Sanquin

CE/IVD-certified tests are not always better than LDT

Some examples

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For Life.

IVDR

From a clinical laboratory perspective

- **CE-IVD (Conformité Européenne - In Vitro Diagnostics)** certification is granted to in vitro diagnostic devices (IVDs) that comply with the **In-Vitro Diagnostics Directive (IVDD 98/79/EC)**. It signifies that the device meets safety and performance requirements for clinical use within the European Economic Area (EEA). CE-IVD certification is an important step for IVDs, as it allows them to be marketed and used in the EEA.
- **IVDR (In Vitro Diagnostic Regulation 2017/746)** will replace the IVDD starting from **May 26, 2022**. Unlike the IVDD, which was a directive, the IVDR is a legally binding regulation. Here are some key points about IVDR:

The aim was to improve quality of laboratory diagnostic services.

Is an CE-IVD certified test always better than an LDT?

→ no, give some examples

EU: IVD CE 2797

Risks:

- Fewer tests available
- For some orphan diseases, no CE/IVD marked test will be available
- IVD testing becomes more expensive
- One test serves all: what if this test is not OK in clinical use, although it fulfills the demands under IVDR
- What if a test is used outside the purpose / in an adapted fashion?

Classes
& consequences

- Class A no transition period; stays self-certification
- Class A Sterile and Class B to be sold untill 26 May 2027
- Class C to be sold untill 26 May 2026
- Class D to be sold untill 26 May 2025

DELAYED

Examples per class:

A) Low risk for individual and low risk for Public Health
eg DNA isolation kit, culture media

B) Intermediate risk for individual and/or low risk for Public Health
eg Pregnancy selftest, urinestrips, ***autoantibody testing, complement analysis***

C) High risk for individual and/or intermediate risk for Public Health
eg HLA typing, PSA screen, bloodsugar selftest

D) High risk for individual and high risk for Public Health
eg HepB blood donor screen, HIV test, ABO bloodgroup reagents

Hospital diagnostics & IVDR

Hospital diagnostics services + specialized regulated bioanalysis

Health institute, ISO15189 accredited

<u>IVDR for hospital diagnostic services:</u>

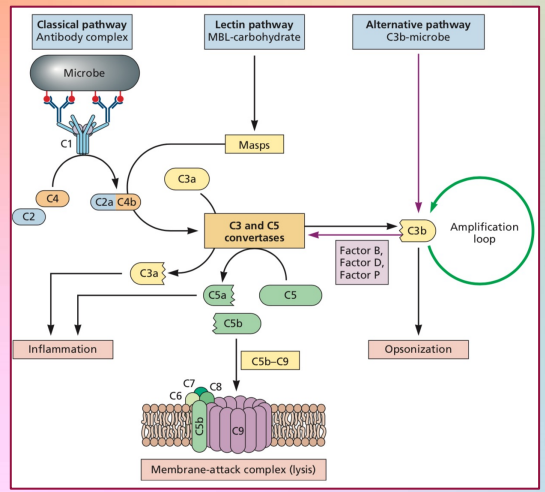
- If non CE-IVD assay is better: document why in dossier. You then are a manufacturer of the test: same rules account (from May 2024, except dossier).
- Dutch IVDR taskforce has formulated ‘rules’ voor adaptation of CE-IVD assay: *small adaptation are allowed (eg own PBS, other inc time)*

<u>IVDR for regulated bioanalysis:</u>

More and more requested/discussed by clients:
what to do and what not and from when?

Complement

Functional assays



CH50 or AP50 measured in context of assessing complement activation, when monitoring complement blocking therapy

→ CH50: 2 commercial CE/IVD marked assays
→ AP50: 1 commercial CE/IVD marked assay

LDT: hemolytic assays

Advantages LDT over commercial kits:

IVD assay	LDT
Not very sensitive	Sensitive and accurate in low range
Black & white result in low range	Reports a rest activity
Unlogic normal range (30-113%)	Logic normal range (70-100%)

The commercial kits may work when adapted, but then lose their CE/IVD certification

AP50 assay

Range

Performance characteristics

120 sera from blood donors were tested in the AP assay and the normal reference range was calculated. The values were expressed in % of the positive control. See Figure 1 and Table 1. In the study no blood donor was below 10 %.

Figure 1 AP assay.

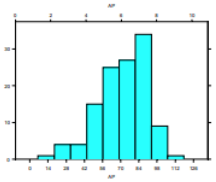
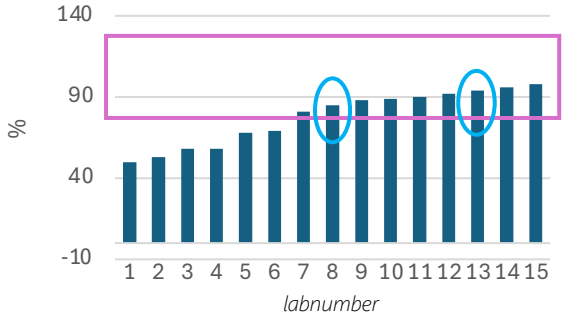


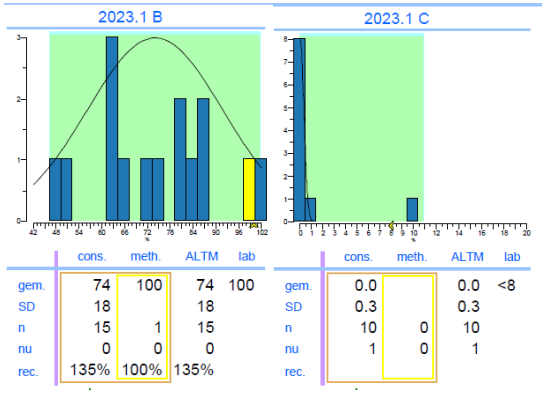
Table 1.

	n	Mean (%)	±2SD (%)	Median (%)
Alternative pathway	120	71	30-113	73

Interlab proficiency testing



Dutch quality survey (SKML)



Healthy donor *C6 deficient Pt*

CE/IVD assay

AP50

Purpose:

To determine if a patient had a complement deficiency or not

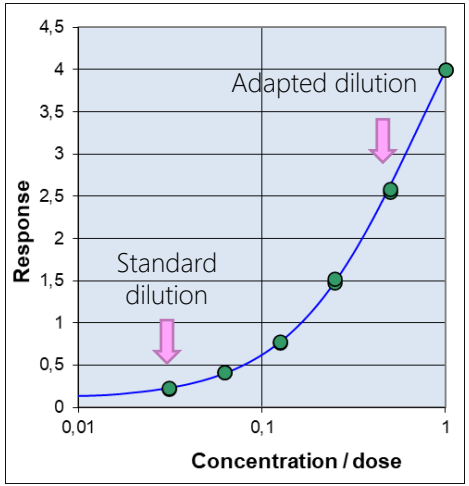
(Alternative use: monitoring therapy or disease)

	IVD assay (%)	LDT (%)
Pt1 (SLE)	10	53
Pt2 (deficiency)	0	<4
Pt3 (on C therapy)	0	17

(normal is 70-130%)

CP: 1:101 (re-test 1:201)
AP: 1:18

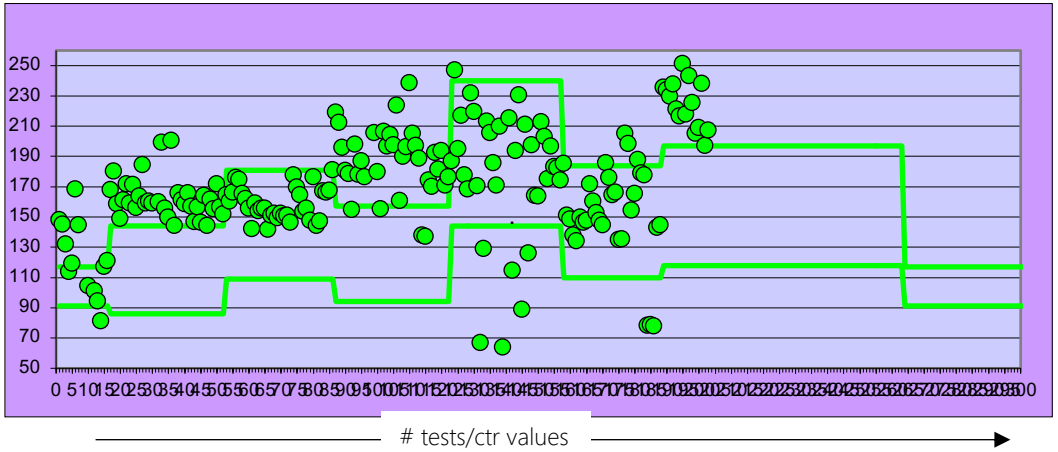
AP50



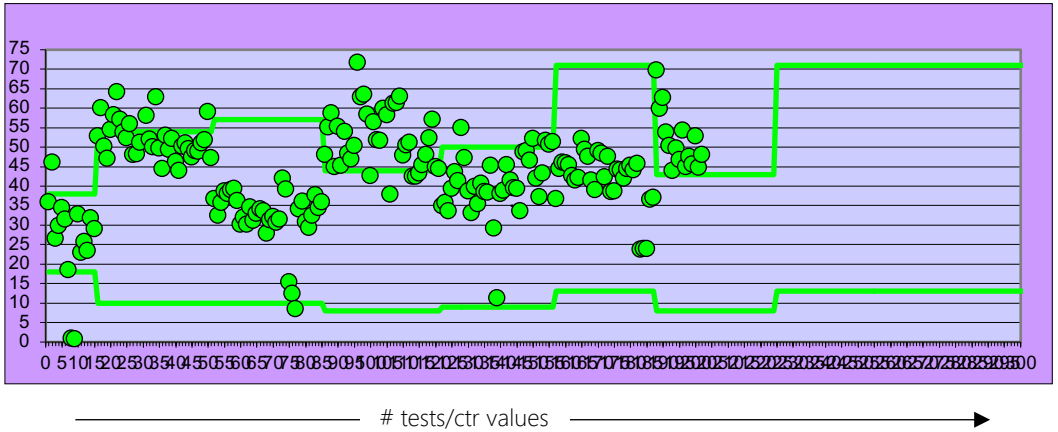
Kit QCs

In-house QC is OK

Kit high QC



Kit low QC



Anti-MOG antibodies

- Biomarker for MOGAD (transverse myelitis + NO)
- Excluding MS

Anti-Myelin-Oligodendrocyte Glycoprotein (MOG) IIFT
Instructions for the indirect immunofluorescence test

ORDER NO.	ANTIBODIES AGAINST	SUBSTRATE	SPECIES	FORMAT SLIDES x FIELDS
FA 1156-1005-50 FA 1156-1010-50	Myelin oligodendrocyte glycoprotein (MOG)	transfected cells control transfection	EU 90	10 x 05 (050) 10 x 10 (100)

Indication: This test kit provides qualitative or semiquantitative in vitro determination of human antibodies of immunoglobulin class IgG against myelin oligodendrocyte glycoprotein (MOG) in patient samples to support the diagnosis of demyelinating diseases of the central nervous system. The fluorescence is either evaluated using the fluorescence microscope (specifications see chapter "Incubation", section "Evaluation") or, following automated image recording by the EUROPattern microscope at the computer screen, optionally supported by the EUROPattern Classifier software. The product is designed for use as IVD.

LOT	Lot description	CE		Storage temperature
IVD	In vitro diagnostic medical device			Unopened usable until

Comparative Study > Neurology. 2019 Mar 12;92(11):e1250-e1255.
doi: 10.1212/WNL.0000000000007096. Epub 2019 Feb 6.

A multicenter comparison of MOG-IgG cell-based assays

Patrick J Waters [1], Lars Komorowski [1], Mark Woodhall [1], Sabine Lederer [1], Masoud Majed [1], Jim Fryer [1], John Mills [1], Eoin P Flanagan [1], Sarosh R Irani [1], Amy C Kunchok [1], Andrew McKeon [1], Sean J Pittock [2]

Affiliations + expand

PMID: 30728305 PMCID: PMC6511109 DOI: 10.1212/WNL.0000000000007096

Abstract

Objectives: To compares 3 different myelin oligodendrocyte glycoprotein-immunoglobulin G (IgG) cell-based assays (CBAs) from 3 international centers.

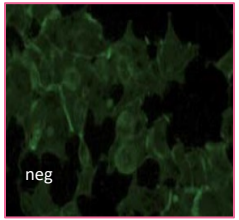
Methods: Serum samples from 394 patients were as follows: acute disseminated encephalomyelitis (28), seronegative neuromyelitis optica (27), optic neuritis (21 single, 2 relapsing), and longitudinally extensive (10 single, 3 recurrent). The control samples were from patients with multiple sclerosis (244), hypergammaglobulinemia (42), and other (17). Seropositivity was determined by visual observation on a fluorescence microscope (Euroimmun fixed CBA, Oxford live cell CBA) or flow cytometry (Mayo live cell fluorescence-activated cell sorting assay).

Results: Of 25 samples positive by any methodology, 21 were concordant on all 3 assays, 2 were positive at Oxford and Euroimmun, and 2 were positive only at Oxford. Euroimmun, Mayo, and Oxford results were as follows: clinical specificity 98.1%, 99.6%, and 100%; positive predictive values (PPVs) 82.1%, 95.5%, and 100%; and negative predictive values 79.0%, 78.8%, and 79.8%. Of 5 false-positives, 1 was positive at both Euroimmun and Mayo and 4 were positive at Euroimmun alone.

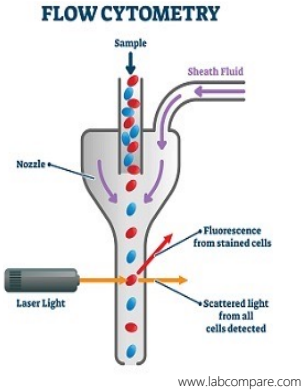
Conclusions: Overall, a high degree of agreement was observed across 3 different MOG-IgG CBAs. Both live cell-based methodologies had superior PPVs to the fixed cell assays, indicating that positive results in these assays are more reliable indicators of MOG autoimmune spectrum disorders.

Anti-MOG
CBA

Fixed cells
(IFT)



www.euroimmun.com



www.labcompare.com

Live cells
(Flowcytometry)

		IFT			
		neg	w pos	pos	total
flow	neg	10	0	1	11
	w pos	5	0	0	5
	pos	1	5	3	9
	total	16	5	4	25

Pos IFT = 36%
Pos flow = 64%

Is more sensitive less specific?

	MOGAD	Not MOGAD
Anti-MOG positive	14	3
Anti MOG weakly positive	15	9
Anti MOG negative	8	132
TOTAL	37	144

Sensitivity: 29/37 = 78%
Specificity: 132/144 = 92%
Likelihood ratio: 9x more likely to have MOGAD with a positive test outcome (positive or weakly positive).

C-activation marker

Adapt dilution or not?

MicroVue™ Bb Plus EIA

The MicroVue Bb Plus EIA kit measures the amount of the complement fragment Bb in human plasma or serum.

Product Specifications		Ordering Information	
Citations	45	For In Vitro Diagnostic Use.	
Specimen	Serum 25 µL, EDTA Plasma 50 µL	**Catalog Number**	A027

Specimen Dilution

Caution: Treat all specimens as if potentially infectious. Do not use heat-inactivated or contaminated specimens.

It is recommended that plasma samples be diluted 1:10 in Specimen Diluent for use in the MicroVue Bb Plus EIA. It is recommended that serum samples be diluted 1:20 in Specimen Diluent. Once diluted, the specimens must be added to the microassay wells within 30 minutes. Do not store or re-use diluted specimens. Any remaining specimens should be discarded.

	Recovery (%)				
dilution	**HC**	**neg ctr**	**spike**	**mid ctr**	**high ctr**
1,125	45	38	nd	nd	nd
2,5	71	60	nd	nd	nd
5	79	78	75	Out of range	Out of range
10	100	100	100	Out of range	Out of range
20	Out of range	Out of range	118	Out of range	Out of range
40	Out of range	Out of range	133	121	Out of range
80	Out of range	Out of range	Out of range	107	Out of range
160	Out of range	Out of range	Out of range	103	100

Conclusion

- Following the kit insert may result in incorrect outcomes
- Check the *intended use / purpose / clinical context* and use the test like that
- Document if you deviate and based on what data
- For many tests IVDR is only to be followed from May 2027

Enter these discussions with the client!

Sanquin

For Life.