

**ELISA kits available from ADI (see details at the web site)**

Catalog#	ProdDescription
4200	Human Anti-Hepatitis B Surface Antigen (anti-HBsAg) IgG ELISA kit
4205	Human Anti-Hepatitis B Surface Antigen (anti-HBsAg) IgM ELISA kit
4220-AHB	Human Anti-Hepatitis B Surface Antigen (anti-HBsAg) ELISA kit, Quantitative
4300-AHG	Human Anti-Hepatitis A Virus IgG (HAV-IgG) ELISA kit, Quantitative
4600	Human Anti-Hepatitis C Virus (Anti-HCV) ELISA kit, Semi-Quantitative
510-100-HRG	Human Anti-Rubella Virus IgG ELISA kit
510-110-HRM	Human Anti-Rubella Virus IgM ELISA kit
520-100-HMG	Human Anti-Mumps Virus (parotitis) IgG ELISA, 96 tests, Quantitative
520-110-HMM	Human Anti-Mumps Virus (parotitis) IgM ELISA, 96 tests, Quantitative
520-120-HMA	Human Anti-Mumps Virus (parotitis) IgA ELISA, 96 tests, Quantitative
520-200-HVG	Human Anti-Varicella Zoster Virus (chickenpox) IgG ELISA, 96 tests, Quantitative
520-210-HVM	Human Anti-Varicella Zoster Virus (chickenpox) IgM ELISA, 96 tests, Quantitative
520-220-HVG	Human Anti-Varicella Zoster Virus (chickenpox) IgA ELISA, 96 tests, Quantitative
530-100-HMG	Human Anti-Measles IgG ELISA kit, 96 tests
530-110-HMM	Human Anti-Measles IgM ELISA kit, 96 tests
530-120-HMA	Human Anti-Measles IgA ELISA kit, 96 tests
970-100-PHG	Human Anti-Polio Virus IgG ELISA kits, 96 tests, Quantitative
540-110-DHM	Human Anti-Polio Virus IgM ELISA kits, 96 tests
600-020-HRV	Human Anti-Rabies Virus IgG ELISA Kit, 96 tests, Quantitative
600-120-HRV	Human Anti-Rabies Virus Glycoprotein (RVG) IgG ELISA Kit, 2x 96 tests,
600-220-HRV	Human Anti-Rabies Virus Nucleoprotein (RV-NP) IgG ELISA Kit, 2x 96 tests,
600-300-100	Human Anti-Meningococcal Group A Oligosaccharides-Diphtheria CRM197 IgG
600-300-105	Human Anti-Meningococcal Group CWY Oligosaccharides-Diphtheria CRM197
600-300-115	Human Anti-Meningococcal Group ACWY Oligosaccharides-Diphtheria CRM197
600-370-CFP	Human Cardiac Fatty acid binding protein (FABP) ELISA kit
600-410-CTN	Human Cardiac Troponin-I (Tn-I) ELISA Kit
600-610-HMY	Human Myoglobin ELISA Kit
700-140-KLM	Human Anti-KLH IgG (total) ELISA Kit, 2x 96 tests, Quantitative
700-160-VAH	Human Anti-Vacmune/Immucotest (KLH) IgG (total) ELISA Kit, 2x 96 tests,
710-140-BSM	Human Anti-BSA IgG (total) ELISA Kit, 2x 96 tests, Quantitative
80170	Human Serum Antibody detection ELISA kit, Qualitative
900-160-83T	Human Anti-Anthrax Protective Antigen 83 (PA83) Ig's ELISA kit
910-160-JEM	Human Anti-Japanese encephalitis virus (JEV) IgG specific ELISA kit
910-170-JEM	Human Anti-Japanese encephalitis virus (JEV) IgM specific ELISA kit
920-040-HAG	Human Anti-Influenza A virus IgG ELISA kit
920-050-HAM	Human Anti-Influenza A virus IgM ELISA kit
920-060-HAA	Human Anti-Influenza A virus IgA ELISA kit
920-400-HBG	Human Anti-Influenza B virus Ig's ELISA kit
930-100-TTH	Human Anti-Tetanus Toxin/Toxoid IgG ELISA kit, 96 tests, Quantitative
940-100-DHG	Human Anti-Diphtheria Toxin/Toxoid IgG ELISA kit, 96 tests, Quantitative
940-110-DHM	Human Anti-Diphtheria Toxin/Toxoid IgM ELISA kit, 96 tests, Quantitative
940-200-DHG	Human Anti-CRM197 (Diphtheria Toxin mutant) IgG ELISA kit
940-210-DHM	Human Anti-CRM197 (Diphtheria Toxin mutant) IgM ELISA kit
950-100-AHA	Human Anti-Adenovirus IgA ELISA kit
950-110-AHG	Human Anti-Adenovirus IgG ELISA kit
950-120-AHM	Human Anti-Adenovirus IgM ELISA kit
960-200-PHA	Human Anti-B. pertussis antigens (Pertussis toxin, FHA and LPS) IgA ELISA kit,
960-220-PHM	Human Anti-B. pertussis antigens (Pertussis toxin, FHA and LPS) IgM ELISA kit,
960-250-PHG	Human Anti-B. pertussis Pertactin IgG ELISA kit
970-100-PHG	Human Anti-Poliomyelitis Virus 1-3 IgG ELISA Kit, 96 tests
980-100-PHG	Human Anti-H. Influenzae B (Hib) polyribosyl phosphate (PRP) IgG ELISA Kit, 96
980-110-PHM	Human Anti-H. Influenzae B (Hib) polyribosyl phosphate (PRP) IgM ELISA Kit, 96
990-100-THA	Human Anti-Mycobacterium Tuberculosis IgA ELISA kit, 96 tests
990-110-THG	Human Anti-Mycobacterium Tuberculosis IgG ELISA kit, 96 tests
990-120-THM	Human Anti-Mycobacterium Tuberculosis IgM ELISA kit, 96 tests
AE-320420-1	Human Crimean-Congo hemorrhagic fever virus (CCHFV) IgG ELISA Kit, 96 tests
AE-320430-1	Human Crimean-Congo hemorrhagic fever virus (CCHFV) IgM ELISA Kit, 96 tests
AE-320520-1	Human Anti-Zaire-Ebola virus IgG ELISA Kit, 96 tests

Instruction Manual No. M-970-100-PHG

## Human Anti- Poliomyelitis Virus 1-3 IgG ELISA KIT

**Cat. # 970-100-PHG; 96 Tests**

**For the detection of IgG antibody to Polio viruses 1-3  
IgG in human serum or plasma.**



**For In Vitro Research Use Only**



**ALPHA DIAGNOSTIC  
INTERNATIONAL**

**India Contact:**

**Life Technologies (India) Pvt. Ltd.**

306, Aggarwal City Mall, Opposite M2K Pitampura, Delhi – 110034  
Ph: +91-11-42208000, 42208111, 42208222

Mobile: +91-9810521400

Fax: +91-11-42208444

Email: [customerservice@atlabs.com](mailto:customerservice@atlabs.com)

Web: [www.atlabs.com](http://www.atlabs.com)



Kit Components (96 tests)	
Polio viruses antigens 1-3 antigen coated strip plate, (8x12 strip or 96 wells) # 970-101	1 plate
<b>Calibrator A</b> (2 mL; Polio virus IgG negative control) #970102A	1 vial
<b>Calibrator B</b> (2 mL; Polio virus IgG Cut-off standards) #970102B	1 vial
<b>Calibrator C</b> (2 mL; Polio virus IgG weak positive control) #970102C	1 vial
<b>Calibrator D</b> (2 mL; Polio virus IgG positive control) #970102D	1 vial
Anti-Human IgG-HRP <b>Conjugate</b> , (15 ml) #970103	1 bottle
<b>Sample Diluent</b> , 60 ml # 970-104	1 bottle
<b>Wash buffer</b> (10X) 60 ml # 970100-WB	1 bottle
<b>TMB Substrate</b> Solution, 15 ml #970100-TMB	1 bottle
<b>Stop Solution</b> , 15 ml # 970100-ST	1 bottle
Complete Instruction Manual	1

### Intended Use

ADI **Polio viruses 1-3 IgG** ELISA Kit is intended for the detection of IgG antibody to Polio virus in human serum or plasma. This kit is for in vitro research use only.

### Introduction

Poliomyelitis, often called polio or infantile paralysis, is an acute viral infectious disease spread from person to person, primarily via the fecal-oral route. Different types of paralysis may occur, depending on the nerves involved. The term poliomyelitis is used to identify the disease caused by any of the three serotypes of poliovirus. Type 1 (Brunhilde): often with severe symptoms Type 2 (Lansing): with milder symptoms Type 3 (Leon): rare, but with severe symptoms. The virus enters the central nervous system in about 3% of infections. Most patients with CNS involvement develop non-paralytic aseptic meningitis, with symptoms of headache, neck, back, abdominal and extremity pain, fever, vomiting, lethargy and irritability. A laboratory diagnosis is usually made based on recovery of poliovirus from a stool sample or a swab of the pharynx. Antibodies to poliovirus can be diagnostic, and are generally detected in the blood of infected patients early in the course of infection. Two types of **vaccines** are used throughout the world to combat polio. The first is Salk vaccine, or inactivated poliovirus vaccine (**IPV**), is based on three wild, virulent reference strains, Mahoney (type 1), MEF-1 (type 2), and Saukett (type 3) polio viruses, grown in a type of monkey kidney tissue culture (Vero cell line), which are then inactivated with formalin. Oral polio vaccine (**OPV** or Sabin's vaccine) is a live-attenuated vaccine, produced by the passage of the virus through non-human cells at a sub-physiological temperature, which produces spontaneous mutations in the viral genome. This vaccine is unable to replicate in the brain. .

**IPOL®**, Poliovirus Vaccine Inactivated, produced by Sanofi, contains three types of polioviruses: Type 1 (Mahoney), Type 2 (MEF-1), and Type 3 (Saukett). IPOL vaccine is replicated in Vero cells. Each dose (0.5 mL) of trivalent vaccine is formulated to contain 40 D antigen units of Type 1, 8 D antigen units of Type 2, and 32 D antigen units of Type 3 poliovirus. Poliovirus Vaccine induces the production of neutralizing antibodies against each type of virus which are related to protective efficacy. There are 57 nucleotide

### QUALITY CONTROL

The test run may be considered valid provided the following criteria are met:

1. The O.D. of the Calibrator should be greater than 0.250.
2. The Ab index for Negative control should be less than 0.9.
3. The Ab Index for Positive control should be greater than 1.2.

### INTERPRETATION

The following is intended as a guide to interpretation of Polio virus IgG test results; each laboratory is encouraged to establish its own criteria for test interpretation based on sample populations encountered.

### Antibody Index Interpretation

<0.9 No detectable IgG antibody to Polio virus.

0.9-1.1 Borderline positive. Follow-up testing is recommended if clinically indicated.

>1.1 Detectable IgG antibody to Polio Virus.

### LIMITATIONS OF THE TEST

1. The test results obtained using this kit serve only as an aid to diagnosis and should be interpreted in relation to the patient's history, physical findings and other diagnostic procedures.
2. Lipemic or hemolyzed samples may cause erroneous results.

### Cross Reactivity

No cross reactivity to *Corynebacterium diphtheriae*.

### Polio Vaccine Testing in Animals & Humans

Human samples obtained from vaccinated individuals tested positive using the polio virus antigen coated used in this kit. Rabbits and mice vaccinated with Ipol vaccine also produced high titered antibodies to the antigens used in the kit.

ADI also cloned and expressed Polio virus 1 (Sabin) viral protein 1 (Polv1-VP1, Cat# POLV15-R-10) and vaccinated mouse and rabbits. The resulting antiserum reacted with the human polio viruses 1-3 antigens used in this test. Human samples obtained from polio-vaccinated individuals also reacted with the recombinant POLV-VP1. Therefore, POLV1-VP1 is highly represented in the antigen mix used in the kit, and the anti-VP1 reacting with the natural polio viruses 1-3 VP1s.

### Specificity

Polio viruses' 1-3 antigens are used in the kit. Therefore, antibodies to all three polio serotype will be detected in this test. Anti-human IgG-HRP conjugate has been optimized to detect all IgG subtypes (IgG1-4) but not the IgM or IgA. It is possible to use anti-IgM specific conjugate to detect mouse IgM polio antibodies.

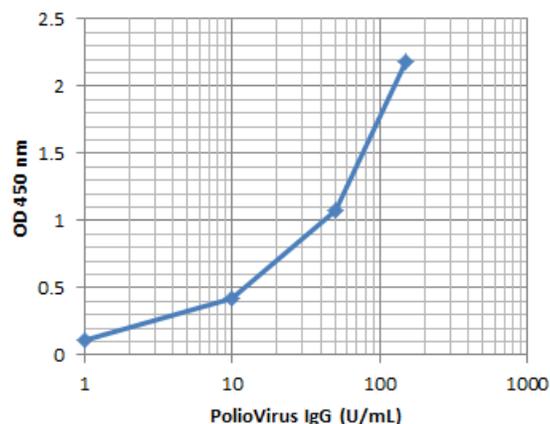
### References:

Pinheiro FP et al; World Health Stat Q 50(3/4):161-169, 1997; Gubler DJ et al; Infect Agents Dis 2:383-393, 1993; Wu SJ et al Clin Diagn Lab Immunol 1997; 4(4):452-7; Lam SK; Clin Diagn Virol 1998;10(1):75-8; Rossi CA; 1995-1996. Am J Trop Med Hyg 1998;59(2):275-8 2008-12-18

## WORKSHEET OF A TYPICAL ASSAY

NOTE: These data are for demonstration purpose only. It must not be used to determine the sample results.

Serum	U/ml	Net A450
Calibrator A (Negative Control)	1	0.109
Calibrator B (Cut off stdl)	10	0.422
Calibrator C (weak positive Control)	50	1.076
Calibrator D (Positive Control)	150	2.184



\*Kit-spec-XL

Typical Std Curve (do not use this for sample calculation)

### CALCULATION OF RESULTS

The mean values for the measured absorptions are calculated after subtraction of the blank values from the controls and standards.

The OD of the calibrators (y-axis, linear) are plotted against their concentration (x-axis, logarithmic) either on semi-logarithmic graph paper or using an automated method. A good fit is provided with cubic spline, 4 parameter logistics or Logit-Log. The initial dilution of unknowns (1:100) has been taken into consideration when reading the results from the graph. Therefore, do not multiply the sample values if used at 1:100 dilution. Results of unknowns of higher predilution (e.g., 1:500 have to be adjusted for the dilution factor and multiplied by 5). Unknowns showing concentrations above the highest calibrator have to be diluted as described in "Assay Procedure" and reassayed.

All samples above the cut-of values may be considered positive for the presence of anti-polio virus IgG.

substitutions which distinguish the attenuated Sabin 1 strain from its virulent parent (the Mahoney serotype), two nucleotide substitutions attenuates the Sabin 2 strain, and 10 substitutions are involved in attenuating the Sabin 3 strain. The primary attenuating factor common to all three Sabin vaccines is a mutation located in the virus's internal ribosome entry site (IRES) which alters stem-loop structures, and reduces the ability of poliovirus to translate its RNA template within the host cell. The attenuated poliovirus in the Sabin vaccine replicates very efficiently in the gut, the primary site of infection and replication, but is unable to replicate efficiently within nervous system tissue.

ADI's human anti-polio virus IgG ELISA kit detects antibodies to the three subtypes of polio viruses that are currently used in vaccines. This serological tests will determine IgG to polio viruses 1-3 that can be due to past illness or by vaccination.

### PRINCIPLE OF THE TEST

ADI's Polio virus IgG ELISA Kit is based on the principle of the enzyme immunoassay (EIA). Diluted patient serum is added to wells coated with purified Polio virus antigen. Polio virus IgG specific antibody, if present, binds to the antigen. All unbound materials are washed away and the enzyme conjugate is added to bind to the antibody-antigen complex, if present. Excess enzyme conjugate is washed off and substrate is added. The plate is incubated to allow the hydrolysis of the substrate by the enzyme that produced blue color. The intensity of the color generated is proportional to the amount of IgG specific antibody in the sample. The color development is terminated by the addition of a stop solution, which changes the color from blue to yellow. The resulting dye is measured spectrophotometrically at the wavelength of 450 nm. The concentration of the IgG antibodies is directly proportional to the intensity of the color.

### MATERIALS AND EQUIPMENT REQUIRED

Adjustable micropipet (5µl, 100µl, 500µl) and multichannel pipet with disposable plastic tips. Bidistilled water, reagent troughs, Orbital shaker, plate washer (recommended) and ELISA plate Reader (450nm).

### PRECAUTIONS

Only for in-vitro use! Do not ingest or swallow! The usual laboratory safety precautions as well as the prohibition of eating, drinking and smoking in the lab have to be followed. All sera and plasma or buffers based upon, have been tested respective to HBsAg, HIV and HCV with recognized methods and were found negative. Nevertheless precautions like the use of latex gloves have to be taken. Serum and reagent spills have to be wiped off with a disinfecting solution (e.g. sodium hypochlorite, 5%) and have to be disposed of properly. All reagents have to be brought to room temperature (18 to 25 °C) before performing the test. Before pipetting all reagents should be mixed thoroughly by gentle tilting or swinging. Vigorous shaking with formation of foam should be avoided. It is important to pipet with constant intervals, so that all the wells of the microtiter plate have the same conditions. When removing reagents out of the bottles, care has to be taken that the stoppers are not contaminated. Further a possible mix-up has to be avoided. The content of the bottles is usually sensitive to oxidation, so that they should be opened only for a short time. In order to avoid a carry-over or a cross-contamination, separate disposable pipet tips have to be used. No reagents from different kit lots have to be used, they should not be mixed among one another. All reagents have to be used within the expiry period. In accordance with a Good Laboratory Practice (GLP) or following ISO9001 all laboratory devices employed should be regularly checked regarding the accuracy and precision. This refers amongst others to microliter pipets and washing or reading (ELISA-Reader) instrumentation. The contact of certain reagents, above all the stopping solution and the substrate with skin, eye and mucosa has to be avoided, because possible irritations and acid burns could arise, and there exists a danger of intoxication.

Applicable **MSDS**, if not already on file, for the following reagents can be obtained from ADI or the web site.

TMB (substrate), H<sub>2</sub>SO<sub>4</sub> (stop solution), and Prolcin-300 (0.1% v/v in standards, sample diluent and HRP-conjugates).

[http://4adi.com/commerce/info/showpage.jsp?page\\_id=1060&category\\_id=2430&visit=10](http://4adi.com/commerce/info/showpage.jsp?page_id=1060&category_id=2430&visit=10)

#### SPECIMEN COLLECTION AND HANDLING

Principally serum or plasma (EDTA, heparin) can be used for the determination. Serum is separated from the blood, which is aseptically drawn by venipuncture, after clotting and centrifugation. The serum or plasma samples can be stored refrigerated (2-8°C) for up to 48 hours, for a longer storage they should be kept at -20 °C. The samples should not be frozen and thawed repeatedly. Lipemic, hemolytic or bacterially contaminated samples can cause false positive or false negative results.

For the performance of the test the samples (not the standards) have to be diluted 1:100 with ready-to-use sample diluent (e.g. 5 µL serum + 495 µL sample diluent). Do not dilute the calibrators.

#### REAGENTS PREPARATION

1. **Dilute Wash buffer** 1:10 with water. (**Dilute 60 ml stock with 540 ml distilled water**) Store diluted buffer at 4°C for 1 month. (If during the cold storage crystals precipitate, the concentrate should be warmed up at 37 degrees C for 15 minutes.

*All reagents must be at room temperature prior to their use.*

#### STORAGE AND STABILITY

The microtiter well plate and all other reagents are stable at 2-8°C until the expiration date printed on the label. The whole kit stability is usually 6 months from the date of shipping under appropriate storage conditions. The unused portions of the standards should be stored at 2-8°C or stored frozen in small aliquots and should be stable for 3 months.

#### TEST PROCEDURE (ALLOW ALL REAGENTS TO REACH ROOM TEMPERATURE BEFORE USE).

Remove required number of coated strips and arrange them on the plate. Store unused strips in the bag. Dilute all samples 1:100 with the sample diluent. It is recommended to prepare a parallel replica plates containing all sample for quick transfer to the coated plate. DO NOT dilute calibrators or controls. **Dilute wash buffer stock (10X) 1:10 with distilled water.**

1. Label or mark the microtiter well strips to be used on the plate
2. Dispense **100 µl** diluent in 1 well to be used as blank. Pipet **100 µl of , calibrators, controls, and diluted samples** into appropriate wells in *duplicate*. See worksheet of a typical set-up on page 5. Cover the plate, mix gently for 5-seconds and **incubate at room temp (25-28°C) for 60 min.**
3. Aspirate the well contents and blot the plate on absorbent paper. Immediately, **wash the wells 3 times** with 300 µl of 1X wash buffer. We recommend using an automated ELISA plate Washer for better consistency. Failure to wash the wells properly will lead to high blank or zero values. If washing manually, plate must be tapped over paper towel between washings to ensure proper washing.
4. Add **100 µl anti-human IgG-HRP conjugate** to all wells leaving one empty for the substrate blank. Mix gently for 5-10 seconds. Cover the plate and **incubate for 30 minutes** at room temp (18-26°C).
5. **Wash the wells 4 times** as in step 3.
6. Add **100 µl TMB substrate solution**. Mix gently for 5-10 seconds. Cover the plate and **incubate for 20 minutes** at room temp. **Blue color** develops in positive controls and samples.
7. Stop the reaction by adding **100 µl of stop solution** to all wells. Mix gently for 5-10 seconds to have uniform color distribution (**blue color turns yellow**).
8. **Measure the absorbance at 450 nm** using an ELISA reader within 60 min.

#### NOTES

Read instructions carefully before the assay. Do not allow reagents to dry on the wells. Careful aspiration of the washing solution is essential for good assay precision. Since timing of the incubation steps is important to the performance of the assay, pipet the samples without interruption and it should not exceed 5 minutes to avoid assay drift. If more than one plate is being used in one run, it is recommended to include a standard curve on each plate. The unused strips should be stored in a sealed bag at 4°C. Do not touch the bottom of the wells.