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Changes: §1, §2, §4, §5, §6, §8, §9, §12, §14, §15.3, References;
Deletions: -

LIAISON® VZV IgM (REF 310860)

1. INTENDED PURPOSE

The LIAISON® VZV IgM assay uses chemiluminescent immunoassay (CLIA) technology for the in vitro qualitative determination of specific IgM antibodies to varicella-zoster virus (VZV) in human serum and plasma samples. This assay is intended as an aid in the determination of immune status to VZV and as an aid in the diagnosis of infection of varicella-zoster virus in individuals including pregnant women.
The test has to be performed on the LIAISON® Analyzer family*.

2. SUMMARY AND EXPLANATION OF THE TEST

Varicella-zoster virus (VZV) is the etiological agent of varicella, also known as chickenpox, and herpes zoster, also known as shingles[1]. Varicella is the manifestation of a primary VZV infection in patients without prior exposure, usually in childhood[2,3]. VZV is a neurotropic human restricted alpha-herpes virus of the Varicellovirus genus, of which the principal characteristic is the capacity for latency[2,4].

Primary VZV infection causes systemic disease (varicella/chickenpox) and induces specific antibody production and cell-mediated immunity. The main feature of varicella is the eruption of itchy vesicles, which after a week form crust. Symptoms such as fever, tiredness and loss of appetite are common[5,10, 13,14]. After the first infection, VZV reaches the sensory ganglia via retrograde axonal transport where it goes through a period of latency. During this latency period, there is no evidence that virus transmission can occur. Decreasing cellular immunity, either by age or by immunosuppression, may cause reactivation of the virus[2,5,8]. Reactivation of VZV results in an infection within the ganglia. This reactivation is denominated as herpes zoster (shingles), frequently complicated by postherpetic neuralgia.

After VZV infection, the first antibody to be produced is IgM, which appears 2 to 5 days after the lesions and the highest titer occurs at 2 to 3.5 weeks. IgM remains for a few months and then disappears. The second antibody to be developed is IgG which appears approximately 5 to 10 days after rash and persists for many years. Tests for anti-VZV IgG and IgM are used to diagnose the presence of the disease or the immunity status against this infection[18].

Diagnostic evaluation of VZV infection may be considered to check immune status and/or identify active infection, when the individual has atypical and/or severe symptoms and the physician wishes to distinguish between VZV infection and other causes. Diagnostic testing may also be performed prior to organ transplantation or when a child, pregnant woman or immunocompromised individual has been exposed to a patient with chickenpox[3,6,12,13].

Serological testing is also recommended for women of childbearing age without a history of varicella or vaccination to examine the value of possible vaccination before pregnancy[20].

Pregnant women who contract varicella are at higher risk for severe disease and, although rare, perinatally acquired VZV can cause significant morbidity and mortality in the newborn[9,13,15-17].

Organ transplant recipients and immunocompromised persons are at increased risk for VZV infection[3,6].

While IgM antibodies are a sign of active disease, IgG antibodies persist after an initial infection or vaccination. Therefore, only a documented increase in IgG levels is a sign of active disease[7,9,10,12,18].

IgG antibodies against VZV generally persist for life after a successful vaccination[7,8,12]. As a result, anti-VZV IgG antibody determination is the most common approach to determine a history of VZV infection or vaccination[7,9]. IgG levels do not quantitatively correlate with the level of immune protection or disease severity[4,7,8,19], but passive transfer of VZV-specific antibodies helps to prevent disease development after exposure to varicella which indicates that humoral immunity does contribute meaningfully to disease prevention[4,7].

3. PRINCIPLE OF THE PROCEDURE

The method for qualitative determination of specific IgM to varicella-zoster virus is an indirect chemiluminescence immunoassay (CLIA). Varicella-zoster virus antigen is used for coating magnetic particles (solid phase) and a mouse monoclonal antibody to human IgM is linked to an isoluminol derivative (isoluminol-antibody conjugate). Calibrators, samples and controls are diluted with buffer H, that contains goat IgG to human IgG as an absorbent reagent to curb interference from human IgG specific to VZV or from rheumatoid factor. During the first incubation, varicella-zoster virus antibodies present in calibrators, samples or controls bind to the solid phase. During the second incubation, the antibody conjugate reacts with varicella-zoster virus IgM already bound to the solid phase. After each incubation, the unbound material is removed with a wash cycle.
Subsequently, the starter reagents are added and a flash chemiluminescence reaction is thus induced. The light signal, and hence the amount of isoluminol-antibody conjugate, is measured by a photomultiplier as relative light units (RLU) and is indicative of varicella-zoster virus IgM concentration present in calibrators, samples or controls.

*(LIAISON®, LIAISON® XL, LIAISON® XS)

4. MATERIALS PROVIDED

Reagent integral

Magnetic particles (2.5 mL)	SORB	Magnetic particles (> 0.25% solid) coated with varicella-zoster virus antigen (partially purified extract of infected cell cultures, Ellen strain) (approx. 100 µg/mL), BSA, PBS buffer, < 0.1% sodium azide.
Calibrator 1 (0.7 mL)	CAL 1	Human serum/plasma containing low varicella-zoster virus IgM levels (approx. 0.4 Index), BSA, phosphate buffer, 0.2% ProClin® 300, an inert yellow dye. The calibrator concentrations are referenced to an in-house antibody preparation.
Calibrator 2 (0.7 mL)	CAL 2	Human serum/plasma containing high varicella-zoster virus IgM levels (approx. 1.5 Index), BSA, phosphate buffer, 0.2% ProClin® 300, an inert blue dye. The calibrator concentrations are referenced to an in-house antibody preparation.
Buffer H (28 mL)	BUF H	Goat IgG to human IgG (absorbent reagent) (≥5%), goat serum, BSA, phosphate buffer, 0.2% ProClin® 300, an inert blue dye.
Conjugate (23 mL)	CONJ	Mouse monoclonal antibodies to human IgM (minimum 10 ng/mL) conjugated to an isoluminol derivative, BSA, PBS buffer, 0.2% ProClin® 300, preservatives.
Number of tests		100

All reagents are supplied ready to use. The order of reagents reflects the layout of containers in the reagent integral.

Materials required but not provided (system related)

LIAISON® XL Analyzer	LIAISON® Analyzer
LIAISON® XL Cuvettes (REF X0016). LIAISON® XL Disposable Tips (REF X0015) or LIAISON® Disposable Tips (REF X0055). LIAISON® XL Starter Kit (REF 319200) or LIAISON® EASY Starter Kit (REF 319300). – – LIAISON® Wash/System Liquid (REF 319100). LIAISON® XL Waste Bags (REF X0025). LIAISON® XL Cleaning Tool (REF 310995) or LIAISON® EASY Cleaning Tool (REF 310996).	LIAISON® Module (REF 319130). – – LIAISON® Starter Kit (REF 319102) or LIAISON® XL Starter Kit (REF 319200) or LIAISON® EASY Starter Kit (REF 319300). LIAISON® Light Check 12 (REF 319150). LIAISON® Wash/System Liquid (REF 319100). LIAISON® Waste Bags (REF 450003). LIAISON® Cleaning Kit (REF 310990). –

LIAISON® XS Analyzer
LIAISON® Cuvettes on Tray (REF X0053). LIAISON® Disposable Tips (REF X0055). LIAISON® EASY Starter Kit (REF 319300). LIAISON® EASY Wash Buffer (REF 319301). LIAISON® EASY System Liquid (REF 319302). LIAISON® EASY Waste (REF X0054). LIAISON® EASY Cleaning Tool (REF 310996).

Additionally required materials

LIAISON® VZV IgM controls (negative and positive) (REF 310861).

5. WARNINGS AND PRECAUTIONS

For *in vitro* diagnostic use.

For Laboratory Professional Use Only.

Visually inspect the integral vials for leaking at the membrane seals or elsewhere. If the vials are found to be leaking, the local customer service should be notified immediately.

All serum and plasma units used to produce the components provided in this kit have been tested for the presence of HBsAg, anti-HCV, anti-HIV-1, anti-HIV-2 and found to be non-reactive. As, however, no test method can offer absolute assurance that pathogens are absent, all specimens of human origin should be considered potentially infectious and handled with care.

6. SAFETY PRECAUTIONS

Do not eat, drink, smoke or apply cosmetics in the assay laboratory.

Do not pipette by mouth.

Avoid direct contact with potentially infected material by wearing laboratory clothing, protective goggles, and disposable gloves. Wash hands thoroughly at the end of each assay.

Avoid splashing or forming an aerosol. All drops of biological reagent must be removed with a sodium hypochlorite solution with 0.5% active chlorine, and the means used must be treated as infected waste.

All samples and reagents containing biological materials used for the assay must be considered as potentially able to transmit infectious agents. The waste must be handled with care and disposed of in compliance with the laboratory guidelines and the statutory provisions in force in each Country.

Any materials for reuse must be appropriately sterilized in compliance with the local laws and guidelines. Check the effectiveness of the sterilization/decontamination cycle. The analyzers should be cleaned and decontaminated on a regular basis. See the Operator's Manual for the procedures.

Do not use kits or components beyond the expiration date given on the label.

Pursuant to EC Regulation 1272/2008 (CLP) hazardous reagents are classified and labeled as follows:

REAGENTS:	CAL 1, CAL 2, BUF H, CONJ
CLASSIFICATION:	Skin sens. 1 H317
SIGNAL WORD:	Warning
SYMBOLS / PICTOGRAMS:	 GHS07 Exclamation mark
HAZARD STATEMENTS:	H317 May cause an allergic skin reaction.
PRECAUTIONARY STATEMENTS:	P261 Avoid breathing dust/fume/gas/mist/vapours/spray. P280 Wear protective gloves/protective clothing/eye protection/face protection. P363 Wash contaminated clothing before reuse.
CONTAINS: (only substances prescribed pursuant to Article 18 of EC Regulation 1272/2008).	reaction mass of: 5-chloro-2-methyl-4-isothiazolin-3-one [EC no. 247-500-7] and 2-methyl-2H -isothiazol-3-one [EC no. 220-239-6] (3:1) (ProClin® 300).

Pursuant to EC Regulation 1272/2008 (CLP), SORB is labeled as EUH210 safety data sheets available on request.
For additional information see Safety Data Sheets available on www.diasorin.com.

7. PREPARATION OF REAGENT INTEGRAL

Please note the following important reagent handling precautions:

Resuspension of magnetic particles

Magnetic particles must be completely resuspended before the integral is placed on the instrument. Follow the steps below to ensure complete suspension:
Before the seal is removed, rotate the small wheel at the magnetic particle compartment until the colour of the suspension has changed to brown. Gentle and careful side-to-side mixing may assist in the suspension of the magnetic particles (avoid foam formation). Visually check the bottom of the magnetic particle vial to confirm that all settled magnetic particles have resuspended. Carefully wipe the surface of each septum to remove residual liquid.
Repeat as necessary until the magnetic particles are completely resuspended.

Foaming of reagents

In order to ensure optimal performance of the integral, foaming of reagents should be avoided. Adhere to the recommendation below to prevent this occurrence:
Visually inspect the reagents, calibrators in particular (position two and three following the magnetic particle vial), to ensure there is no foaming present before using the integral. If foam is present after resuspension of the magnetic particles, place the integral on the instrument and allow the foam to dissipate. The integral is ready to use once the foam has dissipated and the integral has remained onboard and mixing.

Loading of integral into the reagent area

LIAISON® Analyzer

– Place the integral into the reagent area of the analyzer with the bar code label facing left and let it stand for 30 minutes before using. The analyzer automatically stirs and completely resuspends the magnetic particles.
– Follow the analyzer operator's manual to load the specimens and start the run.

LIAISON® XL and LIAISON® XS analyzer

– LIAISON® XL and LIAISON® XS analyzer are equipped with a built-in solid-state magnetic device which aids in the dispersal of microparticles prior to placement of a reagent integral into the reagent area of the analyzer. Refer to the analyzer operator's manual for details.
  a. Insert the reagent integral into the dedicated slot.
  b. Allow the reagent integral to remain in the solid-state magnetic device for at least 30 seconds (up to several minutes). Repeat as necessary.
– Place the integral into the reagent area of the analyzer with the label facing left and let it stand for 15 minutes before using. The analyzer automatically stirs and completely resuspends the magnetic particles.
– Follow the analyzer operator's manual to load the specimens and start the run.

8. STORAGE AND STABILITY OF REAGENT INTEGRAL

Upon receipt, the reagent integral must be stored in an upright position to facilitate later resuspension of magnetic particles. When the reagent integral is stored sealed and kept upright, the reagents are stable at 2-8°C up to the expiry date. Do not freeze. The reagent integral should not be used past the expiry date indicated on the kit and reagent integral labels. After removing the seals, the reagent integral is stable for twelve (12) weeks refrigerated either at 2-8°C or on board the instrument.

9. SPECIMEN COLLECTION AND PREPARATION

The correct specimen type must be used in the assay. Following matrices have been tested and may be used:
– serum;
– heparin plasma;
– citrate plasma;
– EDTA plasma.

Blood should be collected aseptically by venipuncture and the serum or plasma separated from clot, red cells or gel separator, after centrifugation, carefully following the tube manufacturers' instructions and according to good laboratory practices.

Centrifugation conditions of collection tubes may vary depending on the manufacturer. A minimum of 1,000 g for 10 minutes is reported. Use of centrifugation conditions should be evaluated and validated by the laboratory.

Package and label specimens in compliance with applicable regulations covering the transport of clinical specimens and infectious substances.

Specimens may be shipped on dry ice (frozen), on wet ice (for 2°-8°C), following the sample storage limitations described below.

Uncontrolled transport conditions (in terms of temperature and time) can cause inaccurate analytical results. During validation studies, specimen collection tubes commercially available at the time of testing were used. Therefore not all collection tubes from all manufacturers have been evaluated. Blood collection devices from various manufacturers may contain substances which could affect the test results in some cases (Bowen et al., Clinical Biochemistry, 43, 4-25, 2010).

A dedicated study on storage limitations was performed on serum or plasma specimens removed from clot, red cells or gel separator. The following storage conditions showed no significant differences:
– room temperature storage should be avoided;
– 2°-8°C for seven (7) days, otherwise they should be aliquoted and stored deep-frozen (–20°C or below);
– Up to five freeze-thaw cycles, however multiple freeze thaw cycles should be avoided.

If samples are stored frozen, mix thawed samples well before testing. Further centrifugation of specimens removed from red cells, clot or gel separator (suggested between 3,000 and 10,000 g for 10 minutes) is recommended to guarantee the consistency of results whenever one of the following conditions is identified:
– Samples previously centrifuged and stored at 2°-8°C;
– Samples with particulate matter, fibrin, turbidity, lipaemia or erythrocyte debris;
– Samples frozen and thawed;
– Samples requiring repeat testing.

Specimens with a lipid layer on the top should be transferred into a secondary tube, taking care to transfer only the clarified material.

Grossly haemolyzed or lipaemic samples as well as samples containing particulate matter or exhibiting obvious microbial contamination should not be tested. Heat inactivation of the specimens may affect the test results. Check for and remove air bubbles before assaying.

The minimum volume required for a single determination is 170 µL of specimen (20 µL specimen + 150 µL dead volume).

10. CALIBRATION

Test of assay specific calibrators allows the detected relative light unit (RLU) values to adjust the assigned master curve. Each calibration solution allows five calibrations to be performed.

Recalibration in triplicate is mandatory whenever at least one of the following conditions occurs:
– A new lot of reagent integral or of Starter Kit is used.
– The previous calibration was performed more than eight weeks before.
– Control values lie outside the expected ranges.
– **LIAISON® and LIAISON® XL analyzers:** the analyzer has been serviced.
– **LIAISON® XS Analyzer**: after a technical intervention, only if required by the service procedure, as communicated by DiaSorin Technical support or representative.

LIAISON® Analyzer: Calibrator values are stored in the bar codes on the integral label.
LIAISON® XL Analyzer: Calibrator values are stored in the reagent integral Radio Frequency IDentification transponder (RFID Tag).
LIAISON® XS Analyzer: Calibrator values are stored in the reagent integral Radio Frequency IDentification transponder (RFID Tag).

11. ASSAY PROCEDURE

Strict adherence to the analyzer operator's manual ensures proper assay performance.

LIAISON® Analyzer. Each test parameter is identified via the bar codes on the reagent integral label. In the event that the barcode label cannot be read by the analyzer, the integral cannot be used. Do not discard the reagent integral; contact your local DiaSorin technical support for instruction.

LIAISON® XL and LIAISON® XS analyzers. Each test parameter is identified via information encoded in the reagent integral Radio Frequency IDentification transponder (RFID Tag). In the event that the RFID Tag cannot be read by the analyzer, the integral cannot be used. Do not discard the reagent integral; contact your local DiaSorin technical support for instruction.

The analyzer operations are as follows:

1. Dilute samples with buffer H.
2. Dispense calibrators, controls or specimens into the reaction module.
3. Dispense coated magnetic particles.
4. Dispense buffer H.
5. Incubate.
6. Wash with Wash/System liquid.
7. Dispense conjugate into the reaction module.
8. Incubate.
9. Wash with Wash/System liquid.
10. Add the Starter Kit and measure the light emitted.

Warning - *Maintenance with the LIAISON® XL Cleaning Tool (REF 310995) or LIAISON® EASY Cleaning Tool (REF 310996) must be performed (refer to pertinent instruction for use for details).*

12. QUALITY CONTROL

LIAISON® controls should be run in singlicate to monitor the assay performance. Quality control must be performed by running LIAISON® VZV IgM controls REF 310861

(a) at least once per day of use,
(b) whenever a new reagent integral is used,
(c) whenever the kit is calibrated,
(d) whenever a new lot of Starter Reagents is used,
(e) to assess adequacy of performance of the open integral in agreement with guidelines or requirements of local regulations or accredited organizations.

Control values must lie within the expected ranges: whenever one or both controls lie outside the expected ranges, calibration should be repeated and controls retested. If control values obtained after successful calibration lie repeatedly outside the predefined ranges, the test should be repeated using an unopened control vial. If control values lie outside the expected ranges, patient results must not be reported.

The performance of other controls should be evaluated for compatibility with this assay before they are used. Appropriate value ranges should then be established for quality control materials used.

13. INTERPRETATION OF RESULTS

The analyzer automatically calculates varicella-zoster virus IgM levels expressed as index value and grades the results. For details, refer to the analyzer operator's manual.
Calibrators and controls may give different RLU or dose results on LIAISON®, LIAISON® XL and LIAISON® XS, but patient results are equivalent.

The cut-off value discriminating between the presence and the absence of varicella-zoster virus IgM has an index value of 1.00. Sample results should be interpreted as follows:
Samples with varicella-zoster virus IgM levels below an index value of 1.00 should be graded *negative*.
Samples with varicella-zoster virus IgM levels equal to or above an index value of 1.00 should be graded *positive*.
Samples with varicella-zoster virus IgM levels ranging within ± 10% of the cut-off value should be retested in order to confirm the initial result. Samples which are repeatedly positive should be considered positive. Samples which are negative at the second test should be considered negative.

A positive result may indicate recent or reactivated infection. A negative result, however, does not always rule out acute varicella-zoster virus infection, because the patient may still be unable to synthesize varicella-zoster virus specific IgM in spite of the presence of primary infection or virus reactivation. If clinical exposure to varicella-zoster virus is suspected despite a negative finding, a second sample should be collected and tested no less than one week later.

14. LIMITATIONS OF THE PROCEDURE

- A skillful technique and strict adherence to the instructions are necessary to obtain reliable results. Bacterial contamination or heat inactivation of the specimens may affect the test results.
- Users should be aware of possible cross-reactions from IgM antibody to other *Herpesviridae* (e.g., HSV, EBV) arising from the presence of common herpes antigens or simultaneous expression of IgM to other members of the herpes virus family.
- The performance characteristics of the test in newborns have not been established. Results in immunosuppressed subjects should be interpreted with caution.
- Test results are reported qualitatively as positive or negative for the presence of varicella-zoster virus IgM. However, diagnosis of infectious diseases should not be established on the basis of a single test result, but should be determined in conjunction with clinical findings and other diagnostic procedures as well as in association with medical judgement.
- The level of protection in patients, especially those with low titers of varicella-zoster virus IgM, should be determined by a physician.
- Results obtained with LIAISON® VZV IgM assay may not be used interchangeably with values obtained with different manufacturers' assay methods.
- Integrals may not be exchanged between analyzer types (LIAISON®, LIAISON® XL and LIAISON® XS). Once an integral has been introduced to a particular analyzer type, it must always be used on that analyzer until it has been exhausted.

15. SPECIFIC PERFORMANCE CHARACTERISTICS

15.1. Analytical specificity

Analytical specificity may be defined as the ability of the assay to accurately detect specific analyte in the presence of potentially interfering factors in the sample matrix (e.g., anticoagulants, haemolysis, effects of sample treatment), or cross-reactive antibodies.

Interference. Controlled studies of potentially interfering substances or conditions showed that the assay performance was not affected by anticoagulants (citrate, EDTA, heparin), haemolysis (up to 1000 mg/dL haemoglobin), lipaemia (up to 3000 mg/dL triglycerides), bilirubinaemia (up to 20 mg/dL bilirubin), or by freeze-thaw cycles of samples.

Cross-reactions. 40 potentially cross-reactive samples were tested, positive for IgM antibodies to one or more etiologic agent(s). 12 samples were positive for *Borrelia burgdorferi* IgM, 5 samples were positive for rubella virus IgM, 3 samples were positive for EBV IgM, 3 samples were positive for measles virus IgM, 3 samples were positive for parvovirus B19 IgM, 2 samples were positive for hCMV IgM, 2 samples were positive for HSV-1/2 IgM, 2 samples were positive for *Toxoplasma gondii* IgM, one sample was positive for mumps virus IgM, 7 samples were positive for rheumatoid factor (anti-Fc immunoglobulin) antibodies. All potentially cross-reactive samples scored negative by LIAISON® VZV IgM test.

15.2. Precision with LIAISON® Analyzer

Different samples, containing different concentrations of specific analyte, were assayed to estimate repeatability and reproducibility of the assay (i.e., within- and between-assay variability). The variability shown in the tables below did not result in sample misclassification. The results refer to the groups of samples investigated and are not guaranteed specifications, as differences may exist between laboratories and locations.

Repeatability. Twenty replicates were performed in the same run to evaluate repeatability.

Repeatability	A	B	Negative control	Positive control
Number of determinations	20	20	20	20
Mean (index value)	0.556	1.22	0.158	1.44
Standard deviation	0.041	0.06	0.036	0.11
Coefficient of variation (%)	7.4	5.1	22.6	7.4
Min. value	0.421	1.06	0.106	1.22
Max. value	0.598	1.34	0.232	1.61

Reproducibility. Twenty replicates were performed in different days (one or two runs per day) to evaluate reproducibility.

Reproducibility	A	C	Negative control	Positive control
Number of determinations	20	20	20	20
Mean (index value)	0.517	1.53	0.162	1.67
Standard deviation	0.082	0.24	0.030	0.17
Coefficient of variation (%)	15.9	15.7	18.3	10.4
Min. value	0.349	1.12	0.111	1.29
Max. value	0.636	2.12	0.198	1.96

15.3. Precision with LIAISON® XL Analyzer

Different samples, containing different concentrations of specific analyte, were assayed to estimate repeatability and reproducibility of the assay (i.e., within- and between-assay variability). The variability shown in the tables below did not result in sample misclassification.

Repeatability. Twenty replicates were performed in the same run to evaluate repeatability.

Repeatability	1	2	Negative control	Positive control
Number of determinations	20	20	20	20
Mean (index value)	0.666	1.67	0.153	1.85
Standard deviation	0.068	0.14	0.008	0.14
Coefficient of variation (%)	10.2	8.2	4.9	7.3
Min. value	0.515	1.40	0.136	1.49
Max. value	0.758	1.91	0.169	2.07

Reproducibility. Twenty replicates were performed in different days (one or two runs per day) to evaluate reproducibility.

Reproducibility	1	2	Negative control	Positive control
Number of determinations	20	20	20	20
Mean (index value)	0.597	1.76	0.132	1.92
Standard deviation	0.060	0.23	0.016	0.17
Coefficient of variation (%)	9.9	13.0	11.9	8.7
Min. value	0.516	1.34	0.104	1.70
Max. value	0.718	2.13	0.157	2.23

Lot-to-Lot Reproducibility. Six samples tested in singleton on five different LIAISON® XL instruments on four different batches.

Reproducibility	LIAISON® VZV IgM (Code 310860) on LIAISON® XL							
Sample ID	A	B	C	D	E	F	Positive Control	*Negative Control
Mean (Index/RLUs)	0.131	0.209	0.299	0.621	1.06	1.58	1.60	5040
Inter-lot coefficient of variation (%)	8.5	13.9	14.8	14.6	8.2	7.6	6.2	44.4

15.4. Precision with LIAISON® XS Analyzer

A five day precision study was conducted on three LIAISON® XS Analyzers to verify the precision with the LIAISON® VZV IgM Assay. The CLSI document EP15-A3 was consulted in the preparation of the testing protocol.
A coded panel comprised of six (6) frozen samples was used for the study.
The samples could be prepared by pooling samples with similar title in order to represent negative, borderline and positive levels.
The LIAISON® Control VZV IgM set was also included in the five day study.
The coded panel was tested on three LIAISON® XS Analyzers, in six replicates in a single run per day, for 5 operative days.
The mean Index value, standard deviation, and coefficient of variation (%CV) of the results were computed for each of the tested specimens for each of the instruments and across instruments.

Repeatability. Ninety replicates were performed in the same test to evaluate repeatability. 6 serum samples containing different concentration of analyte and kit controls were assayed in 6 replicates per day, over 5 operating days, on 3 units and one reagent lot.

Repeatability	3	4	5	6	7	8	Negative control*	Positive control
Number of determinations	90	90	90	90	90	90	90	90
Mean (index value)	0.360	0.659	0.881	1.30	1.53	1.80	1823	1.39
Standard deviation	0.010	0.019	0.038	0.045	0.051	0.084	105	0.044
Coefficient of variation (%)	2.7	2.9	4.4	3.5	3.3	4.6	5.8	3.2
Min. value	0.297	0.554	0.650	1.14	1.22	1.46	1014	1.13
Max. value	0.391	0.725	0.983	1.44	1.70	2.05	2365	1.56

*Negative control is expressed in RLU because out of the Assay Range

Reproducibility. Ninety replicates were performed in different days (one run per day) to evaluate reproducibility. 6 serum samples containing different concentration of analyte and kit controls were assayed in 6 replicates per day, over 5 operating days, on 3 units and one reagent lot.

Reproducibility	3	4	5	6	7	8	Negative control*	Positive control
Number of determinations	90	90	90	90	90	90	90	90
Mean (index value)	0.360	0.659	0.881	1.30	1.53	1.80	1823	1.39
Standard deviation	0.011	0.024	0.043	0.053	0.073	0.121	149	0.054
Coefficient of variation (%)	3.2	3.7	4.9	4.1	4.7	6.7	8.2	3.9
Min. value	0.297	0.554	0.650	1.14	1.22	1.46	1014	1.13
Max. value	0.391	0.725	0.983	1.44	1.70	2.05	2365	1.56

*Negative control is expressed in RLU because out of the Assay Range

15.5. Diagnostic specificity and sensitivity

A total of 282 samples were tested for the presence of VZV IgM antibodies using LIAISON® VZV IgM test and three CE-marked reference methods. Specimens used for the study included samples from different populations (pregnant women, blood donors, transplant recipients) as well as selected positive samples.
Concordant results obtained with two out of three reference methods were defined as consensus (32 positive results and 246 negative results); for four results consensus was not achieved. Three equivocal results by LIAISON® VZV IgM test were graded positive by consensus.
Diagnostic specificity and sensitivity were calculated upon exclusion of results for which consensus was not achieved.
Diagnostic specificity: 100% (246/246) - 95% confidence interval: 98.51-100%.
Diagnostic sensitivity upon exclusion of equivocal results: 81.25% (26/32) - 95% confidence interval: 63.57-92.80%.
Diagnostic sensitivity upon inclusion of equivocal results (considered as reactive): 90.63% (29/32) - 95% confidence interval: 74.99-98.02%.

A Summary of safety and performance is available on EUDAMED.

For EU only: please be aware that any serious incident that has occurred in relation to this IVD medical device should be reported to DiaSorin Italia S.p.A. and to the Competent Authority of the EU Member State in which the user and/or the patient is established.

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