Panel Member	Day Since 1st Bleed	ELECTRA™ HCV (S/Co)	Abbott PRISM (S/Co)	Abbott 2.0 (S/Co)	Abbott 3.0 (S/Co)	Abbott AxSYM 3.0 (S/Co)	Abbott IMx 3.0 (S/Co)	Diag. Past. Monolisa PLUS (S/Co)	Ortho 3.0 US (S/Co)	Ortho Enhanced SAVe (S/Co)
1	0	0.02	0.1	0.4	0.1	0.1	0.2	0.1	0	0
2	5	This Panel member is no longer available								
3	7	0.06	0.1	1.0	0.3	0.2	0.1	0.2	0	0
4	13	1.23	1.0	1.8	0.8	1.8	1.0	0.9	0.5	0.4
5	16	4.02	3.6	2.9	1.8	7.0	4.6	3.2	3.1	2.1
6	20	7.43	4.3	3.4	1.7	9.0	4.1	3.2	3.6	2.9
7	26	7.25	5.7	3.4	1.9	19.6	8.0	6.2	>5.0	>4.8
8	28	8.9	5.3	3.7	3.9	29.8	11.8	8.4	>5.0	>4.8
9	33	12.87	6.4	>4.4	>4.5	63.4	26.2	10.5	>5.0	>4.8
10	35	12.87	7.3	>4.4	>4.5	62.9	24.8	9.7	>5.0	>4.8

REMARKS

1. Though **ELECTRA™ HCV Ab CLIA** is a reliable screening assay, it should not be used as a sole criterion for diagnosis of HCV infection. Reactive sample should be retested with confirmatory assays like RIBA, HCV DNA by PCR etc.
2. Absence of HCV antibodies does not indicate that an individual is absolutely free of HCV infection.
3. Since various tests for HCV antibodies differ in their performance characteristics and antigens used, their reactivity patterns may differ.
4. As with all diagnostic tests, a definitive clinical diagnosis should not be based on the result of a single test, but should only be made by the physician after all clinical and laboratory findings have been evaluated.
5. Interferences due to heterophile antibodies, Rheumatoid Factors and other non-analyte substances in patient's serum, capable of binding antibodies multivalently and providing erroneous analyte detection in immunoassays, has been reported in various studies. Though **ELECTRA™ HCV Ab CLIA** uses sufficient amounts of HETEROPHILE BLOCKING REAGENT (HBR) to inhibit the majority of this interference; nevertheless, some samples with high titers may still express clinically important assay interference. Both laboratory professionals and clinicians must be vigilant to this possibility of antibody interference. Results that appear to be internally inconsistent or incompatible with the clinical presentation should invoke suspicion of the presence of an endogenous artefact and lead to appropriate in vitro investigative action.

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SYMBOL KEYS

Temperature Limitation	Consult Instructions for use	Date of Manufacture	LOT Batch Number / Lot Number
Manufacturer	IVD *In vitro* Diagnostic Medical Device	This side up	Σ Contains sufficient for <n> tests
Use by	REF Catalogue Number	Do not reuse	

Manufactured by:

Qualpro Diagnostics

A Division of Tulip Diagnostics (P) Ltd.

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1220/VER-06

electra™

HCV Ab CLIA

Chemiluminescence Assay for the Detection of Antibodies to Hepatitis C Virus in Human Serum or Plasma.

FOR IN VITRO DIAGNOSTIC USE ONLY

INTENDED USE

ELECTRA™ HCV Ab CLIA is intended to be used for the detection of antibodies to Hepatitis C Virus in Human Serum or Plasma.

SUMMARY

ELECTRA™ HCV Ab CLIA is a micro-well Chemiluminescence assay (CLIA) which employs highly purified recombinant antigens representing most conserved antigenic segments of Core, NS3, NS4 and NS5 antigens from multiple genotypes. These antigens are so selected that they recognize all six major HCV genotypes of HCV prevalent worldwide. **ELECTRA™ HCV Ab CLIA** is categorised as fourth generation HCV CLIA, based on high sensitivity as compared with licenced third generation EIA kits.

PRINCIPLE

The **ELECTRA™ HCV Ab CLIA** assay is for use on **ELECTRA™ SA** and **ELECTRA™ FA** analyzers. **ELECTRA™ HCV Ab CLIA** works on the principle of chemiluminescence wherein light is produced by a chemical reaction from a substance as it returns from an electronically excited state to the ground state. When catalysed by HRP,the oxidation of luminol by hydrogen peroxide produces an electronically excited form of 3-aminophthalate which on relaxation emits light with maximum intensity at λ=425nm.

ELECTRA™ HCV Ab CLIA micro-well strips are coated with recombinant antigens representing Core, NS3, NS4 and NS5 antigens from multiple HCV genotypes. Samples along with positive and negative controls are added in the coated microwells and incubated. The microwells are washed to remove the unbound component and goat anti-human IgG conjugated to horseradish peroxidase (HRPO) is added. After incubation the wells are washed again to remove unbound components. This is followed by addition of Chemiluminescence substrate. The bound enzyme converts the substrate to a reaction product that emits a photon of light.

The Chemiluminescence thus produced is measured in Relative Light Units (RLU) that are typically proportionate to the amount of analyte present in the sample. The presence or absence of analyte in the sample is determined by comparing the sample RLU with Cutoff which is calculated by **ELECTRA™ SA** and **ELECTRA™ FA** analyzers and expressed as Electra Cutoff Index (E.C.I.). E.C.I. is equivalent to S/Co ratio which is calculated by using sample RLU and calculated Cutoff of specific testing batch. Samples with E.C.I. values greater than or equal to 1.00 are considered reactive and samples with E.C.I. values less than 1.00 are considered non-reactive.

KIT COMPONENTS

ELECTRA™ HCV Ab CLIA has following components:

1. Coated micro-wells: Microwells coated with recombinant antigens derived from multiple genotypes of HCV. Ready to use. 96 Wells: (3x8) x 4 pouches, 192 Wells: (3x8) x 8 pouches.
2. Positive control: Inactivated and stabilized human serum reactive for HCV antibodies with preservatives.
3. Negative control: Inactivated and stabilized human serum non-reactive for HIV-1 and HIV-2, HBsAg and HCV.
4. Conjugate: Anti-human IgG - HRPO conjugate.
5. Conjugate Diluent: Buffered solution containing stabilizing proteins and preservatives.
6. Sample Diluent: Buffered solution containing stabilizing proteins and preservatives.
7. Substrate A: Chemiluminescent substrate containing enhanced luminol solution.
8. Substrate B: Chemiluminescent substrate containing stabilized peroxide solution.
9. Wash buffer: Buffer containing surfactants (20 X). To be diluted 20 times with distilled or deionized water.
10. Microwell holder.
11. Instructions for use
12. Plate sealer.

REF	407020096	407020192
Σ	96 Tests	192 Tests

STORAGE AND STABILITY

1. **ELECTRA™ HCV Ab CLIA** kit is stable at 2-8°C up to the expiry date printed on the label.
2. Coated micro-wells should be used within one month of opening the pouch. Once opened, the pouch must be sealed properly to protect from moisture. In case the desiccant pouch changes color from blue to white, the strips should not be used.

3. Diluted wash buffer is stable up to one week at 2-8°C.
4. Diluted Conjugate must be used immediately.
5. Working Substrate (A+B) must be used immediately.

MATERIAL REQUIRED BUT NOT PROVIDED

1. Manual or automatic pipettor.
2. Pipettor tips.
3. Incubator.
4. Micro-well washer.
5. **ELECTRA™ SA** or **ELECTRA™ FA**.
6. Distilled water.
7. Disposable gloves.
8. Timer/ Stop Watch.

SAMPLE COLLECTION

1. No prior preparation of the patient is required.
2. Collect blood specimen by venipuncture according to the standard procedure.
3. Serum or plasma can be used.
4. Specimen should be free of particulate matter and microbial contamination.
5. Specimen containing precipitate or particulate matter should be centrifuged prior to use.
6. Use of fresh sample is preferred. However, specimen samples can be stored refrigerated for short duration. For long term storage, freeze at -20°C or below. Do not freeze samples in frost-free freezer.
7. Specimen should not be frozen and thawed repeatedly.
8. Do not heat inactivate before use.

PRECAUTIONS

1. Do not pipette any material by mouth.
2. Do not eat, drink or smoke in the area where testing is done.
3. Use protective clothing and wear gloves when handling samples.
4. Immediately clean up any spills with sodium hypochlorite.
5. Dispose off all the reagents and material used as they contain infectious agent.
6. Neutralize acid containing waste before adding hypochlorite.
7. Do not use kit after the expiry date.
8. Do not mix components of one kit with another.
9. Always use new tip for each specimen and reagent.
10. Do not allow liquid from one well to mix with other wells.
11. Do not let the strips dry in between the steps.
12. Do not expose the working substrate to direct light.

REAGENT PREPARATION

1. Dilute wash buffer 20 times (for example add 5 ml concentrated buffer to 95 ml distilled or deionized water).
2. Dilute conjugate 50 times (for example add 20 µl concentrated Conjugate to 980 µl Conjugate Diluent).

No. of Strips	1	2	3	4	5	6	7	8	9	10	11	12
Conjugate	10 µl	20 µl	30 µl	40 µl	50 µl	60 µl	70 µl	80 µl	90µl	100 µl	110 µl	120 µl
Conjugate Diluent	490µl	980µl	1470 µl	1960µl	2450µl	2940µl	3430µl	3920µl	4410µl	4900µl	5390µl	5880 µl

3. Working Substrate: Mix Substrate A and Substrate B (1:1 ratio) in equal volume before addition to the micro-wells.

No. of Strips	1	2	3	4	5	6	7	8	9	10	11	12
Substrate A	250 µl	450 µl	650 µl	850 µl	1050µl	1250µl	1450µl	1650µl	1850µl	2050µl	2250µl	2450µl
Substrate B	250 µl	450 µl	650 µl	850 µl	1050µl	1250µl	1450µl	1650µl	1850µl	2050µl	2250µl	2450µl

TEST PROCEDURE

1. Bring all the reagents and specimen to room temperature before use.
2. Take out required number of strips and immediately close the pouch.
3. Well A1 must be used as blank.
4. Add 200 µl Sample Diluent in each well except well A1.
5. Add 10 µl of NC's to B1 & C1 and PC's to D1 & E1 respectively.
6. Add 10 µl of samples to remaining wells.
7. Gently shake the plate to mix thoroughly. Apply plate sealer and incubate for 20 minutes at 37°C.
8. Wash each well using a semi-automated micro-plate washer (Preferably LisaWash models) with diluted wash buffer for 6 wash cycles giving 30 seconds soak time for each wash cycle and Blot dry.
9. Add 50 µl Diluted Conjugate in each well except well A1 and incubate for 20 minutes at 37°C.
10. Wash six times as described in step 8 and Blot dry.
11. Add 50 µl of working Substrate (A+B) in all the micro-wells. Keep away from direct light while adding the substrate.
12. Cover the Electra microplate and incubate for 10 mins at room temperature (18-25°C) in dark.
13. Read the Electra micro-plate exactly at 10 mins in **ELECTRA™ SA** or **ELECTRA™ FA.** If Electra micro-plate is not read between 10- 15 mins, the test results should be considered as invalid.

TEST VALIDATION CRITERIA

1. The individual RLU of negative controls should be less than 200000 for **ELECTRA™ SA** and **ELECTRA™ FA.**
2. The individual RLU of positive controls should be more than 7500000 for **ELECTRA™ SA** and **ELECTRA™ FA.**

CALCULATIONS

The cutoff value is calculated by the software of **ELECTRA™ SA** and **ELECTRA™ FA** using the mean of negative control RLU and a factor.
Factor for **ELECTRA™ SA** and **ELECTRA™ FA** is 800000

SAMPLE DATA

For **ELECTRA™ SA** and **ELECTRA™ FA:**
Considering the Cutoff for this batch is 800069.

Well	RLU	E.C.I.	Interpretation
Blank	39		
NC	68		
NC	70		
PC	14986969		
PC	14995136		
Sample 1	10480190	13.09	Reactive
Sample 2	41821	0.05	Non-reactive
Sample 3	5625266	7.03	Reactive
Sample 4	124176	0.155	Non-reactive

INTERPRETATION OF RESULTS

1. Results are interpreted in E.C.I. This is determined by dividing the RLU of the sample by the Cutoff value calculated for that specific run.
2. Samples with E.C.I. values greater than or equal to 1.00 are considered Reactive and samples with E.C.I. values less than 1.00 are considered Nonreactive.
3. Samples that are initially reactive in **ELECTRA HCV Ab CLIA** should be retested in duplicate. Repeat reactivity is highly predictive of the presence of HCV antibodies.
4. As with all immunoassays, the **ELECTRA HCV Ab CLIA** may yield nonspecific reactions due to other causes, particularly when testing in low prevalence populations. A repeatedly reactive specimen should be tested further with supplementary confirmatory assays.
5. A grey zone of ± 10% is recommended.

PERFORMANCE CHARACTERISTICS

1. **ELECTRA™ HCV Ab CLIA** was evaluated with 729 samples out of which 718 were negative samples and 11 were HCV positive samples. The results were compared with commercially available HCV EIA.

Specimen Data	Total	ELECTRA™ HCV Ab	Other HCV EIA
Total Specimens	729	729	729
HCV Reactive	11	11	11
HCV Non-reactive	718	718	718

2. **ELECTRA™ HCV Ab CLIA** was also evaluated against Reactive and Non-reactive panels provided by national Institute of Biologicals (NIB), India. In this evaluation, HCV reactive panel ID C1-C100 consisting of 100 HCV positive samples were tested and showed 100% correlation. HCV Non-reactive panel ID N1-N300 consisting of 300 negative samples was also evaluated and showed 100% correlation.
3. **ELECTRA™ HCV Ab CLIA** was also evaluated using the serial dilution of 11 HCV Positive samples.
4. **ELECTRA™ HCV Ab CLIA** was also evaluated using British Working Standard for anti-HCV, NIBSC Code: 14/240.
5. **ELECTRA™ HCV Ab CLIA** was also evaluated using AccuVert HCV Seroconversion panel (PHV920(M)) from Seracare.