

# ELISA Flex: Human IgG high sensitivity

3850-1A-6 |

ELISA Flex kit for quantitative determination of native human IgG in solution, e.g. serum/plasma samples or cell supernatants.

<b>The kit includes</b>		<b>3850-1A-6 for 6 plates</b>	
Capture mAb:	MT145 (0.5 mg/ml)	300 µl	
Detection mAb:	MT78, biotin (0.5 mg/ml)	50 µl	
Streptavidin-ALP		80 µl	
Purified human IgG ELISA standard		1 vial	
Standard reconstitution buffer A5		1 ml	

To ensure total recovery of the stated quantity, vials have been overfilled.

## Shipping and storage

Shipped at ambient temperature. All reagents should be stored at 4-8 °C upon receipt, except the standard which should be stored at -20 °C. Antibodies are supplied in sterile-filtered PBS with sodium azide (0.02%). Streptavidin-ALP is supplied in 0.1 M Tris buffer with 0.002% Kathon CG. The expiry date indicates how long unopened products, stored according to instructions, are recommended for use.

# General and Preparations

## Specificity

The kit contains a matched pair of monoclonal antibodies (mAbs) specific for the Fc part of human IgG. The mAbs cross-react with IgG from non-human primates (NHP). Please visit [www.mabtech.com](http://www.mabtech.com) for reactivity on NHP species.

## Standard range

0.1-10 ng/ml

## Calibration

The ELISA standard has been calibrated against an international standard from the National Institute of Biological Standards and Control (NIBSC), Potters Bar, Hertfordshire EN6 3QG, UK. One  $\mu\text{g}$  of supplied standard equals 11 mU NIBSC-standard. Please note that the calibration is batch specific.

## Analysis of serum and plasma samples

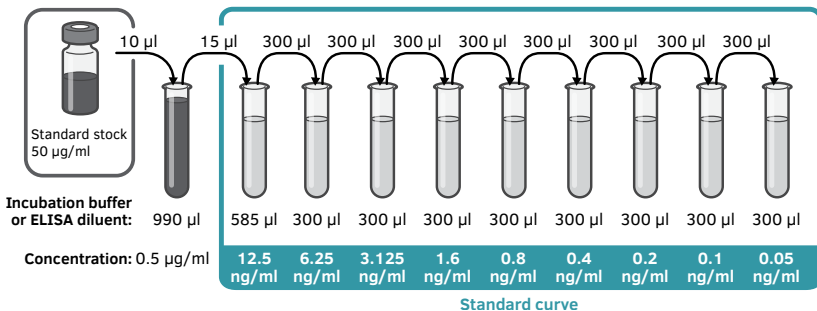
Analysis of serum/plasma requires the use of ELISA diluent (product code: 3652-D2). The ELISA diluent blocks heterophilic antibodies, commonly found in serum/plasma, from cross-linking the assay antibodies, thereby preventing false positive read-outs. The ELISA diluent should be used for dilution of standard, samples, and detection antibody.

## Reconstitution of ELISA standard

Reconstitute the ELISA standard to a stock solution of 50  $\mu\text{g}/\text{ml}$  by adding 0.5 ml of the standard reconstitution buffer. Allow the standard to dissolve for 5 minutes and mix thoroughly. The standard should be kept in aliquots at  $-20^\circ\text{C}$ . Avoid repeated freeze-thaw cycles.

## Preparation of standard curve

Prepare within 30 minutes of use. Volumes are sufficient for duplicates.



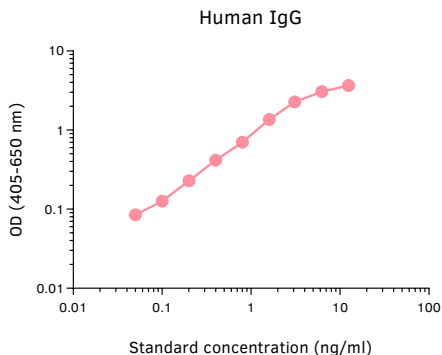
# Protocol

## Day 1

1. Add 100  $\mu\text{l}$ /well of capture mAbs MT145 diluted to 2  $\mu\text{g}/\text{ml}$  in PBS, pH 7.4. Use high protein binding ELISA plates. Incubate overnight at 4-8  $^{\circ}\text{C}$ .

## Day 2

2. Empty the plate and add 200  $\mu\text{l}$ /well of PBS with 0.05% Tween 20 and 0.1% BSA (incubation buffer) to block the plate. Incubate for 1 hour at room temperature.
3. Wash the plate 5 times with PBS containing 0.05% Tween 20 (300  $\mu\text{l}$ /well).
4. Add 100  $\mu\text{l}$ /well of samples or standards diluted in incubation buffer or ELISA diluent. Include assay background control, i.e. wells without standard. Incubate for 2 hours at room temperature.
5. Wash as above.
6. Add 100  $\mu\text{l}$ /well of detection mAb MT78-biotin diluted to 0.25  $\mu\text{g}/\text{ml}$  in incubation buffer or ELISA diluent. Incubate for 1 hour at room temperature.
7. Wash as above.
8. Add 100  $\mu\text{l}$ /well of Streptavidin-ALP diluted 1:1000 in incubation buffer. Incubate for 1 hour at room temperature.
9. Wash as above.
10. Add 100  $\mu\text{l}$ /well of pNPP substrate (product code: 3652-P10) and incubate the plate for approximately 60 minutes.
11. Measure the optical density in an ELISA reader at 405 nm. Preferably use a reader capable of subtracting a reference wavelength of between 570 and 650 nm. Representative standard curve shown below.



Developed and manufactured by MABTECH AB, Sweden, whose quality management system complies with the standards ISO 9001:2015 & ISO 13485:2016.



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**Mabtech AB (Head Office)**  
Sweden  
Tel: +46 8 716 27 00  
mabtech@mabtech.com

**Mabtech, Inc.**  
USA  
Tel: +1 513 871-4500  
mabtech.usa@mabtech.com