



*Blood and Transplant*



# Evaluation of Abbott Architect Rubella IgG Assay Product code 6C17

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## Introduction

The Microbiological Diagnostics Assessment Service (HPA-MiDAS) in conjunction with the National Blood Service carried out evaluations of six assays for the Abbott Architect i2000SR analyser. The object of the evaluations was to assess the ability of the Architect HBsAg, HIV Ag/Ab combo, Anti-HCV, Rubella IgG, anti-CMV and Syphilis TP assays to detect serological evidence of each respective marker in human serum and plasma specimens.

The results of the evaluation of the Rubella IgG assay are presented in this report. The kits were tested against a panel of serum/plasma samples found to be either reactive or unreactive by relevant screening assays used in Europe. In addition, several sequential blood collections from individuals undergoing seroconversion for the relevant marker (chosen to compare directly with a range of other assays), and national quality control samples were incorporated in this evaluation.

Abbott Diagnostics provided all equipment, reagents and consumables required for this evaluation. They were responsible for the training of the operators in the use of the analyser and for the installation and ongoing maintenance and repair of any faulty equipment. The Architect analyser was installed in the National Transfusion Microbiology Reference Laboratory (NTMRL), North London Blood Centre, where all testing took place.

HPA-MiDAS staff were responsible for the testing of the evaluation panel which consisted of anti-rubella negative and positive samples, seroconversion and performance panels and quality control samples. NTMRL staff were responsible for the testing of a panel of weakly positive samples and the NBS Rubella Lot Release Testing panel.

## Description of the assay

The Abbott Architect Rubella IgG assay is a two-step sandwich chemiluminescent microparticle immunoassay (CMIA) for the quantitative determination and qualitative detection of IgG antibody to rubella virus in human serum or plasma. IgG antibody to rubella present in the sample binds to rubella antigen coated paramagnetic particles. After a wash step, anti-human IgG acridinium-labelled conjugate is added. Following a further wash step, pre-trigger solution (hydrogen peroxide) and trigger solution (sodium hydroxide) are added. The resulting chemiluminescent reaction is measured in relative light units (RLUs) which are directly proportional to the amount of rubella antibody present in the sample.

The assay was evaluated to determine its ability to detect IgG antibody to rubella. The Architect Rubella IgG assay was run on the i2000SR analyser at the NTMRL. HPA-MiDAS provided a panel of samples which comprised rubella antibody positive and negative specimens. The NTMRL, NLBS provided a panel of weakly positive samples and the samples used for the NBS Lot Release Programme.

**Table 1. Assay information**

<b>General</b>	
Assay name	Architect Rubella IgG
Manufacturer/UK agent	Abbott Diagnostics
Product number	6C17
Number of tests per pack	100 / 500
Sample volume (including 'dead volume')	150µL
<b>Presentation</b>	
Assay type	Two-step chemiluminescent sandwich immunoassay
Solid phase	Paramagnetic microparticles coated with partially purified rubella virus
Conjugate	Acridinium-labelled anti-human IgG (mouse, monoclonal)
Substrate	Pre-trigger - hydrogen peroxide solution Trigger - sodium hydroxide solution
Negative control	1
Positive control	2
Reading wavelength	n/a - chemiluminescent
Cut-off computation	n/a - curve fit. 10.0IU/mL cut-off value
Equivocal zone	5.0 - 9.9IU/mL
<b>Stages</b>	
Preparation/sample well loading	5 minutes
Specimen volume	25µL
Incubation status	37°C
Sampling time - 1 sample	1 minute
Time to completion (from initial loading of first sample)*	
- 1 sample	37 minutes
- 10 samples	43 minutes
- 100 samples	97 minutes
<b>Additional equipment requirement</b>	
Centrifuge	
Latex/nitrile gloves & personal protective equipment	
<b>Note:</b> * These data were observed timings by the evaluator. Information provided by Abbott Diagnostics: Throughput approximately 50 tests in the first hour and 100 tests per hour after the first result is generated.	

## Evaluation panel and method

A total of 798 samples were included in the evaluation panel.

The NTMRL panel comprised 65 samples referred to NTMRL for further testing and which gave results between 9 and 15IU/mL and 15 samples that comprise the NBS Rubella Lot Release Testing panel, Table 2.

The evaluation panel used by the HPA-MiDAS totalled 718 specimens, Table 2. Of the 718 specimens, 545 specimens were blood donations obtained from the NBS (261 previously screened and 284 previously unscreened for antibody to rubella), 29 were clinical samples, 11 from the HPA Quality Control Reference Unit and 50 from commercial sources. The rubella antibody status of the samples was ascertained by testing with three commercially available rubella antibody EIA test kits<sup>1</sup>. The Bioelisa Rubella IgG colour and Mercia Rubella IgG assays had been standardised by the manufacturer against the 1st International Rubella Standard (RUBI-1-94) and the Captia Rubella IgG assay against the 2nd British Standard (67/182). For

those samples which gave discordant results, further testing by EIA and in-house GACELISA was carried out in the Virus Reference Department, HPA, and by single radial haemolysis at the Microbiology Department, Royal Preston Hospital. From the results obtained it was determined that 329 specimens were rubella IgG antibody negative and 295 were rubella IgG antibody positive, however 11 specimens remain for which the status is not certain<sup>1</sup>.

In addition, seventy-three samples were the constituent members of three seroconversion panels and one performance panel. Nine quality control samples and one British standard were also tested.

**Table 2. Evaluation panel**

Specimen Category	Number
<b>NTMRL</b>	
1 Anti-Rubella IgG 9-15IU/mL (ante-natal patients' samples)	65
2 Rubella Lot Release Panel	15
<b>HPA-MiDAS</b>	
<b>1 Anti-Rubella IgG negative n=329</b>	
Screened NBS Blood donations	234
Unscreened NBS Blood donations	13
Quality Control Reference Unit (ESL/HPA)	9
BCP	45
Kings College Hospital	28
<b>2 Anti-Rubella IgG positive n=295</b>	
Screened NBS Blood donations	25
Unscreened NBS Blood donations	263
Quality Control Reference Unit (ESL/HPA)	2
BBI	5
<b>3 Anti-Rubella IgG indeterminate n=11</b>	
Screened NBS Blood donations	2
Unscreened NBS Blood donations	8
Kings College Hospital	1
<b>4 Seroconversion panels</b>	
BBI: RP001	15
BBI: RP011	20
BBI: RP014	13
<b>5 Performance panel</b>	
BBI: PTR201	25
<b>6 Quality Control specimens</b>	
HPA anti-rubella QC1	3x 1
Accurun 140 rubella positive control	3x 1
Accurun 25 multi-marker positive control	3x 1
Accurun 810 multi marker negative control	3x 1
Virotrol ToRCH positive control	3x 1
Virotrol-I multi-marker positive control	3x 1
Viroclear negative control	3x 1
Viroclear ToRCH positive control	3x 1
Bio-Rad Liquicheck ToRCH Plus positive control	3x 1
NIBSC 2nd British anti-Rubella Standard 67/182 (80IU/mL)	3x 1
<b>TOTAL (number of tests)</b>	<b>798</b>
<b>Notes:</b>	
BBI = Boston Biomedica Inc (SeraCare Inc). BCP = BioClinical Partners (Zeptometrix Inc.).	
NIBSC = National Institute for Biological Standards and Control.	

The method described in the kit insert was strictly followed. Abbott Architect is a fully automated analyser; all processing steps are performed on the instrument. The Architect assay parameters are factory set and defined in the system software.

A daily maintenance program is followed each day, the steps for which are prompted on the display screen. Principally, Probe Conditioning Solution and sodium hypochlorite solution are loaded onto the analyser by the operator and the analyser completes the program automatically. This process takes approximately 20 minutes. A weekly maintenance program is also required in which the sample, reagent and wash probes are cleaned with cotton-wool swabs soaked in distilled water and the air filters are cleaned.

Prior to running the analyser, test reagents, pre-trigger solution, trigger solution, wash buffer and reaction vessels are loaded onto the analyser and automatically primed and loaded as appropriate. The latter two may be added at any time irrespective of the analyser mode.

Prior to running a new batch of an assay, a calibration must first be performed. The calibrators are provided in dropper bottles and an appropriate volume is placed into sample cups and loaded onto the analyser. The calibration is valid for all subsequent tests using that particular lot number; it is not time limited. For the Rubella IgG assay, the calibrator pack consists of a series of six calibrators with rubella antibody concentrations of 0.0, 5, 15, 75, 250 and 500IU/mL. A calibration curve is produced by the analyser against which the RLU reading from each sample is compared to give the number of international units per mL.

Three rubella antibody kit controls are provided by Abbott. It is recommended that these are run at least once within every 24 hours that the test is in use. The rubella antibody controls consist of a Negative Control (0.0IU/mL, range 0-0.04IU/mL), Positive Control 1 (25IU/mL, range 0.15-0.35IU/mL) and Positive Control 2 (300IU/mL, range 180-420IU/mL).

Samples may be loaded in their primary tubes, if suitable for the analyser, or aliquotted into Architect sample cups. Once ordered and the analyser put into 'running' mode, sample processing is initiated by the loading of the samples onto the analyser. The reactions occur in the following processing sequence: -

- A reaction mixture is formed combining sample, sample diluent and microparticles in the reaction vessel.
- After the first incubation is complete, the reaction mixture undergoes a wash step. A magnetic field is applied to retain the paramagnetic microparticles within the reaction vessel during the wash procedure.
- The anti-human IgG/acridinium conjugate is then added and a further incubation takes place.
- Following a second wash step, pre-trigger (hydrogen peroxide) and trigger (sodium hydroxide) solutions are added to the reaction vessel
- The resultant chemiluminescent signal is measured and expressed as Relative Light Units.

The time taken from loading a sample to obtaining a result was 37 minutes for the Rubella IgG assay. Subsequent results are obtained every 36 seconds, assuming continuous loading of samples. (18 seconds is a set cycle time per reaction vessel, for which two are required for the Rubella IgG assay, and does not vary.)

The cut-off sensitivity for the Abbott Architect Rubella IgG assay is set by the manufacturer at 10IU/mL. The results are expressed as follows: 0.0-4.9IU/mL is considered negative, 5.0-9.9IU/mL equivocal and  $\geq 10$ IU/mL positive. All results for this assay are presented as IU/mL in

this report. The assay was standardised by Abbott Diagnostics against the 1st Rubella International Standard (RUBI-1-94).

## Specificity

Three hundred and twenty-nine anti-rubella IgG negative specimens were tested in the Architect Rubella IgG assay, lot number 17563UN07. On initial testing, 326 samples were unreactive (*ie* <4.9IU/mL) to give a specificity of 99.1% (95% confidence interval 97.4-99.8%), 2 specimens (0.6%) were equivocal and one specimen (0.3%) was reactive in the Architect Rubella IgG assay. Table 3 shows further analysis of the results along with the results of three other assays currently under evaluation.

The initially false positive specimen was unreactive after retesting in duplicate. Both specimens that gave initially equivocal results (5.49 and 4.68IU/mL) were again equivocal after retesting (5.88/5.53 and 6.11/5.12, respectively), to give a final specificity of 99.4% (95% confidence interval 97.8-99.9%). Table 4 shows further analysis of the results along with the results of three other assays currently under evaluation.

**Table 3. Initial specificity**

Assay (Manufacturer/agent)	Product code	Number tested	Number positive >10IU (reactive rate)	Number equivocal 5-9.9IU (reactive rate)	Number negative <4.9IU (specificity)	95% confidence interval %	Range IU	Mean IU	Median IU
Architect Rubella IgG	6C17-25	329	1 (0.3%)	2 (0.6%)	326 (99.1%)	97.4-99.8	0-158.3	0.73	0.14
Assay	Product code	Number tested	Number positive >11IU (reactive rate)	Number equivocal 9.1-10.9IU (reactive rate)	Number negative <9IU	95% confidence interval %	Range IU	Mean IU	Median IU
Bioelisa Rubella IgG (Biokit/Launch)	3000-1219	329	0 (0.0%)	0 (0.0%)	329 (100%)	98.9-100	<9.2 - <9.2	<9.2	<9.2
Captia Rubella IgG (Trinity Biotech)	2325300	329	2 (0.6%)	1 (0.3%)	326 (99.09%)	97.4-99.8	2.5-17.2	2.9	2.6
Mercia Rubella IgG (Microgen Bioproducts)	M5066	329	2 (0.6%)	0 (0.0%)	327 (99.39%)	97.8-99.9	2.7-14.7	3.8	3.5

**Table 4. Repeat specificity**

Assay	Product code	Number tested	Number positive >10IU (reactive rate)	Number equivocal 5-9.9IU (reactive rate)	Number negative <4.9IU (specificity)	95% confidence interval %	Range IU	Mean IU	Median IU
Architect Rubella IgG	6C17-25	329	0 (0.0%)	2 (0.6%)	327 (99.4%)	97.8-99.9	0-5.6	0.3	0.1
Assay	Product code	Number tested	Number positive >11IU (reactive rate)	Number equivocal 9.1-10.9IU (reactive rate)	Number negative <9IU	95% confidence interval %	Range IU	Mean IU	Median IU
Bioelisa Rubella IgG (Biokit/Launch)	3000-1219	329	0 (0.0%)	0 (0.0%)	329 (100%)	98.9-100	<9.2 - <9.2	<9.2	<9.2
Captia Rubella IgG (Trinity Biotech)	2325300	329	0 (0.0%)	1 (0.3%)	328 (99.7%)	98.3-100	2.5-9.7	2.8	2.6
Mercia Rubella IgG (Microgen Bioproducts)	M5066	329	0 (0.0%)	0 (0.0%)	329 (100%)	98.9-100	2.7-7.6	3.7	3.5

## Sensitivity

Two hundred and ninety-five specimens positive for rubella IgG were tested in the Architect Rubella IgG assay, lot number 17563UN07, by HPA-MiDAS. On initial testing, 288 were reactive by the assay (ie  $\geq 10$  IU/mL) to give an initial sensitivity of 97.63% (95% confidence interval 95.6-99.2%), six (2.03%) were equivocal (ie  $\geq 5.0 \leq 9.9$  IU/mL) and one (0.34%) was unreactive. The results are presented in Table 5 with three other assays currently under evaluation.

Following retests, the negative sample and six equivocal samples were again unreactive and equivocal, respectively, Table 6. The six samples were also tested using the second lot of the Architect Rubella IgG (Lot number 14966UN06). The S/CO values obtained were very similar to those for the main lot (Lot number 17563UN07).

**Table 5. Initial sensitivity**

Assay	Product code	Number tested	Number negative <4.9IU	Number equivocal 5.0-9.9IU	Number positive >10IU (Sensitivity)	95% confidence interval %	Range IU	Mean IU	Median IU
Architect Rubella IgG	6C17	295	1 (0.34%)	6 (2.03%)	288 (97.63%)	95.2-99.0	4.4->500	73	51.1
Assay	Product code	Number tested	Number negative <9IU	Number equivocal 9.1-10.9IU	Number positive >11IU (Sensitivity)	95% confidence interval %	Range IU	Mean IU	Median IU
Bioelisa Rubella IgG (Biokit/Launch)	3000-1219	295	0 (0%)	7 (2.37%)	289 (98.0%)	95.6-99.2	<9.23->54.19	39.9	43.3
Captia Rubella IgG (Trinity Biotech)	2325300	295	6 (2.03%)	10 (3.39%)	279 (94.6%)	91.3-96.9	2.5-31.1	17.5	17.8
Mercia Rubella IgG (Microgen Bioproducts)	M5066	295	10 (3.39%)	6 (2.03%)	276 (93.6%)	90.1-96.1	5.4-160.0	24.3	20.5

**Table 6. Repeat sensitivity**

Assay	Product code	Number tested	Number negative <4.9IU	Number equivocal 5.0-9.9IU	Number positive >10IU (Sensitivity)	95% confidence interval %	Range IU	Mean IU	Median IU
Architect Rubella IgG	6C17	295	1 (0.34%)	6 (2.03%)	288 (97.63%)	95.2-99.0	4.5->500	73	51.1
Assay	Product code	Number tested	Number negative <9IU	Number equivocal 9.1-10.9IU	Number positive >11IU (Sensitivity)	95% confidence interval %	Range IU	Mean IU	Median IU
Bioelisa Rubella IgG (Biokit/Launch)	3000-1219	295	0 (0%)	1 (0.33%)	294 (99.7%)	98.1-100	<9.23->54.19	39.7	43.2
Captia Rubella IgG (Trinity Biotech)	2325300	295	2 (0.68%)	2 (0.66%)	291 (98.6%)	96.6-99.6	6.9-31.1	17.8	17.8
Mercia Rubella IgG (Microgen Bioproducts)	M5066	295	2 (0.68%)	0 (0%)	293 (99.3)	97.6-99.9	7.1-160.0	24.9	20.7

NTMRL tested 65 specimens that were selected from the NTMRL database as being between 9 and 15IU/mL by the Biokit Bioelisa Rubella IgG Colour assay. The samples had been previously screened as negative or equivocal by the Microgen Mercia Rubella G assay, Product code M5066, and referred to NTMRL for further testing.

Forty-nine of the 65 samples were nonreactive by the Architect Rubella IgG assay (lot number 17563UN07), giving reactivities between 0.655 - 4.919IU/mL and 16 were within the greyzone with reactivities between 5.02 – 8.763, Figure 1.

The NBS Rubella Lot Release panel comprises 15 samples that are positive for Rubella IgG with reactivities ranging from weakly to strongly positive. These samples were tested by NTMRL and all 15 samples were detected by the Architect Rubella IgG assay, with reactivities ranging from 29.54 to 93.48IU/mL, Table 7.

**Table 7. Results of Rubella Lot Release Testing Panel**

Panel/Ctrl Member	Expected Ratios	Expected Result	Architect Rubella IgG	
			IU/mL	Result
1	>0.6	Positive	29.54	Reactive
2	>1	Positive	79.37	Reactive
3	>1	Positive	93.48	Reactive
4	>1	Positive	83.18	Reactive
5	>1	Positive	80.84	Reactive
6	>1.4	Positive	89.98	Reactive
7	>1.4	Positive	83.93	Reactive
8	>0.3	Positive	44.14	Reactive
9	>0.4	Positive	55.05	Reactive
10	>0.5	Positive	54.28	Reactive
11	>0.6	Positive	44.46	Reactive
12	>0.7	Positive	52.36	Reactive
13	>0.8	Positive	61.69	Reactive
14	>0.8	Positive	67.23	Reactive
15	>0.8	Positive	51.89	Reactive

## Indeterminate specimen results

HPA-MiDAS also tested 11 samples for which a final status of positive or negative had not been determined during previous characterisation testing. The results of initial testing only in the Architect Rubella IgG assay (the samples were not repeat tested) and the results from previous characterisation testing are shown in *Appendix Table 12*.

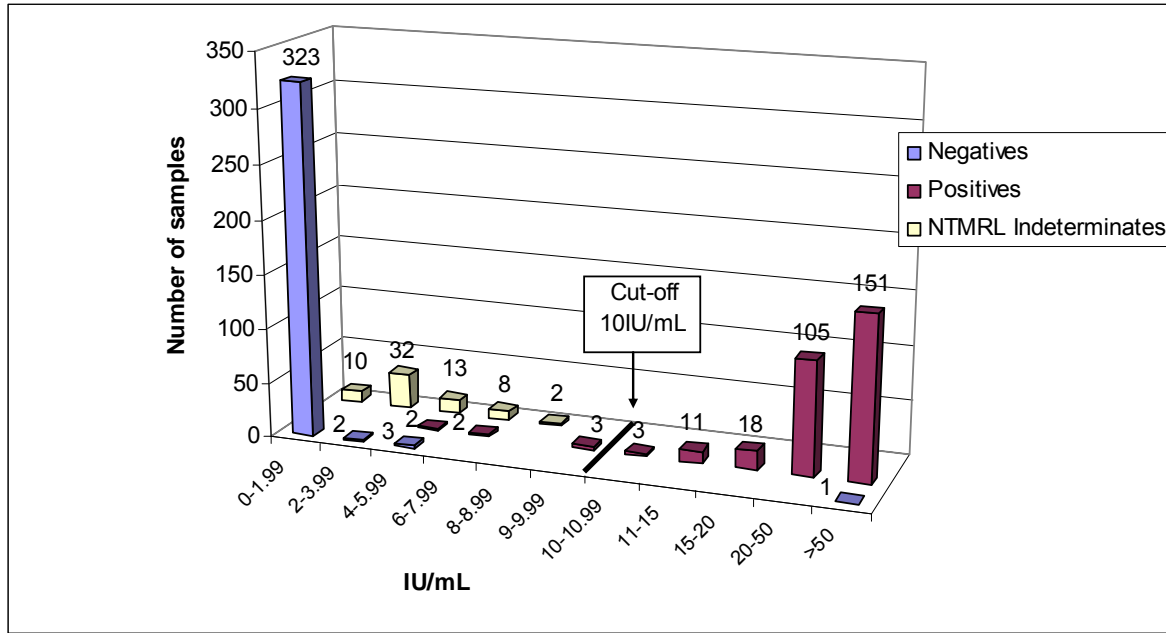
## Distribution of initial reactivities

The distribution of initial reactivities for the 329 rubella IgG negative and the 295 rubella IgG positive specimens, tested by HPA-MiDAS; and the 65 rubella IgG samples (9-15IU/mL), tested by NTMRL, is shown in Figure 1. Assays with good discrimination have few or no samples wrongly classified and few reactions close to the cut-off.

For the HPA-MiDAS samples, the Architect Rubella IgG assay gave one initially reactive result for the negative samples and one negative and six equivocal results for the positive samples. The discordantly reactive samples gave the same results when retested in duplicate.

For the NTMRL samples, 49 of the 65 samples were nonreactive (<4.9IU/mL) and 16 were equivocal (5-9.9IU/mL) in the Architect Rubella IgG assay. These specimens were not retested.

**Figure 1. Distribution of initial reactivities**



**Note.** The scale used for the IU/mL values is not continuous

### Commercial panel sensitivity

Three commercial seroconversion panels were tested in both batches of the Architect Rubella IgG assay evaluated. Of the 48 specimens represented in the three panels, 32 were detected by the Architect Rubella IgG assay. Complete data for the three panels are available for three other rubella assays. The results indicate that, In a comparison with these three assays, the Architect Rubella IgG assay gave highest score, however the number of panels is limited, Tables 8 and 13

**Table 8. Comparative seroconversion sensitivity for 3 panels**

Panel	Number of specimens in panel	Architect Rubella IgG	Bioelisa Rubella IgG	Captia Rubella IgG	Mercia Rubella IgG	Abbott* Rubazyme-M IgM	Abbott* Rubazyme IgG	Gull Labs* ELISA Rubella IgM	Gull Labs* ELISA Rubella IgG
		IU/mL	IU/mL	IU/mL	IU/mL	S/CO	S/CO	S/CO	S/CO
RP001	15	9 (21)	7 (28)	8 (24)	6 (31)	9 (21)**	8 (24)	9 (21)**	7 (28)
RP011	20	15 (19)	15 (19)	15 (19)	15 (19)	15 (19)**	15 (19)	16 (16)**	15 (19)
RP014	13	8 (19)	7 (21)	8 (19)	4 (33)	8 (19)**	8 (19)	8 (19)**	8 (19)
<b>Total</b>	<b>48</b>	<b>32</b>	<b>29</b>	<b>31</b>	<b>25</b>	<b>32</b>	<b>31</b>	<b>33</b>	<b>30</b>

**Notes:**

All panels taken from male individuals following vaccination with Meruvax II Vaccine.  
 The total for each assay was calculated by summing the correct positive reactions for each of the panels. A higher score suggests higher sensitivity. The number in parenthesis is the number of days from the initial bleed to the first positive sample.  
 \* Results were extracted from BBI / BCP data sheets  
 \*\* IgM detection assays therefore panels began positive but had one or more negative results later on in the panel.

One commercial rubella IgG performance panel, PTR201, was also tested in both lots of the Architect Rubella IgG assay evaluated. The panel consisted of 23 rubella IgG positive and two rubella IgG negative samples. Comparative data for three other assays are available, see Table 9.

For both lots of the Architect Rubella IgG assay (17563UN07 and 14966UN06), similar S/CO ratios were obtained for panel PTR201, *Appendix* Table 13. Of the 23 rubella IgG panel members, 22 were reactive and one was equivocal (please note that insufficient of one sample was available for testing in Lot 17563UN07, however the sample was highly reactive in Lot 14966UN06). The two rubella IgG negative panel members were unreactive in both lots.

**Table 9. Comparative sensitivity for one performance panel**

Panel	Number of specimens in panel	Architect Rubella IgG	Bioelisa Rubella IgG	Captia Rubella IgG	Mercia Rubella IgG	Abbott* EIA Rubella IgG	Abbott* IMx Rubella IgG	Murex* Rubella Total (Latex Agg.)
PTR201	25	22**	22	23	21	23	23	23

**Notes:** The total for each assay was the number of reactive samples for the panel

\*Data taken from BBI data sheet.

\*\* Sample PTR201-20 has not been tested in Lot 1 however this sample was strongly positive in Lot 2. Sample number PTR 201-22 was equivocal in both Lot 1 and Lot 2 of the assay.

## Lot comparison

A subset of the main evaluation panel was tested in a second lot of the assay (Lot number 14966UN06). Forty rubella IgG positive specimens, 20 negative specimens, three seroconversion panels, one performance panel and ten quality control samples were compared, Tables 10, 11 and 13. The IU/mL results obtained from both lots were similar for all the samples compared.

**Table 10. Comparison of two lots of Architect Rubella IgG**

Specimen category	Number of specimens	Number of initial reactive specimens (>10IU)	
		17563UN07	14966UN06
Rubella IgG negative	20	0	0
Rubella IgG positive	40	40	40
RP001	15	9	9
RP011	20	15	15
RP014	13	8	8
PTR201	24	21	21
<b>Total</b>	<b>132</b>	<b>93</b>	<b>93</b>

**Notes.** PTR201-20 not tested in lot 1 therefore not included in table.  
PTR201-22 equivocal in both assays.

**Table 11. Comparison of 9 quality control samples and 2nd British Standard**

Quality control sample	Architect Rubella IgG							
	Lot 17563UN07 (IU/mL)				Lot 14966UN06 (IU/mL)			
	Test 1	Test 2	Test 3	Mean	Test 1	Test 2	Test 3	Mean
HPA anti-rubella QC1	22.91	24.92	23.78	23.87	25.17	23.70	22.26	23.71
Accurun 140 rubella positive control	18.97	18.01	19.09	18.69	17.78	18.01	17.60	17.80
Accurun 25 multi-marker positive control	22.54	22.75	24.81	23.37	22.24	21.84	22.51	22.19
Accurun 810 multi marker negative control	122.59	116.96	118.60	119.38	105.04	99.02	103.78	102.61
Virotrol ToRCH positive control	16.03	14.37	15.48	15.30	14.67	15.00	14.70	14.79
Virotrol-I multi marker control	18.02	18.08	17.60	17.90	16.54	16.77	15.41	16.24
Viroclear negative control	0.29	0.24	0.16	0.23	0.33	0.33	0.30	0.32
Viroclear ToRCH positive control	35.89	37.94	37.40	37.08	33.44	33.78	31.71	32.97
Bio-Rad Liquicheck ToRCH Plus positive control	14.09	15.03	14.33	14.48	14.73	13.16	14.30	14.06
NIBSC 80IU 2nd British Standard	58.04	61.39	64.19	61.21	60.97	57.59	62.69	60.42
NIBSC 80IU 2nd British Standard 1:10 dilution*	6.88	7.40	6.73	7.00	5.85	6.14	6.13	6.04

Notes. \*Standard reconstituted in 1 mL distilled water and an aliquot of a 1:10 dilution tested.

A quality control sample/statistical assay control should be chosen to have a reactivity within the linear dynamic range of the assay. Our findings suggest that the rubella IgG positive controls that give IU/mL between 15 and 50 would be suitable for use in the Architect CMV IgG assay.

## Conclusion

The evaluation of the Architect Rubella IgG assay resulted in an initial reactive rate of 0.9%. Of the 329 rubella IgG negative specimens tested one was reactive (158.3IU/mL) and two were equivocal (5.0IU/mL and 5.5IU/mL). The repeat reactive rate was 0.6%, whereby the two equivocal specimens were again equivocal following retests.

The initial and repeat sensitivity of the Architect Rubella IgG assay was 97.6%. Of the 295 Rubella IgG positive specimens tested, one was nonreactive (4.35IU/mL) and six were equivocal on initial testing. After retesting, all seven samples had similar IU/mL values.

Three seroconversion panels were tested for which the Architect Rubella IgG assay detected 32 of the total of 48 specimens. Three other assays with which it was compared detected 29, 31 and 25 of the 48 specimens.

A small lot comparison was undertaken in which both lots of the Architect Rubella IgG assay gave very similar results for all samples tested.

The results obtained from the 'indeterminate' samples illustrate difficulties (which are not assay dependent) that may be encountered in determining the negative/positive rubella antibody status of individuals, particularly where the antibody level is near or around the accepted immune cut-off of 10IU/mL.

## Reference

1. **Burgess C, Perry KR** (2008). Evaluation of three Rubella IgG Enzyme Immunoassays. NBSR06005.  
Available at [www.hpa-midas.org.uk/reports](http://www.hpa-midas.org.uk/reports)

Appendix

Table 12. Results of indeterminate samples

Specimen number	MiDAS test results										Virus Reference Division Test Results			Preston	
	Architect Rubella IgG	Assay 1			Assay 2			Assay 3			Microimmune Rubella IgM Capture EIA	Dade Behring Enzygnost Rubella IgG	In-house GACELISA	Result	SRH
	Initial result IU/mL	Initial result IU (OD/CO)	Repeat results IU (OD/CO)		Initial result IU (OD/CO <sup>#</sup> )	Repeat results IU (OD/CO <sup>#</sup> )		Initial result IU (OD/CO)	Repeat results IU (OD/CO)		OD Qualitative result	IU (OD*)	Qualitative result	Final status	Zone (mm) <sup>†</sup>
05R0118A	10.8	<9.2 (0.73)	9.3 (0.92)	10.5 (1.05)	8.5 (1.13)	8.8 (1.17)	9.4 (1.23)	7.0 (0.77)	8.4 (0.81)	10.9 (1.10)	0.021 negative	7 (0.20) positive	positive	Positive	7
05R0119A	2.1	12.7 (1.21)	12.3 (1.2)	11.0 (1.1)	9.8 (1.26)	7.4 (1.02)	7.4 (1.01)	10.0 (1.01)	10.4 (1.06)	8.7 (0.80)	0.032 negative	9 (0.24) positive	positive	Positive	0†
05R0145a	6.2	12.2 (1.17)	18.0 (1.5)	19.5 (1.6)	14.4 (1.62)	8.7 (1.16)	9.6 (1.25)	6.1 (0.62)	10.9 (1.03)	9.0 (0.85)	0.017 negative	10 (0.27) positive	positive	Positive	0†
05R0170a	10.3	<9.2 (0.79)	<9.2 (0.66)	9.9 (0.98)	8.7 (1.16)	5.0 (0.65)	6.1 (0.84)	10.6 (1.07)	10.8 (1.09)	11.6 (1.17)	0.009 negative	9 (0.229) positive	positive	Positive	7
05R0172a	12.9	10.9 (1.08)	<9.2 (0.37)	<9.23 (0.90)	8.7 (1.15)	7.6 (1.03)	8.2 (1.11)	6.8 (0.70)	12.0 (1.20)	13.8 (1.36)	0.013 negative	10 (0.267) positive	positive	Positive	7
05R0283a	9.6	9.7 (0.97)	<9.2 (0.91)	11.53 (1.13)	9.7 (1.25)	10.2 (1.30)	9.9 (1.27)	15.3 (1.47)	12.2 (1.23)	13.0 (1.30)	0.015 negative	8 (0.23) positive	positive	Positive	8
05R0360a	10.1	13.2 (1.23)	14.1 (1.33)	12.8 (1.23)	9.5 (1.24)	7.6 (1.03)	8.5 (1.13)	7.3 (0.69)	11.3 (1.14)	9.7 (0.96)	0.019 negative	8 (0.22) positive	positive	Positive	8
05r0043a	8.4	9.7 (0.98)	<9.2 (0.79)	9.4 (0.95)	12.9 (1.51)	9.2 (1.21)	8.6 (1.15)	6.6 (0.68)	8.2 (0.76)	9.6 (0.95)	0.011 negative	9 (0.23) positive	equivocal	Indeterminate	7
05R0052a	3.4	<9.2 (0.51)	<9.2 (0.84)	<9.2 (0.81)	10.3 (1.31)	7.7 (1.04)	8.1 (1.10)	5.4 (0.53)	7.2 (0.62)	7.2 (0.62)	0.013 negative	7 (0.19) equivocal	negative	Indeterminate	4
05R0290a	5.8	9.8 (0.98)	<9.2 (0.77)	<9.2 (0.92)	8.3 (1.12)	6.6 (0.91)	5.9 (0.82)	10.5 (1.05)	8.0 (0.67)	10.4 (1.06)	0.014 negative	8 (0.197) equivocal	positive	Indeterminate	8
06R0112a	9.7	10.3 (1.02)	11.3 (1.11)	14.0 (1.30)	9.8 (1.27)	8.0 (1.08)	7.6 (1.03)	9.2 (0.93)	12.0 (1.26)	12.9 (1.37)	0.023 negative	11 (0.288) positive	negative	Indeterminate	7

Notes. <sup>#</sup> Assay 2: OD/CO value 1.28 is equivalent to 10IU for this batch number.  
<sup>\*</sup> Enzygnost assay cut-off value = 0.200  
<sup>‡</sup> 15IU control ≅ 8mm  
<sup>†</sup> Results following absorption. In initial testing sample test zone = sample control zone.

**Table 13. Architect Rubella IgG results for three commercial seroconversion panels (RP001, RP011 and RP014) and one performance panel (PTR201).**

Panel	IU/mL		Panel	IU/mL	
	17563UN07	14966UN06		17563UN07	14966UN06
RP001-01	0.63	0.65	PTR201-01	15.39	15.66
RP001-02	0.49	0.63	PTR201-02	95.19	85.06
RP001-03	0.70	0.64	PTR201-03	116.48	109.06
RP001-04	0.45	0.52	PTR201-04	149.89	136.53
RP001-05	0.62	0.71	PTR201-05	0.31	0.45
RP001-06	3.53	4.14	PTR201-06	19.52	18.95
RP001-07	19.47	21.07	PTR201-07	51.96	51.84
RP001-08	27.88	27.13	PTR201-08	39.34	31.40
RP001-09	21.70	23.27	PTR201-09	98.67	79.95
RP001-10	69.27	68.88	PTR201-10	55.92	48.90
RP001-11	62.35	59.08	PTR201-11	41.59	39.18
RP001-12	51.31	42.32	PTR201-12	344.24	265.57
RP001-13	48.08	47.10	PTR201-13	80.40	78.54
RP001-14	44.52	44.46	PTR201-14	12.91	13.54
RP001-15	43.79	42.66	PTR201-15	275.07	247.44
RP011-01	0.41	0.40	PTR201-16	1.22	1.13
RP011-02	0.45	0.37	PTR201-17	56.79	52.26
RP011-03	0.42	0.41	PTR201-18	21.61	23.16
RP011-04	0.33	0.60	PTR201-19	33.47	31.17
RP011-05	3.39	3.77	PTR201-20	NT	109.41
RP011-06	30.72	30.58	PTR201-21	80.29	69.30
RP011-07	84.30	74.63	PTR201-22	7.64	8.49
RP011-08	90.83	86.65	PTR201-23	42.11	38.61
RP011-09	80.96	76.68	PTR201-24	16.50	17.78
RP011-10	78.43	76.75	PTR201-25	93.53	82.30
RP011-11	79.14	72.52			
RP011-12	80.04	73.95			
RP011-13	87.23	73.22			
RP011-14	82.79	64.39			
RP011-15	84.28	71.10			
RP011-16	82.22	81.64			
RP011-17	78.70	67.39			
RP011-18	75.02	70.71			
RP011-19	74.09	65.61			
RP011-20	82.81	66.35			
RP014-01	0.21	0.40			
RP014-02	0.26	0.26			
RP014-03	0.34	0.33			
RP014-04	0.45	0.52			
RP014-05	1.44	1.53			
RP014-06	25.84	27.59			
RP014-07	40.13	40.62			
RP014-08	52.74	54.15			
RP014-09	55.26	57.83			
RP014-10	55.44	57.31			
RP014-11	57.78	56.75			
RP014-12	60.19	54.34			
RP014-13	60.38	60.82			