VIDRL & WHO

Measles IgM Proficiency Panel 00702

Final Report

Prepared by Jennie Leydon, Michaela Riddell

&

Dr Mike Catton

Director

VIDRL

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Abbreviations

ANOVA	Analysis of variance
CDC	Centers for Disease Control and Prevention
EMRO	Eastern Mediterranean Regional Office
N	Negative
OD	Optical density
P	Positive
QA	Quality assurance
S/CO	Sample/cut-off ratio
VIDRL	Victorian Infectious Diseases Reference Laboratory
WHO	World Health Organization

Measles IgM proficiency panel 2002

Panel number: 00702

Introduction

As the world moves towards control of measles, confirmation of clinically diagnosed measles by IgM serology will become increasingly important. Proficiency testing is an important part of measles laboratory programs as both false positive and false negative results can occur with some of the commonly used measles IgM enzyme immunoassays (EIA).

Aim

The aims of this panel are to

1. Assess the proficiency of laboratories with-in the WHO laboratory network
2. Identify problems with any assays routinely used in these laboratories
3. Check the accuracy of data reporting.

Methods

Panel composition

This panel was a renumbered version of the 00801 panel distributed between promulgation of the correct results of the 00801 panel and distribution of the next annual panel (00102)

All samples were undiluted serum samples, comprising

10 Measles IgM positive (sourced from 1999 measles outbreak in Victoria, Australia)

5 Measles IgM negative (VIDRL staff volunteers)

2 Parvovirus IgM positive (Diagnostic sera)

2 Rubella IgM positive (Diagnostic sera)

1 Dengue IgM positive (supplied by the WHO Arbovirus Reference Lab, Qld. Australia)

All samples were negative for HIV, Hepatitis BsAg & Hepatitis C.

Table 1 details the composition of the panel by sample number and validated result of testing.

WHO Panel Results

PANEL 00801

Sample	Measles IgM	Rubella IgM	Status
00702001	Positive	Negative	MEASLES IgM POSITIVE
00702002	Positive	Negative	"
00702003	Positive	Negative	"
00702004	Positive	Negative	"
00702005	Positive	Negative	"
00702006	Negative	Negative	PARVO IgM POSITIVE
00702007	Negative	Positive	RUBELLA IgM POSITIVE
00702008	Negative	Negative	DENGUE IgM POSITIVE
00702009	Negative	Negative	PARVO IgM POSITIVE
00702010	Negative	Negative	MEASLES IgM NEGATIVE
00702011	Negative	Negative	"
00702012	Negative	Negative	"
00702013	Positive	Negative	MEASLES IgM POSITIVE
00702014	Positive	Negative	"
00702015	Negative	Positive*	MEASLES IgM NEGATIVE
00702016	Positive	Negative	MEASLES IgM POSITIVE
00702017	Positive	Negative	"
00702018	Negative	Negative	MEASLES IgM NEGATIVE
00702019	Negative	Positive	RUBELLA IgM POSITIVE
00702020	Positive	Negative	MEASLES IgM POSITIVE

- ***False positive result

- **Table 1:** Panel composition detailing IgM status of panel number.

Validation of panel

The panel was tested at VIDRL using two methods for Measles IgM: and two methods for Rubella IgM.

DadeBehring Enzygnost® anti- measles virus IgM

Chemicon Light Diagnostics Measles IgM Capture Enzyme Immunoassay

Beckman Access Chemiluminescent Rubella IgM
DiaSorin ETI-RUBEK-M reverse PLUS capture assay

Before general distribution the panel was tested for measles IgM by CDC, Atlanta USA and the Central Public Health Laboratory, Colindale UK. The results obtained by the other two laboratories confirmed those obtained by VIDRL.

Distribution of panel

Results were returned from 17 laboratories from WHO EMRO region.

Each laboratory has been assigned a dedicated number. This number is known only to VIDRL and that laboratory.

The countries that returned results were:
Bahrain
Cyprus
Egypt
Iran
Iraq
Jordan
Lebanon
Morocco
Oman
Pakistan
Qatar
Saudi Arabia
Syria
Tunisia
United Arab Emirates
Yemen

Analysis

Analysis was performed by laboratory and panel number. A range of kits was used by participating laboratories. The group using Dade Behring kits was the only one large enough to compare results as there were insufficient numbers for comparison in the other groups. The proportion of correct results, based on the positive/negative interpretation reported by the laboratory, was calculated by laboratory and according to kit used.

The Dade Behring group

The laboratory assigned optical density (OD) values and interpretation (positive/negative) were recorded for each of the panel numbers. The positive/negative cut off was assumed to be 0.2 unless stated otherwise. OD values for all positive samples were graphed for all laboratories and inspected for normality. This inspection was repeated separately for all negative samples .

0Other kit groups

Three laboratories used the Chemicon assay, two laboratories used the Virion assay. The remaining five laboratories used a variety of assay methods. No comparative analysis was attempted since there were insufficient numbers of laboratories using the same kit for meaningful analysis. The OD values obtained by laboratories using commercial EIA kits was plotted but no further comparisons could be made.

Reporting of kit details

Lot numbers

All laboratories supplied lot or reagent details

Expiry dates

1 laboratory did not supply an expiry date.

Results analysed by kit

Kit Details

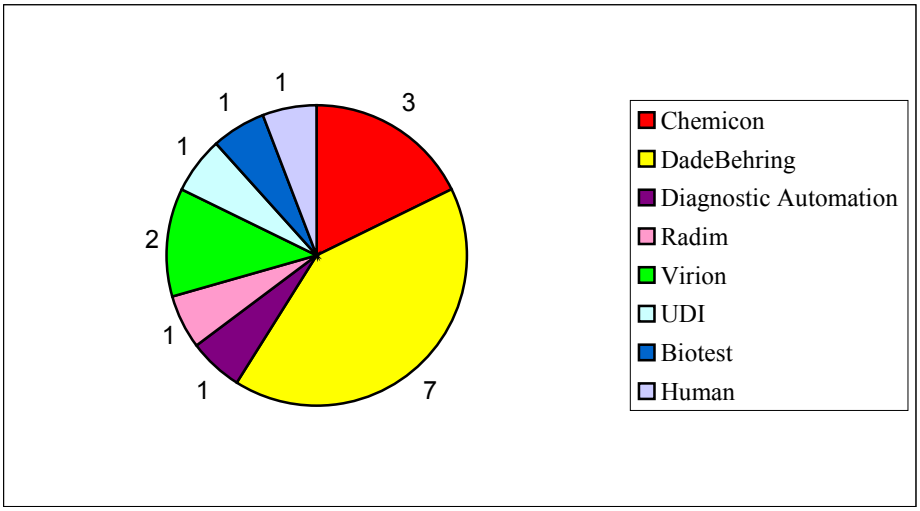

Figure 1: Methods in use for measles IgM testing

Table 2 demonstrates the number of correct results by kit.

Assay	Number of labs using kit	Number of samples correctly identified				
		20	19	18	14	11
Chemicon	3	2			1	
Dade Behring	7	4	2*	1		
Other	7	6				1

- *1 laboratory only tested 19 samples

Table 2: The assay used versus the number of correct results that each laboratory achieved.

Assay	Number of laboratories using assay	Proportion of laboratories with all positives correct	Proportion of laboratories with all negatives correct
Dade Behring	7	71%	71%
Other commercial kits	10	100%	80%

Table 3: The proportion of laboratories correctly identifying all positive and negatives by assay.

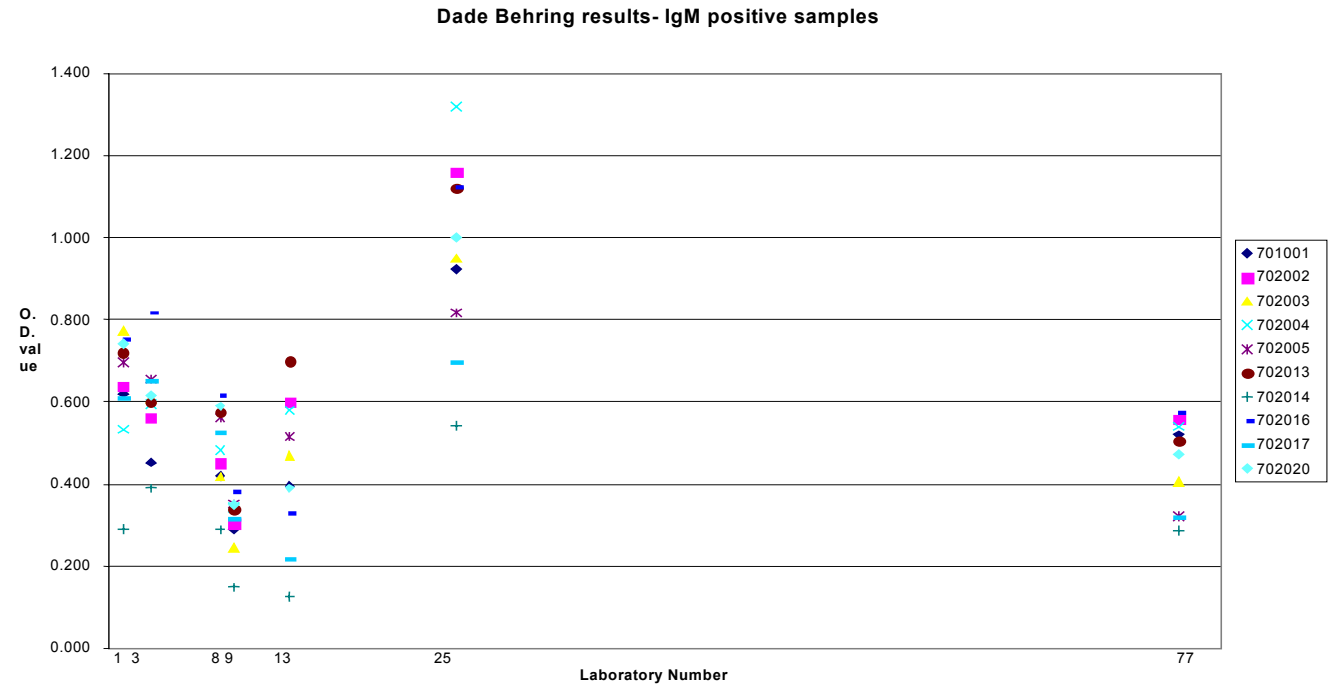

Figure 2: OD values for each positive panel sample by laboratory for laboratories using the DadeBehring kit. The cut-off is 0.2 OD units.

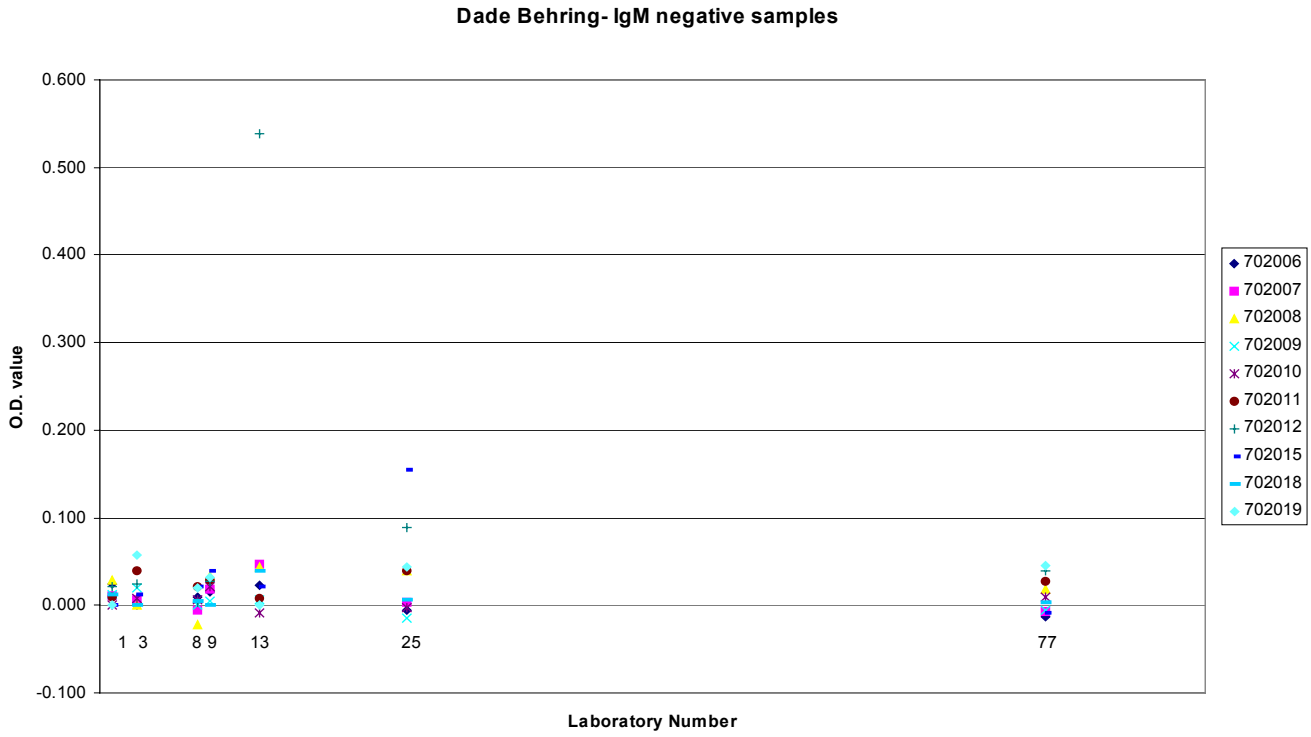

Figure 3: OD values for each negative panel sample by laboratory for laboratories using the Dade Behring assay. The cut-off is 0.2 OD units, a true negative is <0.1OD units and equivocal range 0.1- 0.2 OD units.

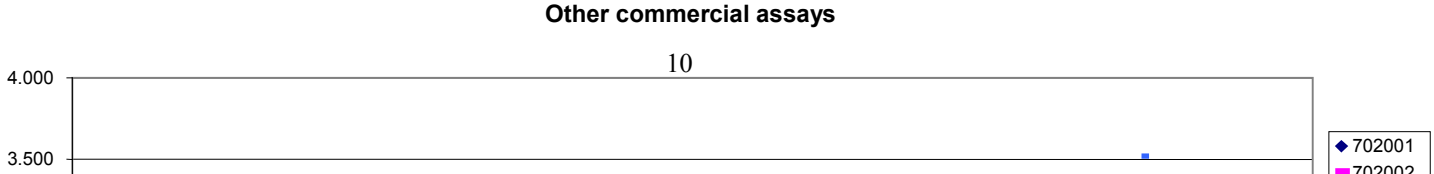

Figure 4: OD value of all panel samples for laboratories using a selection of commercial assays other than DadeBehring.
Laboratories 5 & 10 used the Chemicon assay and laboratories 37 & 74 used the Virion assay.

Results analysed by panel number

Twelve (12) laboratories achieved a perfect score (20/20)

SCORE	NUMBER OF LABS
20/20	12
19/20	2
18/20	1
14/20	1
11/20	1
TOTAL	17

Table 4: Scores achieved by participating laboratories assessed by the P/N interpretations as returned to VIDRL.

Results by panel number

Panel no.	001	002	003	004	005	006	007	008	009	010
Measles IgM status	P	P	P	P	P	N	N	N	N	N
Number correct	17 100%	17 100%	17 100%	17 100%	17 100%	15 88%	17 100%	15 88%	15 88%	16 94%

Panel no.	011	012	013	014	015	016	017	018	019	020
Measles IgM status	N	N	P	P	N	P	P	N	N	P
Number correct	15 88%	15 88%	17 100%	15 88%	14 82%	17 100%	17 100%	16 94%	15 88%	17 100%

Table 5: Percentage of laboratories reporting correct result for each individual panel number.

Analysis of discrepant results

Panel no.	006	008	009	010	011	012	014	015	018	019
Measles IgM status	N	N	N	N	N	N	P	N	N	N
Positive	2	2	1		2	2		2	1	1
Negative							1			
Equivocal			1	1			1	1		
Not tested										1
Total	***2***	***2***	***2***	***1***	***2***	***2***	***2***	***3***	***1***	***2***

Table 6: Details of result classification for those panel samples which were not correctly identified by reporting laboratories.

Rubella serology

Fifteen laboratories returned results for rubella IgM. Nine laboratories tested the whole panel for Rubella IgM, and six laboratories tested between 3 and 11 samples.

Kit Details

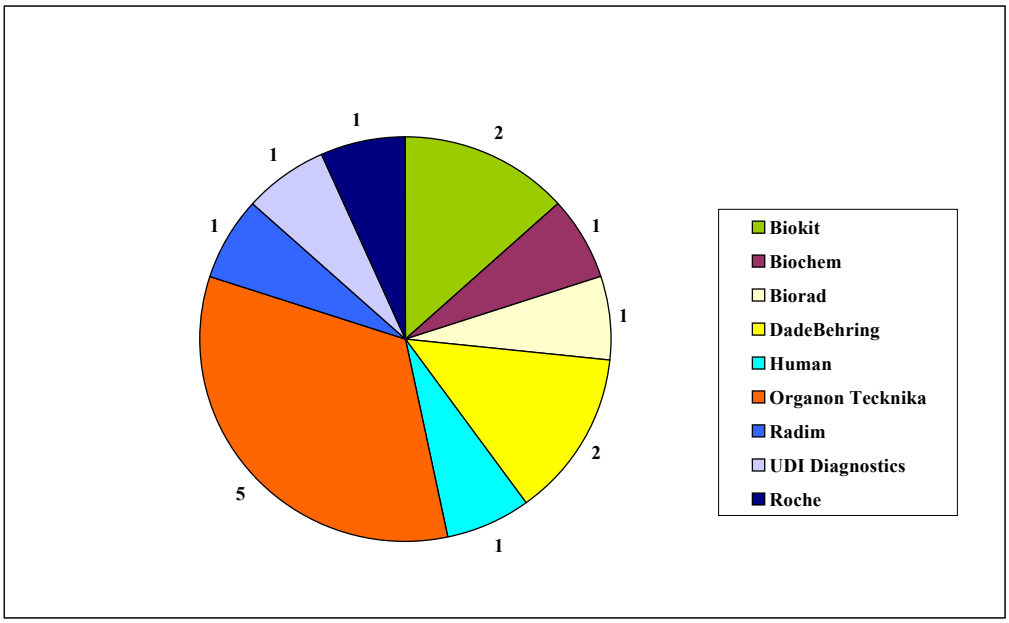

Figure 5: Methods in use for rubella IgM testing

Analysis of discrepant results

Panel No.	001	005	012	017
Diagnosis	Measles	Measles	Healthy adult	Measles
Rubella IgM POS		1	1	2
Rubella IgM EQUIV	1		1	

Table 7: Details of result classification for those panel samples which were not correctly identified by reporting laboratories for rubella IgM.

Discussion

Measles

The panel was distributed to National and Regional Measles reference laboratories within the WHO EMRO measles network and 17 laboratories returned results for analysis.

The results overall were very encouraging. Eighty-two percent of laboratories achieved a score of 90% or greater. A score of 100%was achieved by 71% of laboratories. The Dade Behring Enzygnost IgM measles kit was used by 41% of laboratories for investigating measles so we were able to compare the reactivity of samples for these laboratories. The number of users of other kits was too small for any comparisons.

Among users of Dade Behring EIAs there were 3 samples where discrepant results were reported.

1. Sample 012, reported positive by lab 013 (healthy volunteer)
2. Sample 014, reported negative by lab 019 (actually in equivocal range) and equivocal by lab 013(this sample was from a young adult with laboratory confirmed measles by serology &PCR)
3. Sample 015, reported equivocal by lab 025 (healthy volunteer)

Laboratory 025 had O.D. values noticeably higher than the other Dade Behring users and may wish to review their protocols.

There were 10 sera where discrepant results were reported. One was a measles IgM positive sample the other nine were measles IgM negative samples (5 healthy volunteers, 2 parvovirus IgM positive, 1 dengue IgM positive and 1 rubella IgM positive). Two laboratories reported the majority of discrepant results. These two laboratories (010 and 075) were using Chemicon and Biotest assays respectively. Laboratory 075 did not give a batch number or expiry date for the kit used.

Most users of alternative assays to DadeBehring had perfect scores. The exceptions were laboratories 010 and 075 using Chemicon & Biotest assays respectively. Laboratory 010 used the same kit and lot number as laboratory 011 who achieved a perfect score. These laboratories obtained false positive results on nine samples (010, 011, 012, 015, 018, 006, 009, 008, & 019, 5 healthy volunteers, 2 parvovirus IgM positives,1 dengue IgM positive and 1 rubella IgM positive respectively)..The discrepant results were very close to the cut-off value in contrast to the true positives which gave significant O.D. values.

Good data reporting is just as important as obtaining the correct result. Three laboratories did not supply a cut-off value and one laboratory did not supply any values for the samples tested .

Rubella

Although not designed as a rubella QAP panel,testing this measles QAP for rubella gave an opportunity for some comparative rubella testing on well characterised samples.
Sample 015 was an unsatisfactory sample for Rubella IgM testing as it is known to be IgM positive by a number of commercial kits but is not from a patient with a clinical illness. Excluding sample 015 from the analysis 67% of laboratories reported all samples that they tested correctly, 2 laboratories reported only one incorrect result and another 2 laboratories had 2 incorrect results. One laboratory tested 10 samples and reported them all positive as they diluted all samples 1:100 (due to insufficient volumes) and then multiplied the result by 100.

Excluding the results of the laboratory that altered the kit protocol and reported all samples as positive there was 4 false positive results reported :3 were measles IgM positive samples and one was a healthy volunteer.