TNP COATED SURFACE

The Biomat product is a 96 well coated microplate with TNP (trinitrophenol) and a protein to block non-specific binding sites and to maintain stable activity.
Trinitrophenol is an hapten that can be attached to carrier proteins such as KLH (Keyhole Limpet Hemocyanin) or ovoalbumin; when this complex is injected into appropriate animal models, produce an immune response which is measured as changes in the levels of anti TNP IgM and IgG. Thanks to these changes, the researchers can assess the impact of pharmacologic or genetic manipulations on the studied immune system.

Example of applications:

- used as biomarkers of immunotoxicity
- assessing efficacy of vaccines, including dosage, adjuvantcy, route of immunization and timing
- determination of immune status relative to controls

Product specifications

Available configurations
96-well microplates with 12 removable 8-well strips.

Coating

TNP-BSA is coated using 100 µl/well. The strips are post-coated (blocked) for low non specific binding and long-term stability.

Binding capacity
Microplate was saturated with mouse IgG anti TNP at a concentration of 0.125 µg/ml (12.5 ng/well) in an ELISA format using goat anti mouse IgG-HRP as detector and TMB as substrate (see Technical notes 41 and Figure 1 and 2 for data and experiment details).
The Biomat TNP microplate shows a nominal **binding capacity of ~ 0.250 µg/ml of mouse IgG anti TNP**

Uniformity
Microplates show a **CV% less than 5** when used as a sandwich of mouse IgG anti TNP in an ELISA format using goat anti mouse IgG -HRP as detector and TMB as substrate.

Storage and Stability
The microplates, under the indicated storage conditions 2-8 °C, are stable until the expiration date printed on the label.
If opened, store in closed pouch with desiccant and use within the expiration date.

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Binding capacity test

1. Add 100 µl of different concentrations of monoclonal mouse IgG anti TNP, from 0.0005 to 0.500 µg/ml, diluted in Sample Diluent, (*Biomat* code 400-1-100) to the wells of TNP coated plate and incubate for 60 minutes at room temperature
2. Empty the wells and wash with Wash Buffer, (*Biomat* code 200-1-100) four times
3. Add 100 µl/well of goat anti-mouse IgG-HRP (*Jackson ImmunoResearch* code 115-035-003), diluted 1:25,000 in Diluent for HRP conjugate, (*Biomat* code 400-2-100) and incubate for 60 minutes at room temperature
4. Empty the wells and wash with Wash Buffer, (*Biomat* code 200-1-100) four times
5. Add 100 µl/well of TMB substrate solution, (*Biomat* code 500-1-100) and incubate 15 minutes at room temperature
6. Stop the substrate reaction by adding 100 µl /well of sulphuric acid, (*Biomat* code 600-1-100) and read the optical density values at 450 nm

The data show that a plateau has got starting with a mouse IgG anti TNP concentration of 0.250 µg/ml.

This concentration means the well binding capacity we can express as:

- µg/well = 0.0250 (25 ng/well)

Figure 1

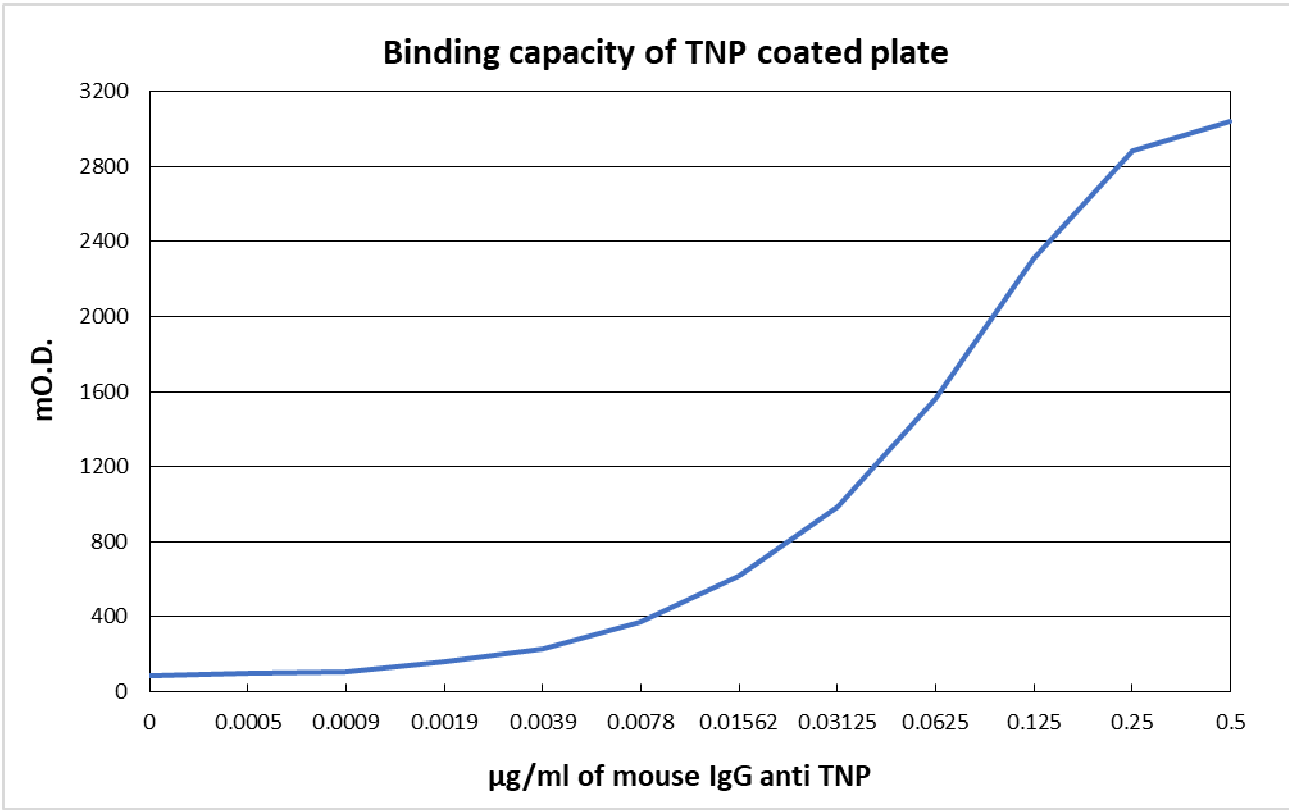

Figure 2: the figure gives an idea of the dilution factor to apply to the serum/plasma of the immunized mouse under evaluation; where k means a dilution of 1:1,000