Alveolar Larvage (BAL) / Bronchial Washing / Bronchial Brushing Culture & Sensitivity

a) Broncho Alveolar Lavage (BAL): 50 to 200 ml of sterile saline is infused into the distal bronchoalveolar tree and subsequently suction out about 10 ml fluid containing cells and microorganisms located at the alveolar level from each specimen trap. Place the fluid in sterile tubes, and submit the specimens to the laboratory.
b) Bronchial Washing: Saline is injected through the bronchoscope and subsequently aspirated from the airways.
c) Bronchial Brushing: A brush is advanced through the bronchoscope and used to abrade suspicious lesions to obtain cells. Bronchial brushings are preferable to washings because the latter specimens are more diluted.
d) Place aspirate or washing in sterile container and send to laboratory immediately. Do not refrigerate the specimen

6.4 Guidelines for Mycobacteriology Test:

6.4.1 Sputum for AFB Detection

a) Sputum is preferably collected when the patient first wakes up in the morning.
b) Gargle mouth before sputum collection and ask the patient to spit directly into a sterile container. Ensure that expectorate is sputum and not saliva.
c) Send the specimen immediately to the laboratory. If delay is unavoidable store it in a refrigerator.

6.4.2 Sterile Body Fluid (Other than CSF) For AFB Detection

a) Gastric Content (Gastric Lavage Fluid) - this method is used to examine for *M. tuberculosis* when sputum specimens are unavailable.
b) Early morning fasting specimen is preferred for mycobacterial culture.
c) The gastric contents of 5 - 10 ml are aspirated and placed in a sterile container.
d) Immediately send the specimen to laboratory.
e) Handle all specimens with the safety precautions necessary for working with *M. tuberculosis.*

6.5 Guidelines for Serology Tests:

a) Each specimen should accompany by fully completed Microbiology Request Form.
b) Blood collected in plain container should be allowed to clot by standing undisturbed at room temperature.
c) Do not refrigerate the specimen (haemolysis increases during refrigeration).
d) Clear, non-haemolysed specimens are preferred (false positivity is a problem with haemolysed specimens).
e) Serum is the recommended and best specimen for serological testing.
f) For Hepatitis, HIV and syphilis screening, tests are performed only on Thursday (once per week) for patient. For urgent cases, please inform Microbiology Unit staff before submit the test request.

REQUESTED TEST	VOLUME OF SPECIMEN NEEDED
For single serology test (Adult)	3 ml of blood in one plain tube
For single serology test (Neonates)	Not less than 1.5 ml of blood per bullet.

g) For Needle Prick Injury Cases (NPI):
   i. Please inform lab staff immediately before sending sample.
   ii. Specimens are send in pairs (except unknown source) and note on the form whether it is victim / staff or patient / source.
   iii. State the status of NPI case (Review or New case).
   iv. **New case**: HBsAg, HCVAb, HIV Ag/Ab and RPR test will be done in 1 working day from the date sample is received.
   Repeat / Review case: HBsAg, HCVAb, HIV Ag/Ab and RPR test will be done within 7 days from the date sample is received.

6.6 Guideline for Outsource Samples

a) Those tests which are not offered by the Microbiology Unit will be outsourced to Referral Laboratory (Refer **APPENDIX: LIST OF REFERRAL LABORATORY**).
b) Prior arrangement must be made with Microbiology Unit staff before submission of specimen.
c) Each specimen should accompany by fully completed Microbiology (ADL) Request Form.

d) For combination of many tests (e.g., TORCHES), please send more blood / serum volume in one plain tube (more than 3 ml) instead of many tubes but lesser serum volume.
e) Swab specimens for viral culture should be transported using appropriate transport medium - Viral Transport Medium (VTM) to prevent drying, and send immediately the specimen to the laboratory on wet ice or a cold pack. Do not freeze or hold the specimen at room temperature.
f) Microbiology Unit (ADL) staff will send the outsourced specimens to the Referral Laboratories only on Monday and Tuesday morning.

7.0 SPECIMEN REJECTION

7.1 Besides the rejection criteria stated in Chapter 1, 10.1, Microbiology Unit reserves the right to refuse processing any specimen that has any of the following characteristics:
i. Inappropriate temperature of the specimen during transportation.
ii. Prolonged transportation time / transport media already dried up.
iii. Specimen collected in inappropriate transport medium.
iv. Presence of contamination detected on gross examination of the specimen.
v. Specimen received in formalin or other fixative for culture.
vi. Saliva as a respiratory specimen (instead of sputum).
vii. Two or more specimens collected within a 24 hour period (unless requested by the laboratory).
viii. Anaerobic specimen not sent under conditions which will preserve anaerobic organism.
ix. Unsuitable specimen for culture (for example, swab specimen for anaerobic culture, or Foley catheter tip for culture).
x. Culture for Gonococci is not suitable for specimen HVS (only accept for endocervical swabs, cervical swab and vaginal discharge).

7.2 The clinic / ward / test requester will be notified before a statement of specimen rejection is issued by the laboratory. Processing of compromised specimen is the onus of the test requester and microbiology consultation should be sought.
7.3 Any compromised specimen that cannot be easily re-collected, or that has been harvested as part of an invasive procedure (i.e. aspirates, sterile body fluids and tissues) will be processed only upon obtaining documented agreement from the test requester. The laboratory reports will contain the specification of the specimen's compromised qualities to indicate that the result generated may not be accurate and test results should be interpreted with caution.

8.0 RELEASE OF RESULTS

8.1 Preliminary report

a) Preliminary report is available when result is crucial to patient management.
b) For Blood C&S test:
i. Routine blood culture bottle (Aerobic, Anaerobic, Paeds) are incubated in BACTEC machine for up to 5 days incubation and 28 days for SBE/IE,

Brucella/Cat Scratch Disease and blood fungal, while 42 days for blood Mycobacterium.

ii. If positive blood culture bottle is flagged by machine, Gram Stain of the blood culture is performed and will be verbally informed immediately to the ward / clinic / test requester. Then, Primary Antibiotic Susceptibility Test (AST) will be informed verbally on the next day (if applicable). All telephoned communications is recorded and printed reports will be followed.

c) For CSF C&S test:

i. All CSF microscopic result will be informed via phone immediately to the ward / clinic / test requester. All telephone communications is recorded.

ii. If CSF culture is growth (positive), result will be informed via phone immediately to the ward / clinic / test requester and a printed report will followed.

8.2 Notification Result

a) When a result warrants a notification of a communication disease under the Notification Communicable Disease of Malaysia via Prevention and Control of Infectious Disease Act 1998, results that is ready to dispatch will be informed via phone immediately to Unit Kawalan Jangkitan (UKJ) IPPT and respective ward / clinic / test requester, whether it is new or review case.

b) Critical Notification Values For Clinical Microbiology List :

TYPE OF SPECIMEN / TEST	ORGANISM
Blood & CSF C&S	All Positive final result
All type of specimens for C&S	*Methicillin Resistant Staphylococcu aureus (MRSA)* *Extended Spectrum Beta Lactamases (ESBL)* *Carbapenem Resistant Enterobacteriaceae (CRE)* *Multidrug Resistant Organisms (MRO)* – include *Acinetobacter* *Vancomycin Resistant Enterococci (VRE)* *Salmonella spp./ typhi* *Shigella* spp *Streptococcus pneumonia* *Streptococcus Group A* *Haemophilus influenzae* *Neisseria gonorrhoeae* *Vibrio cholera* *Aeromonas hydrophila* *Burkholderia mallei/ pseudomallei* *Clostridium perfringenes* *Corynebacterium diphtheria* Enterohemorrhagic *Escherichia coli* EHEC) *Mycobacterium tuberculosis*
Serology Test	All Positive testing results

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9.0 APPENDIX: LIST OF REFERRAL LABORATORY

1. Department of Medical Microbiology & Parasitology
   School of Medical Sciences,
   Universiti Sains Malaysia,
   16150 Kubang Kerian,
   Kelantan.
   Tel: 09-7676286
   Fax: 09-7676289

2. Pathology Department
   Ground Floor, Block B,
   Hospital Pulau Pinang.
   Tel: 04-2225145
   Fax: 04-2225155

3. Institute for Medical Research (IMR)
   Jalan Pahang,
   50588 Kuala Lumpur.
   Tel: 03-2698 6033
   Fax: 03-56939335

4. Makmal Kesihatan Awam Kebangsaan (MKAK)
   Lot 1853, Kg Melayu Sungai Buloh,
   47000 Sungai Buloh,
   Selangor.
   Tel: 03-6156 5109
   Fax: 03-6140 2249