

ab285278– Salmonella Typhi IgG ELISA Kit

For the quantitative measurement of Salmonella Typhi IgG.

For research use only - not intended for diagnostic use.

For overview, typical data and additional information please visit:

<http://www.abcam.com/ab285278>

Storage and Stability

On receipt entire kit should be stored at 4°C, protected from light. Upon opening, use kit within 6 months. Avoid freeze-thaw cycles. Keep microwells sealed in a dry bag with desiccants. Do not expose reagent to heat, sun, or strong light.

Materials Supplied

Item	Quantity	Storage Condition
Microplate	12 strips X 8 wells	4°C
Sample Diluent	2 X 25 ml	4°C
Calibrator	1 ml	4°C
Positive Control	1 ml	4°C
Negative Control	1 ml	4°C
Enzyme conjugate	12 ml	4°C
TMB Substrate	12 ml	4°C
Stop Solution	12 ml	4°C
Wash Buffer (20X)	25 ml	4°C

Materials Required, Not Supplied

These materials are not included in the kit, but will be required to successfully utilize this assay:

- Microplate reader capable of measuring absorbance at 450 nm
- Absorbent paper
- Adjustable pipettes and pipette tips

Reagent Preparation

- Prepare reagents within 30 minutes before the experiment.
- Spin briefly to bring down all components to the bottom of the tubes.

Wash Buffer: Prepare 1X Wash buffer by adding the contents of the bottle (25 ml, 20X) to 475 ml of distilled or deionized water. Store at room temperature (18-26°C).

Sample Preparation

Collect blood specimens & separate the serum immediately. Specimens may be stored refrigerated at (2-8°C) for 7 days. Store frozen at (-20°C) for up to six month. Avoid multiple freeze-thaw cycles. Prior to assay, frozen sera should be completely thawed and mixed well.

Assay Protocol

Δ Note: Bring all reagents and samples to room temperature. Gently mix all reagents before use.

1. Place the desired no. of coated strips into the holder. Replace any unused microwell strips back into the aluminum bag, seal, and store at 2-8°C.
2. Negative control, positive control, and calibrator are ready to use. Prepare 1:101 dilution of test samples, by adding 5 µl of the sample to 0.5 mL of sample diluent. Mix well.
3. Dispense 100 µl of diluted sera, calibrator and controls into the appropriate wells. For the reagent blank, dispense 100 µl sample diluent in 1A well position. Tap the holder to remove air bubbles from the liquid and mix well. Incubate for 20 minutes at room temperature.
4. Remove liquid from all wells. Wash wells three times with 300 µl of 1X wash buffer. Blot on absorbance paper or paper towel.
5. Dispense 100 µl of enzyme conjugate to each well and incubate for 20 minutes at room temperature.
6. Remove enzyme conjugate from all wells. Wash wells three times with 300 µl of 1X wash buffer. Blot on absorbance paper or paper towel.
7. Dispense 100 µl of TMB substrate and incubate for 10 minutes at room temperature.
8. Add 100 µl of stop solution.
9. Read O.D. at 450 nm using ELISA reader within 15 min. A dual wavelength is recommended with reference filter of 600-650 nm

Calculation:

- Check Calibrator Factor (CF) value on the calibrator bottle. This value might vary from lot to lot. Make sure you check the value on every kit.
- Calculate the cut-off value: Calibrator OD x Calibrator Factor (CF). Calculate the Ab (Antibody) Index of each determination by dividing the O.D. value of each sample by cut-off value.

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 fall within the range specified on the COA/label.

Interpretation

The following is intended as a guide to interpretation of IgG antibody 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 antibody to IgG antibody by ELISA.
- 0.9 - 1.1 Borderline positive. Follow-up testing is recommended if clinically indicated.
- >1.1 Detectable antibody to IgG antibody by ELISA.

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.

Precision

Intra-assay precision n=16 mean values in sera are as followed:

Serum	Intra-assay precision
1	1.62
2	0.83
3	0.15

Inter-assay precision n=10 mean values in sera are as followed:

Serum	Inter-assay precision
1	1.14
2	0.76
3	0.18

Technical Support

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