# LABORATORY ASSESSMENT OF

# HAEMATOLOGY ONCOLOGY

# IMMUNOPHENOTYPING

## First Edition 2018

Australasian Cytometry Society Laboratory Assessment Document

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## Contents

The Australasian Cytometry Society (ACS) was established in 1979 and incorporated in 1992 with the aim of promoting research, development and applications in, and to disseminate knowledge of flow cytometry.

A function of the ACS is to assist with development and application of clinical flow cytometry applications for hospitals and laboratories in the diagnosis and treatment of disease. This includes the preparation of guidelines, assessments and education programs. Guidelines produced by the ACS are issued as reference material to provide laboratories and accrediting agencies with minimum requirements for testing considered acceptable for good laboratory practice.

Failure to follow these guidelines and assessments may pose a risk to public health and patient safety.

## SCOPE

The ACS *'Laboratory Assessment of Leukaemia Lymphoma Immunophenotyping'* is an ACS document to be read in conjunction with ACS documents '*Guidelines for Clinical Flow Cytometry Laboratory Practice*' and '*Guidelines for Leukaemia Lymphoma Immunophenotyping'*. These documents outline guidelines for good medical pathology practice where the primary consideration is patient welfare, and where the needs and expectations of patients, laboratory staff and referrers (both for pathology requests and inter-Laboratory referrals) are safely and satisfactorily met in a timely manner.

This document is for use in assessing laboratories providing clinical flow cytometry services.

## ABBREVIATIONS

| | |
|---|---|
| ACD | Acid Citrate Dextrose |
| CSF | Cerebrospinal fluid |
| EDTA | Ethylene-diaminetetraacetic acid |
| RPMI | Roswell park Memorial institute medium, a sample preservative |
| Hanks | Hank's balanced Salt Solution, a sample preservative |
| FNAB | Fine Needle Aspirate Biopsy |

## DEFINITIONS

| | |
|---|---|
| CD | Cluster definition number used to identify individual markers eg CD3 for the pan T cell antigen |
| Competent clinical flow cytometrist | means a person who has a minimum of two years clinical flow cytometry experience, and who has been documented to be competent in clinical flow cytometry according to the Laboratory's Quality System |
| count | means to acquire data on a flow cytometer |
| Guideline | means a consensus recommendation for best medical laboratory practice for a procedure, method, staffing resource or facility |
| Guidelines for Clinical Flow Cytometry Laboratory Practice (GCFCLP) | means the overarching document broadly outlining standards for good clinical flow cytometry laboratory practice where the primary consideration is patient welfare, and where the needs and expectations of patients, Laboratory staff and referrers (both for pathology requests and inter-Laboratory referrals) are safely and satisfactorily met in a timely manner. |
| marker | means an antibody directed to an antigen of interest in or on a cell used for diagnostic purposes |

# INTRODUCTION

This ACS document, together with '*Guidelines for Clinical Flow Cytometry Laboratory Practice',* and *'Guidelines for Leukaemia Lymphoma Immunophenotyping'* is intended to be used in clinical flow cytometry laboratories to assist good practice in relation to flow cytometry for use during laboratory assessments.

Guidelines referred to are intended to serve as consensus recommendations for best medical laboratory practice have been developed by ACS members and associates with reference to other guidelines as published in peer reviewed journals.

The Guidelines are not Standards. The Guidelines should be read in conjunction with the current version of the ACS '*Guidelines for Clinical Flow Cytometry Laboratory Practice'.* For clarification Standards are described as:

• A Standard is the minimum requirement for a procedure, method, staffing resource or laboratory facility that is required before a laboratory can attain accreditation. The use of the verb 'must' in standards indicates mandatory requirements for pathology practice.

In each section of this document, points deemed important for practice are identified as either 'Guidelines' or 'Commentaries', as follows:

• A Guideline is a consensus recommendation for best medical laboratory practice for a procedure, method, staffing resource or facility. Guidelines are prefaced with a 'G' (e.g. G2.2**)**. The use of the word 'should' in each Guideline within this document indicates a recommendation for good pathology practice.

• A Commentary may be provided to give clarification to the Guidelines as well as to provide examples and guidance on interpretation. Commentaries are prefaced with a 'C' (e.g. C1.2) and are placed where they add the most value.

Note: ACS documents can be accessed at: www.cytometry.org.au

# LABORATORY ASSESSMENT OF HAEMATOLOGY ONCOLOGY IMMUNOPHENOTYPING

This assessment is intended for use in assessing laboratory processes according to the ACS '*Guidelines for Leukaemia Lymphoma Immunophenotyping'.*

Assessment 'Point' refers directly to the ACS guideline reference number.

Comments for noncompliance can be made in the table in Appendix.

## 1. PREANALYTICAL PHASE

| POINT | DESCRIPTION | COMPLY |
|---|---|---|
| **G1.1** | **Specimen Collection and Storage** | |
| G1.1.1 | EDTA samples are tested < 48 hrs; lithium heparin < 72 hr; ACD <72 hrs (not BM) | |
| G1.1.2 | Peripheral blood, bone marrow aspirates stored at 18-25$^{o}$C | |
| G1.1.3 | Tissue biopsies in isotonic medium (phosphate buffered saline, Hanks, RPMI), stored at 4°C | |
| G1.1.4 | CSF processed < 8 hrs after collection | |
| G1.1.5 | Fluids are tested < 24 hrs or < 72 hrs if stored at 4°C. | |
| G1.1.6 | Sample type, site, time of collection is provided on sample tube/container with patient identifiers. This information is included in the final report. | |
| G1.1.7 | Total white cell count, differential on all peripheral blood samples is performed at the laboratory initiating the request | |
| G1.2 | Specimen Transport | |
| G1.2 | Samples are delivered to the laboratory as soon as possible to minimise loss of cell viability. | |
| C1.2.(i) | Tissue samples are transported in tissue culture media, saline or similar and are analysed within 24 hrs | |
| C1.2.(iii) | Delay in sample testing is indicated in the final report. | |
| G1.3 | Test Requests | |
| G1.3 | Requests for leukaemia/lymphoma testing include relevant clinical observations and history to assist with appropriate screens undertaken. | |
| C1.3(ii) | Previous abnormal immunophenotypes is noted on the test request or by search of laboratory records. | |

| | | |
|---|---|---|
| G1.4 | Antibody Reagents | |
| G1.4 | Fluorescent marker antibodies used in panels are validated by clinical correlation following IVD standards. | |
| G1.5 | Sample Preparation | |
| G1.5 | Samples are prepared with total white cell count ≤ 20 x 10^9/L | |

**2. ANALYTICAL PHASE**

| POINT | DESCRIPTION | COMPLY |
|---|---|---|
| G2.1 | Sample Analysis | |
| G2.1.1 | A minimum of 5,000 cellular events in the target gate (e.g. lymphocytes, blasts) are acquired where possible. | |
| G2.1.2 | Secondary sample/assay tubes have patient name or part thereof and at least one identifier. Barcode alone is not acceptable. | |
| G2.1.3 | Analysis is performed using CD45, FSC and SSC parameters. | |
| C2.1.4 | Analysis is performed by a competent flow cytometrist with documented competency in clinical flow cytometry analysis of leukaemia and lymphoma. | |
| G2.2 | Performance Measures | |
| G2.2.1 | Within-run positive and negative control results are checked to demonstrate appropriate reactivity. | |
| C2.2.1 | Where absolute numbers (eg cells/uL for blood samples) are reported, a control reagent is periodically tested with specified ranges for the analytes measured, and reasons for deviations determined. | |
| G2.2.2 | Account is made for all populations tested: Lymphosums calculated on blood and bone marrows, CD45 negative populations are examined in leukaemia screens, all lineages tested in tissues and body fluids. | |

**3. POST ANALYTICAL PHASE**

| POINT | DESCRIPTION | COMPLY |
|---|---|---|
| 3.1 | Reports | |
| 3.1.1 | Reports identify any abnormal population gated, its size, markers tested, relevant staining reactivities, and a diagnosis where possible. | |
| G3.1.2 | Reports are completed in a timely manner, no longer than 5 working days for leukaemia and lymphoma immunophenotyping. | |

## REFERENCES

For backgorund on the methods, interpretation and publications refer to the references cited in ACS documents '*Guidelines for Clinical Flow Cytometry Laboratory Practice*' and '*Guidelines for Leukaemia Lymphoma Immunophenotyping'.*

## NPAAC Reference Documents

*Requirements for Medical Pathology Services*, First Edition, 2013
Print ISBN: 978-1-74241-913-8, Online ISBN: 978-1-74241-914-5, Publications approval number: 10207

*Regulatory Requirements for In-house IVDs*, Version 2.0, March 2016, Australian Government, Department of Health, Therapeutic Goods Administration

*Requirements for the Development and Use of In-house In Vitro Diagnostic Devices (IVDs,*) Third Edition 2014
ISBN: 1 74186 158 6; Online ISBN: 1 74186 159 4; Publications Approval Number: 3957

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Further ACS clinical flow cytometry guidelines documents are available on the website: www.cytometry.org.au

Email: clinicalguidelines@cytometry.org.au

**APPENDIX**

The table below may be used for laboratory assessment records and reviews.

**FLOW CYTOMETRY LABORATORY ASSESSMENT**

Laboratory Assessed: Date:

Procedure Assessed: Assessment by:

| POINT | COMMENT | RECOMMENDATION | ACTION |
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| POINT | COMMENT | RECOMMENDATION | ACTION |
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