ACCESS®
Immunoassay System

HIV combo
REF **A59428**

Σ 2 x 50

For the qualitative detection of HIV p24 antigen and antibodies to HIV-1/O/2 in human serum and plasma using the Access Immunoassay Systems.

ACCESS®
Immunoassay System

HIV combo Calibrators
REF **A59429**

The Access HIV combo Calibrators are intended to calibrate the Access HIV combo assay using the Access Immunoassay Systems.

ACCESS®
Immunoassay System

HIV combo QC
REF **A59430**

For monitoring the system performance of the Access HIV combo assay.

IVD CE 0459

A97207E - [EN] - 2017/03

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For the qualitative detection of HIV p24 antigen and antibodies to HIV-1/O/2 in human serum and plasma using the Access Immunoassay Systems.

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A97207E - [EN] - 2017/03

1 Intended Use

The Access HIV combo assay is a paramagnetic-particle, chemiluminescent immunoassay for the qualitative detection of HIV-1 p24 antigen and antibodies to HIV-1 (groups M and O) and HIV-2 in human serum and plasma (Li heparin, K2-EDTA, K3-EDTA and CPDA-1), using the Access Immunoassay Systems. The Access HIV combo assay is intended to be used as an aid in the diagnosis of HIV-1 or HIV-2 infection and as screening test for blood and plasma donors. The assay is not intended for the testing or screening of pooled specimens. An Access HIV combo assay result does not distinguish between the detection of HIV-1 p24 antigen, HIV-1 or HIV-1-O or HIV-2 antibodies.

2 Summary and Explanation of the Test

Acquired Immunodeficiency Syndrome (AIDS) is a virus-induced infectious disease expressed by a deep cellular immunity deficiency. Two types of viruses related to the lentivirus group were isolated from lymphocytes of patients with AIDS or its early syndromes[1,2,3].

The first virus called HIV-1 (Human Immunodeficiency Virus) was initially isolated in France and subsequently then in the USA. The second virus called HIV-2 was identified in two patients of African origin and found to be the origin of a new AIDS focus in West Africa[3,4,5,6].

The knowledge of the genetic variability of HIV strains was gained from the sequencing of the GAG, POL and ENV genes of representative strains for each subtype[7].

A phylogenetic analysis enabled different groups of HIV-1 to be distinguished: group M (Major), group N (non-M, non-O), group O (Outlier) and group P[8,9,10,11,12,13].

The group M of the HIV-1 includes 9 subtypes (A, B, C, D, F, G, H, J and K)[11] and circulating recombinant forms (CRFs)[11,14]. The geographic distribution of the various subtypes is now fairly well defined[15,16]. Some HIV-1 variants have only 70% homology for GAG and POL genes with the main isolates, and only 50% for the ENV gene. These differences may account for the failure to diagnose the disease in some patients[17]. The various HIV-2 strains show common antigenic features with the simian immunodeficiency virus SIV, whichever viral protein is considered (envelope and core proteins; heterology: 30%). They show less than 40% homology with the envelope proteins of HIV-1[3,18,19,20].

However, HIV-2 is less pathogenic than HIV-1, shows slower progression to disease, lower viral titers, and lower rates of vertical and horizontal transmission[21,22,23,24].

HIV antigens and antibodies appear and are detectable at different stages of the infection[25,26,27].

Current diagnosis of HIV infection requires the detection of anti-HIV serum antibodies using an ELISA method[28,29,30]. However, there is a mean period of 3 weeks between exposure and the appearance of the first antibodies. During this period, p24 antigen may be detected in most people infected by HIV-1, whatever their geographical origin[31,32]. The Access HIV combo assay allows the simultaneous detection of both HIV-1 and HIV-2 antibodies. This assay also uses monoclonal antibodies in the reagents to detect HIV-1 p24 antigen prior to seroconversion, thereby decreasing the seroconversion window and improving early detection of HIV infection[33,34,35,36].

3 Principles of The Procedure

The Access HIV combo assay is a sequential two-step immunoenzymatic (“sandwich”) assay.

In the first test step, sample, coated paramagnetic particles, biotinylated monoclonal antibodies to p24 and particle additive are combined. The paramagnetic particles are coated with recombinant HIV-1 protein, HIV-1-O / HIV-2 polypeptides, and monoclonal antibodies against HIV-1 p24 antigen.

After incubation in a reaction vessel, materials bound to the solid phase are held in a magnetic field while unbound materials are washed away.

In the second test step, 3 polypeptides and streptavidin labelled with alkaline phosphatase and also conjugate additive are then added.

After incubation, the unbound reagents are removed by separation in a magnetic field and by washing.

A chemiluminescent substrate Lumi-Phos 530 is added to the vessel and light generated by the reaction is measured with a luminometer. The light production is a function of the amount of enzyme conjugate present at the end of the reaction. The light quantity measured for a sample allows a determination of the presence of anti-HIV-1, or HIV-2 antibodies and/or antigen p24, by comparison to a cut-off value defined during the assay calibration on the instrument. If the light production is equal to or greater than the cut-off value, the sample is considered reactive in the Access HIV combo assay.

4 Product Information

4.1 Description

Access HIV combo Reagent Packs

Identification on label	Description	Presentation/ preparation A59428
R1a Paramagnetic particles	**Paramagnetic particles:** coated with recombinant HIV-1 protein (gp 160), HIV-1-O (gp 41) and HIV-2 (gp 36) polypeptides and monoclonal antibodies to p24 HIV-1 antigen, suspended in TRIS buffered saline, with 0.1% sodium azide and ProClin 300 (0.25%).	2 x 50 tests Ready to use
R1b Conjugate additive	**Conjugate additive:** TRIS buffered saline, with 0.1% sodium azide and ProClin 300 (0.25%).	
R1c Particle additive	**Particle additive:** TRIS buffer saline with biotinylated monoclonal antibodies to p24 HIV-1, with 0.1% sodium azide and ProClin 300 (0.25%).	
R1d Conjugates	**Conjugates:** HIV-1, HIV-1-O, HIV-2 polypeptides and streptavidin conjugated with alkaline phosphatase, with 0.1% sodium azide and ProClin 300 (0.25%).	

4.2 Storage and Handling Conditions

- Store upright and refrigerate at 2 to 10°C.
- Refrigerate at 2 to 10°C for a minimum of two hours before use on the instrument.
- Stable until the expiration date stated on the label when stored at 2 to 10°C (reagent pack unopened).
- Mix the new, unpunctured packs by gently inverting them until the particles are in solution and no longer adhere to the seal or sides of the well. Do not invert packs that have been punctured.
- Stable at 2 to 10°C for 56 days on board after initial use.
- Signs of possible deterioration are a broken elastomeric layer on the pack or control values out of range.
- If the reagent pack is damaged (i.e. broken elastomer), discard the pack.

5 Warnings and Precautions

- For *in vitro* diagnostic use. For healthcare professional use only.

5.1 Health and Safety Precautions

- This test kit should only be handled by qualified personnel trained in laboratory procedures and familiar with their potential hazards. Wear appropriate protective clothing, gloves and eye/face protection and handle appropriately in accordance with Good Laboratory Practices.
- Biological spills: human source material spills should be treated as potentially infectious. Spills not containing acid should be immediately decontaminated, including the spill area, materials and any contaminated surfaces or equipment and with appropriate chemical disinfectant that is effective on the potential biohazards of the samples in question (commonly a 1:10 dilution of household bleach, 70-80% ethanol or isopropanol, an iodophor such as 0.5% Wescodyne™ Plus, etc.) and wiped dry. Spills containing acid should be appropriately absorbed (wiped up) or neutralized, the area flushed with water and wiped dry; materials used to absorb the spill may require hazardous waste disposal. The area should subsequently decontaminated with one of the chemical disinfectants.
- Dispose of all specimens and material used to perform the test as though they contain an infectious agent. Laboratory, chemical or bio-hazardous waste must be handled and discarded in accordance with all local, regional and national regulations.
- For hazard and precaution recommendations related to any chemical components in this test kit, please refer to the pictogram(s) featured on the labels and the information supplied in the section 5.2. The Safety Data Sheet (SDS) is available at www.bio-rad.com.

5.2 Precautions Related to the Procedure

Warning:

H317: May cause an allergic skin reaction.
P280: Wear protective gloves/protective clothing/eye protection/face protection.
P333+P313: If skin irritation or rash occurs: Get medical advice/attention.
P302+P352: If on skin: Wash with plenty of soap and water.
P501: Dispose of contents/container in accordance with local/regional/national/international regulations.

6 Specimens

1. Serum (including serum separator tubes) and plasma (Li Heparin including plasma separator tubes, K2-EDTA, K3-EDTA and CPDA-1) are the recommended samples.
2. **Do not heat the samples.**
3. Observe the following recommendations for handling, processing, and storing blood samples[37]:
   - Collect all blood samples observing routine precautions for venipuncture.
   - Allow serum samples to clot completely before centrifugation.
   - Ensure that all residual fibrin and cellular matter has been removed prior to analysis.
   - Follow blood collection tube manufacturer's recommendations for centrifugation.
   - Keep tubes tightly stoppered at all times.
   - Store samples at room temperature (15 to 23°C) for no longer than twenty-four hours.
   - If the assay is not completed within twenty-four hours, refrigerate the samples at 2 to 8°C.
   - If the assay is not completed within 8 days at 2 to 8°C, or for shipment of samples, freeze at -20°C or below.
   - Thaw samples no more than 3 times. A study of 25 fresh non-reactive sera and 25 fresh reactive sera exhibited no clinically significant dose changes after three freeze-thaw cycles.
   - After thawing, the sample must be thoroughly mixed, centrifuged again at 3,000 g for 10 minutes and transferred into a cup in order to remove any suspended fibrin particles or aggregates liable to yield false-positive results.
4. Each laboratory should determine the acceptability of its own blood collection tubes and serum separation products. Variations in these products may exist between manufacturers and, occasionally, from lot to lot.

7 Procedure

7.1 Material Required

7.1.1 Materials Provided

R1 Access HIV combo Reagent Packs

7.1.2 Materials Required but Not Provided

1. Access HIV combo Calibrators
   Provided as one HIV-Ab negative serum and one anti-HIV-1 Ab positive serum
   Cat. No. A59429
2. Quality control materials:
   - Access HIV combo QC, provided as one HIV-Ab negative serum, one anti-HIV-1 positive serum and one HIV-1 antigen positive in Tris Buffer

   Cat. No. A59430
   - Access HIV combo QC4 & QC5, provided as one anti-HIV-2 antibodies positive serum and one anti-HIV-1-O antibodies positive serum in human negative serum.

   Cat. No. B22822
   - Other commercial control sera
3. Access Substrate
   Cat. No. 81906

4. Access 2:
   Wash buffer: Access Wash Buffer II, Cat. No. A16792
5. UniCel® DxI®:
   Wash buffer: UniCel DxI Wash Buffer II, Cat. No. 16793
6. Systems:
   Access 2, UniCel DxI (UniCel DxI 600, UniCel DxI 800, UniCel DxC 880i, UniCel DxC 860i, UniCel DxC 680i, UniCel DxC 660i).

7.2 Assay Procedure

1. Refer to the appropriate system manuals and/or Help system for a detailed description of installation, start-up, principles of operation, system performance characteristics, operating instructions, calibration procedures, operational limitations and precautions, hazards, maintenance and troubleshooting.
2. Mix contents of new (unpunctured) reagent packs by gently inverting pack several times before loading on the instrument. Do not invert open (punctured) packs.
3. One hundred ten (110) µL of sample is used for each determination (in addition to dead volume).
4. Time to first result is approximately 60 minutes.
5. The system default unit of measure for sample results is the Signal/Cut-off (S/CO) ratio.

7.3 Calibration

An active calibration point is required for all tests. For the Access HIV combo assay, calibration is required every 56 days. Consequently, for the Access HIV combo assay, calibration is required every 56 days using C0 and C1 from the Access HIV combo Calibrators kit.
Refer to the appropriate system manuals and/or Help system for information on calibration theory, configuring calibrators, entering calibrator test requests and reviewing calibration data.

7.4 Quality Control

Quality control materials simulate the characteristics of patient samples and are essential for monitoring the system performance of immunochemical assays. Quality control is recommended at least, every 24 hours[38] and on system start-up prior to running patient samples. Include Access HIV combo QC and Access HIV combo QC4 & QC5 kits or other commercially available quality control materials that cover at least two levels of analyte. More frequent use of these controls or the use of additional controls is left to the discretion of the user based on good laboratory practices or laboratory accreditation requirements and applicable laws. Follow the manufacturer's instructions for reconstitution and storage. Each laboratory should establish mean values and acceptable ranges to ensure proper performance. Quality control results that do not fall within acceptable ranges may indicate invalid test results. Examine all test results generated since obtaining the last acceptable quality control test point for this analyte. The Access HIV combo assay has been evaluated at a room temperature range of 18-32°C. For optimal results, assay calibration and patient sample testing should be conducted under similar temperature conditions.
If ambient laboratory temperature varies by more than ± 5°C from the temperature of calibration, review quality control results and recalibrate as necessary. Refer to the appropriate system manuals and/or Help system for complete information about reviewing quality control results.
All manufactured and commercialized reagents are under subject to a comprehensive quality system starting from the reception of raw materials right up to the ultimate commercialization of the product.
Each lot is submitted to a quality control and is only released onto the market if it conforms to the acceptance criteria.

7.5 Calculation / Interpretation of the Results

Patient test results are calculated automatically by the system software using the cut-off value determined by active calibration. Results (Signal/Cut-Off = S/CO) are reported to be "reactive" or "non-reactive" as a function of their relationship with the "cut-off" (signal greater than or signal equal to or less than the cut-off value). However, results ~10% lower than the "cut-off value" should be cautiously interpreted and retested in duplicate. This recommended gray zone (from 0.9 to less than 1.0) should be stored by the user in the system software (refer to the appropriate system manuals and/or Help system for complete instructions on gray zone for a qualitative assay). In this way a distinctive mark will automatically be reported, enabling rapid identification of a result situated in the gray zone. Patient test results can be reviewed using the Sample Results screen. Refer to the appropriate system manuals and/or Help system for complete instructions on reviewing results.

First result analysis:

- Any sample with a ratio (S/CO) lower than 0.9 is considered to be non-reactive with the Access HIV combo assay.
- Samples with a ratio (S/CO) ≥ 0.9 and < 1 are in the gray zone and should be retested in duplicate before final interpretation.
- Samples with a ratio (S/CO) greater than or equal to 1, are initially considered to be reactive with the Access HIV combo and such samples should be retested in duplicate before final interpretation.

Second result analysis:

All samples that were initially reactive or in the gray zone should be retested in duplicate using the Access HIV combo assay:

- If the results of the duplicates are < 1.0 S/CO, the sample must be considered non-reactive (negative) for the Access HIV combo assay.
- If one of the 2 results is ≥ 1.0 S/CO, the initial result is repeatable and the sample is declared as "reactive" for the Access HIV combo assay.

However, in accordance with local regulations, it is necessary to analyze any "reactive" sample by supplementary tests, including at least a confirmatory method to clearly establish the positive result.

Table 1: Access HIV combo result interpretation

Result Ratio: Signal/Cut-Off			Interpretation	Supplementary tests
First Result Analysis	S/CO < 0.9	Non-reactive	HIV-1 p24 and/or HIV-1/HIV-1-O/HIV-2 Ab not detected	NA
	S/CO ≥ 1.0	Reactive	"Initial Reactive"	Retest in duplicate
	0.9 ≤ S/CO < 1.0	Gray zone	"Initial Reactive"	Retest in duplicate
Second Result Analysis	Retest in duplicate: if the 2 results are < 1.0	Non-reactive	HIV-1 p24 and/or HIV-1/HIV-1-O/HIV-2 Ab not detected	NA
	Retest in duplicate: if one of the 2 results is ≥ 1.0	Reactive	HIV-1 p24 and/or HIV-1/HIV-1-O/HIV-2 Ab detected "Repeat Reactive"	Confirmatory test

8 Test Limitations

1. The Access HIV combo assay is strictly limited to the detection of HIV-1 antigen and HIV-1/HIV-1-O/HIV-2 antibodies in human serum or plasma (Li heparin, K2-EDTA, K3-EDTA and CPDA-1). The performance characteristics using other sample types have not been established or are limited.
2. The Access HIV combo results should be interpreted in light of the total clinical presentation of the patient, including: clinical history, data from additional tests and other appropriate information.
3. For assays employing antibodies, the possibility exists for interference by heterophile antibodies in the patient sample. Patients who have been regularly exposed to animals or have received immunotherapy or diagnostic procedures utilizing immunoglobulins or immunoglobulin fragments may produce antibodies, e.g. HAMA, that interfere with immunoassays. Additionally, other heterophile antibodies such as human anti-goat antibodies may be present in patient samples[39,40]. Such interfering antibodies may cause erroneous results. Carefully evaluate the results of patients suspected of having these antibodies.
4. Transplant patient samples have to be tested before freezing.
5. Performance has not been established using cadaver samples or body fluids other than human serum and plasma.
6. The magnitude of the measured result, above the cut-off, is not indicative of the total amount of antibody and/or antigen present.
7. A non-reactive result indicates that the tested sample contains no antigen and no antibodies detectable with Access HIV combo assay. This does not preclude the possibility of infection by HIV-1 and/or HIV-2.
8. For an infection to be declared, a reactive result obtained with the Access HIV combo assay should be confirmed by an appropriate method.

9. Immunocompromised individuals and conditions such as severe infection and immunosuppressive drug therapy can result in the suppression of antibody levels below the detection threshold of the assay. Results obtained with such samples should be interpreted with caution.

9 Performance Characteristics

9.1 Precision Measurement

The precision of the Access HIV combo assay was determined by the analysis of 13 samples: 1 negative sample, 1 low-positive sample (Low 1), 1 sample close to cut-off (low 2), 1 medium-positive sample for HIV-1, HIV-2, HIV-1-O and HIV Ag.
The repeatability was assessed by testing these 13 samples in one run with 30 replicates on 1 system. The CVs were determined.
The intermediate precision was assessed by testing these 13 samples on 1 lot, in duplicate, in 2 different runs per day (am and pm), and by two operators for a period of 20 days.
The inter-lot precision was assessed by testing these 13 samples in 5 replicates with 4 different lots using 4 different calibrator lots. The results are shown in the tables below:

9.1.1 Repeatability

N=30		Mean (signal/cut-off)	% C.V.
Negative samples		0.28	10.6
Low 1 samples	**HIV-1**	2.19	4.1
	HIV-2	2.20	4.7
	HIV-1-O	1.91	2.6
	HIV-1-Ag	2.40	5.0
Low 2 samples	**HIV-1**	0.96	5.9
	HIV-2	0.95	4.4
	HIV-1-O	1.16	4.6
	HIV-1-Ag	1.20	4.6
Medium 1 samples	**HIV-1**	2.86	5.8
	HIV-2	3.81	3.4
	HIV-1-O	3.34	4.2
	HIV-1-Ag	3.30	3.7

The coefficients of variation for samples are less than 12%.

9.1.2 Intermediate Precision

N=80		Mean (signal/cut-off)	% C.V.
Negative samples		0.30	10.1
Low 1 samples	**HIV-1**	2.35	5.6
	HIV-2	2.37	5.1
	HIV-1-O	1.88	4.6
	HIV-1-Ag	2.35	7.6
Low 2 samples	**HIV-1**	1.02	5.6
	HIV-2	1.03	5.6
	HIV-1-O	1.15	4.9
	HIV-1-Ag	1.17	4.9
Medium 1 samples	**HIV-1**	3.04	5.1
	HIV-2	3.99	4.9
	HIV-1-O	3.23	4.6
	HIV-1-Ag	3.12	4.7

The coefficients of variation for samples are less than 12%.

9.1.3 Inter-lot Precision

N=20		Inter Cal % C.V.	Inter RP % C.V.	Total % C.V.
Negative samples		12.1	12.3	15.0
Low 1 samples	**HIV-1**	11.0	7.4	11.4
	HIV-2	9.8	9.0	12.4
	HIV-1-O	10.2	6.5	10.8
	HIV-1-Ag	8.3	7.0	9.5
Low 2 samples	**HIV-1**	10.3	6.2	10.7
	HIV-2	10.3	7.2	11.3
	HIV-1-O	10.2	5.5	10.3
	HIV-1-Ag	10.4	14.8	16.9
Medium 1 samples	**HIV-1**	9.8	5.7	10.4
	HIV-2	10.2	11.0	13.9
	HIV-1-O	8.5	10.4	12.1
	HIV-1-Ag	11.0	13.0	15.5

The coefficients of variation for samples are less than 20%.

9.2 Diagnostic Performance

9.2.1 Diagnostic Specificity

The specificity of the Access HIV combo assay demonstrated a specificity ≥ 99.5%. This specificity was investigated by testing the following samples:

Sample Type	IR specificity			RR specificity		
	n	%	95% Confidence Interval	n	%	95% Confidence Interval
Blood donors	7656/7664	99.90	[99.79-99.95%]	7664/7664	100.00	[99.95-100%]
Selected hospitalized patients	1961/1969	99.59	[99.20-99.82%]	1966/1969	99.85	[99.56-99.97%]
Not selected hospitalized patients	1121/1122	99.91	[99.50-100%]	1121/1122	99.91	[99.50-100%]
Pregnant women	200/200	100.00	[98.17-100%]	200/200	100.00	[98.17-100%]
Overall mean	10938/10955	**99.84**	[99.75-99.91%]	10951/10955	**99.96**	[99.91-99.99%]

9.2.2 Diagnostic Sensitivity

Sensitivity studies with Access HIV combo were performed by testing confirmed HIV Ab samples, specimens from acute infected patients, commercial seroconversion panels and HIV Ag samples (neat or diluted).

Clinical sensitivity

Confirmed HIV Ab positive samples

The HIV-1 sensitivity was investigated on 674 confirmed positive samples and found equal to **100%** (95% CI: 99.45 – 100%).

255 samples were tested including genotyped subtypes and variants:
- Group M (236): A(21), B(56), C(17), D(13), F(10), G(15), H(7), J(3), K(1), CRF: 01(13) - 02(44) - 05(1) - 06(7) - 08(1) - 09(5) - 10(1) - 11(5) - 12(1) - 13(2) - 14(6) - 15(3) - 19(3) - 27(1)
- Group O (17)
- Group N (2)

The HIV-2 sensitivity was evaluated by testing 126 well-documented samples and declared equal to **100%** (95% CI: 97.11-100%).

Specimens from acute infected patients and from commercial seroconversion panels

- The HIV-1 sensitivity on preseroconversion and perseroconversion was investigated on 86 specimens.
- Seroconversion sensitivity of the Access HIV combo assay was evaluated by testing sequential specimens from 61 well–documented commercial HIV seroconversion panels (with 131 early seroconversion samples).

  Table 2 shows results from 6 seroconversion panels.

Table 2: Seroconversion panels

Panel	Sample ID	Days after 1st bleed	Access® HIV combo (S/CO)	PCR*	Western Blot*
BBI 9012	9012-05	14	0.53	Positive	Negative
	9012-06	16	1.21	Positive	Negative
	9012-07	21	25.36	Positive	Negative
BBI 9017	9017-04	10	0.32	Positive	Positive
	9017-06	13	1.19	Positive	Positive
	9017-07	17	3.48	Positive	Positive
	9017-08	20	4.15	Positive	Positive
	9017-09	24	2.44	Positive	Positive
	9017-10	28	5.67	Positive	Positive
	9017-11	31	42.27	Positive	Positive
BBI 9022	9022-07	23	0.77	Positive	Negative
	9022-08	25	5.81	Positive	Negative
	9022-09	32	161.31	Positive	Negative
PRB 950	PRB950-01	0	0.29	Negative	Positive
	PRB950-02	18	1.12	Positive	Positive
	PRB950-03	21	8.03	Positive	Positive
	PRB950-04	28	21.15	Positive	Negative
BBI 9034	9034-10	42	0.28	Negative	Negative
	9034-11	47	1.75	Positive	Negative
	9034-12	51	20.47	Positive	Negative
Zeptometrix 6243	6243-06	20	0.37	Positive	Indeterminate
	6243-07	25	1.37	Positive	Indeterminate
	6243-08	27	1.89	Positive	Indeterminate
	6243-09	30	6.68	Positive	Indeterminate
	6243-10	32	18.06	Positive	Indeterminate

* Data from the vendors.

HIV-1 Antigen samples
Sensitivity = 100% (104/104) (95% CI: 96.52 – 100%)
Sensitivity of the assay was evaluated by testing 104 well-documented samples:

- 44 HIV Ag culture cells supernatants of HIV-1 group M from the following genotypes including: 10A, 5B, 8C, 5D, 10E, 1F, 2G, 1H, 2J

- 21 HIV-Ag commercial positive samples
- 39 HIV-Ag positive samples from the 86 serum samples at different stages of seroconversion

Fresh samples
103 HIV positive samples were tested within 1 day after blood collection.

9.3 Analytical Sensitivity

The Access HIV combo assay shows an analytical sensitivity < 2 IU/mL to HIV-1 p24 Antigen.
The regression analysis of NIBSC 90/636 Panel WHO and Bio-Rad Internal HIV Ag Standard enabled the assay sensitivity limit to be determined.

9.4 Analytical Specificity

9.4.1 Cross-reactivity Study

477 Samples were been tested from patients showing different pathologies or status not linked to the HIV: pregnant women, rheumatoid factor, cirrhotic, chronic renal failure, dialysis, transplants, patients under lenograstim, human anti-mouse Ig, antinuclear antibodies, mycoplasma pneumoniae, erythrovirus B19, myeloma and other viral or bacterial infections (HAV, HBV, HCV, Rubella, Toxoplasmosis, Syphilis, Mumps, Measles, CMV, HSV, EBV, VZV, HTLVI, Malaria, Flu vaccinated patients).
Specificity was equal to 98.10% (414/422) (95% CI: 96.30 – 99.18%) without the frozen transplant population (see Test Limitations, point no. 4).
Five non-specific reactions were found with:

- VZV positive samples (7.7%)
- EBV positive samples (6.7%)
- HCV positive samples (2.9%)
- Rheumatoid factor (7.1%)
- Syphilis positive samples (2.3%)

9.4.2 Interference Study

Samples containing up to 200 mg/L and 300 mg/L for unconjugated and conjugated bilirubins respectively, up to 90 g/L albumin, lipemic samples containing the equivalent of 30 g/L triolein (triglyceride) and hemolyzed samples containing up to 2 g/L hemoglobin, do not affect the results.

ACCESS®

Immunoassay System

HIV combo Calibrators

REF **A59429**

The Access HIV combo Calibrators are intended to calibrate the Access HIV combo assay using the Access Immunoassay Systems.

IVD CE 0459

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1 Intended Use

The Access HIV combo Calibrators are intended to calibrate the Access HIV combo assay for the qualitative detection of HIV-1 antigen and HIV-1/HIV-1-O/HIV-2 antibodies in human serum and plasma (Li heparin, K2-EDTA, K3-EDTA and CPDA-1) using the Access Immunoassay Systems.

2 Summary and Explanation of the Test

The Access HIV combo Calibrators are used to establish calibration (determine the cut-off value) for the Access HIV combo assay. By comparing the light intensity generated by a sample to the cut-off value, the presence or absence of HIV-1 antigen and/or HIV-1/HIV-1-O/HIV-2 antibodies in the sample is determined.

3 Product Information

3.1 Description

Access HIV combo Calibrators

Identification on label	Description	Presentation/ preparation A59429
C0 **Negative Calibrator**	**Negative Calibrator:** (non-reactive) human serum for HIV-1 antigen and HIV-1/HIV-1-O/ HIV-2 antibodies with 0.1% sodium azide and 0.25% ProClin 300.	1 x 1.7 mL Ready to use
C1 **Positive Calibrator**	**Positive Calibrator:** (reactive) human serum for anti-HIV-1 antibodies with 0.1% sodium azide and 0.25% ProClin 300.	1 x 1.7 mL Ready to use
Calibration card	**1**	

3.2 Storage and Handling Conditions

- Store upright and refrigerate at 2 to 10°C.
- Mix contents by gently inverting before use. Avoid bubble formation.
- Stable until the expiration date stated on the label when stored at 2 to 10°C.
- Vial is stable at 2 to 10°C for 120 days after initial use.
- Out-of-range control values are a possible sign of deterioration.

4 Warnings and Precautions

- For *in vitro* diagnostic use. For healthcare professional use only.

4.1 Health and Safety Precautions

- This test kit should only be handled by qualified personnel trained in laboratory procedures and familiar with their potential hazards. Wear appropriate protective clothing, gloves and eye/face protection and handle appropriately in accordance with Good Laboratory Practices.
- Human source material used in the preparation of the calibrators has been tested and found non-reactive for Hepatitis B surface antigen (HBsAg), antibodies to Hepatitis C virus (HCV), antibodies to Human Immunodeficiency virus (HIV-1 and HIV-2) and HIV-1 antigen, except Calibrator C1, that is positive for HIV-1 antibodies. Because no known test method can offer complete assurance that infectious agents are absent, handle reagents and patient samples as if capable of transmitting infectious disease.
- Biological spills: human source material spills should be treated as potentially infectious. Spills not containing acid should be immediately decontaminated, including the spill area, materials and any contaminated surfaces or equipment and with appropriate chemical disinfectant that is effective on the potential biohazards of the samples in question (commonly a 1:10 dilution of household bleach, 70-80% ethanol or isopropanol, an iodophor such as 0.5% Wescodyne™ Plus, etc.) and wiped dry. Spills containing acid should be appropriately absorbed (wiped up) or neutralized, the area flushed with water and wiped dry; materials used to absorb the spill may require hazardous waste disposal. The area should subsequently be decontaminated with one of the chemical disinfectants.

- Dispose of all specimens and material used to perform the test as though they contain an infectious agent. Laboratory, chemical or bio-hazardous waste must be handled and discarded in accordance with all local, regional and national regulations.
- For hazard and precaution recommendations related to any chemical components in this test kit, please refer to the pictogram(s) featured on the labels and the information supplied in the section 4.2. The Safety Data Sheet (SDS) is available at www.bio-rad.com.

4.2 Precautions Related to the Procedure

Warning:

H317: May cause an allergic skin reaction.
P280: Wear protective gloves/protective clothing/eye protection/face protection.
P333+P313: If skin irritation or rash occurs: Get medical advice/attention.
P302+P352: If on skin: Wash with plenty of soap and water.
P501: Dispose of contents/container in accordance with local/regional/national/international regulations.

- This product contains human or animal components. Handle with care.

5 Procedure

Refer to the appropriate system manuals and/or Help system for information on calibration theory, configuring calibrators, entering calibrator test requests and reviewing calibration data.

Calibration
The Access HIV combo Calibrators are provided as negative (C0) and positive (C1). The Access HIV combo assay requires a calibration curve (determination of the cut-off value) every 56 days in order to have an active “calibration” for only one lot of reagents well identified by its bar code. At the end of the 56 days or if another lot of reagents is loaded on the system, the curve is automatically invalidated.
Each calibration requires 220 µL of the C0 calibrator (determination in duplicate) and 330 µL of the C1 calibrator (determination in triplicate) in addition to the sample container and system dead volume.
One drop is equal to approximately 40 µL.

6 Test Limitation

If there is evidence of microbial contamination or excessive turbidity in a reagent, discard the vial.

ACCESS®
Immunoassay System

HIV combo QC

REF **A59430**

For monitoring the system performance of the Access HIV combo assay.

IVD CE 0459

A97207E - [EN] - 2017/03

1 Intended Use

The Access HIV combo QC is intended for monitoring the system performance of the Access HIV combo assay.

2 Summary and Explanation of the Test

Quality control materials simulate the characteristics of patient samples and are essential for monitoring the system performance of the Access HIV combo assay. In addition, they are an integral part of good laboratory practices[38,41-47]. When performing assays with Access reagents for HIV-1 antigen and anti HIV-1/HIV-1-O/HIV-2 antibodies, include quality control materials to validate the integrity of the assays. The assayed values should fall within the acceptable range if the test system is working properly.

3 Product Information

3.1 Description

Access HIV combo QC

Identification on label	Description	Presentation/ preparation A59430
QC1 **Negative QC**	**Negative QC:** Human serum negative (non-reactive) for HIV-1 antigen and anti HIV-1/HIV-1-O/HIV-2 antibodies, with 0.1% sodium azide and 0.25% ProClin 300.	2 x 4.4 mL Ready to use
QC2 **Anti-HIV-1**	**Anti-HIV-1, positive QC:** Human serum positive (reactive) for anti-HIV-1 antibodies with 0.1% sodium azide and 0.25% ProClin 300.	2 x 4.4 mL Ready to use
QC3 **HIV-1 Ag**	**HIV-1 Ag, positive QC:** Purified HIV-1 antigen heat inactivated with a chaotropic agent in Tris Buffer with 0.1% ProClin 300	2 x 4.4 mL Ready to use
QC card	**1**	

3.2 Storage and Handling Conditions

- Store upright and refrigerate at 2 to 10°C.
- Mix contents by gently inverting before use. Avoid bubble formation.
- QC are stable until the expiration date stated on the label when stored at 2 to 10°C.
- Vial is stable at 2 to 10°C for 120 days after initial use.
- Out-of-range quality control values are a possible sign of deterioration.
- Refer to the QC value card for mean values and standard deviations (SDs).

4 Warnings and Precautions

- For *in vitro* diagnostic use. Healthcare professional use only.

4.1 Health and Safety Precautions

- This test kit should only be handled by qualified personnel trained in laboratory procedures and familiar with their potential hazards. Wear appropriate protective clothing, gloves and eye/face protection and handle appropriately in accordance with Good Laboratory Practices.
- Human source material used in the preparation of the control has been tested and found non-reactive for Hepatitis B surface antigen (HBsAg) and antibodies to Hepatitis C virus (HCV). Because no known test method can offer complete assurance that infectious agents are absent, handle reagents and patient samples as if capable of transmitting infectious disease.
- Biological spills: human source material spills should be treated as potentially infectious.
  Spills not containing acid should be immediately decontaminated, including the spill area, materials and any contaminated surfaces or equipment and with appropriate chemical disinfectant that is effective on the potential biohazards of the samples in question (commonly a 1:10 dilution of household bleach, 70-80% ethanol or isopropanol, an iodophor such as 0.5% Wescodyne™ Plus, etc.) and wiped dry.

Spills containing acid should be appropriately absorbed (wiped up) or neutralized, the area flushed with water and wiped dry; materials used to absorb the spill may require hazardous waste disposal. The area should subsequently be decontaminated with one of the chemical disinfectants.

- Dispose of all specimens and material used to perform the test as though they contain an infectious agent. Laboratory, chemical or bio-hazardous waste must be handled and discarded in accordance with all local, regional and national regulations.
- For hazard and precaution recommendations related to any chemical components in this test kit please refer to the pictogram(s) featured on the labels and the information supplied in the section 4.2 The Safety Data Sheet (SDS) is available at www.bio-rad.com.

4.2 Precautions Related to the Procedure

Warning:

H317: May cause an allergic skin reaction.
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P333+P313: If skin irritation or rash occurs: Get medical advice/attention.
P302+P352: If on skin: Wash with plenty of soap and water.
P501: Dispose of contents/container in accordance with local/regional/national/international regulations.

- This product contains human or animal components. Handle with care.

5 Procedure

The Access HIV combo QC should be treated in the same way as patient specimens and run in accordance with the instructions accompanying the instrument and/or method being used.
To process the Access HIV combo QC, 110 µL of sample is required for each of the 3 levels in addition to the sample container and system dead volume (single determination). One drop is equal to approximately 40 µL.
Because samples can be processed at any time in a “random access” format rather than a “batch” format, quality control materials should be included in each 24-hour time period[38]. More frequent use of controls or the use of additional controls is left to the discretion of the user based on good laboratory practices or laboratory accreditation requirements and applicable laws.
Refer to the appropriate system manuals and/or Help system for information on quality control theory, configuring controls, entering quality control sample test requests and reviewing quality control data.

6 Test Limitations

1. The use of the Access HIV combo QC has not been established with assays other than the Access HIV combo assay.
2. Quality control results that do not fall within acceptable ranges may indicate invalid test results. Examine all test results generated since obtaining the last acceptable quality control test point for this analyte.
3. If there is evidence of microbial contamination or excessive turbidity in a reagent, discard the vial.

7 Expected Values

The expected means (×) and SDs (σ) for the Access HIV combo QC1, QC2 and QC3 are provided on the QC value card contained in the kit. Each laboratory should establish its own acceptability criteria by selecting the QC rules to be applied to the control results. Individual control results should fall within the initial acceptance range. However, each laboratory should update the mean and SD once sufficient data has been collected[38,45].
Given that specific levels of reactivity may vary by assay manufacturer, by procedure, by lot number and by laboratory, each laboratory should determine the specific level of reactivity and establish its own range of acceptable values[38,47]. The acceptable range might include all values within ± 2 SD of the mean of 20 data points out of 20 determinations over a period of 30 days[45].

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