

CATALOG NUMBER: RAG0021

RECOMBINANT ANTIGEN: outer membrane channel protein TolC (OMP50) of *Salmonella typhi* (Parkhill *et al.*, 2001)

DESCRIPTION: some of the antigenic regions of *Salmonella typhi* antigen OMP has been prepared as a recombinant antigen fused to a his-tag in its N-terminus.

PRESENTATION: dry powder (stabilized with 15% trehalose)

SOURCE: Escherichia coli

biotech

MOLECULAR WEIGHT: determined by SDS-PAGE, the protein band is between molecular markers of 35,000-25,000 Da, while relative molecular mass calculated from amino acid sequence is 32,080.30 Da.

BATCH COMPOSITION:

| COMPONENTS | COMPOSITION |
|-----------------------|------------------------------------|
| his-OMP | recombinant antigen with a his-tag |
| | in its N-terminus |
| Storage buffer before | 50 mM MES pH 6.5, 0.1 M KCl and |
| lyophilisation | 0.1% polyoxyethylene (10) tridecyl |
| | ether |

QUALITY CONTROL:

1. PROTEIN CONCENTRATION DETERMINED ESPECTROPHOTOMETRICALLY

DO₂₈₀ = 0.695 A _{0.1 %} (=1 g/l) = 0.543 CONCENTRATION*: 1.28 mg/ml

* The measurement of the protein concentration has been performed with the theoretical extinction coefficient of the recombinant protein obtained from Gill and vonHippel, 1989. It is recommended that the users carry out their absorbance determinations to avoid equipment variabilities regarding final concentration, mainly in reproducibility analysis.

2. PURITY CONTROL IN SDS-PAGE: 12%

kDa Mw OMP

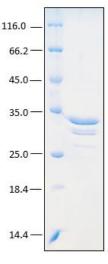


Figure 1. SDS-PAGE analysis (12%) of 2 μ l of recombinant OMP. Purity is > 90% as determined by gel electrophoresis. Bands slightly smaller correspond to the same protein.

LOT NUMBER:

3. POSITIVE CONTROL DETECTION BY AN ELISA ASSAY

The assay was performed by an "in-house" ELISA developed at Rekom Biotech. Each end user should carry out his own titration for a particular application.

The assay was performed with a goat positive control serum and a negative human serum. The value of the positive/negative ratio obtained is between 3-4.

4. ABSENCE OF PRECIPITATION AFTER A FREEZING AND THAWING CYCLE: ok

LOT SPECIFICATIONS:

1. RECONSTITUTION: with approx. 0.68 of sterile doubledistilled water, a final concentration of approx. 1.28 mg/ml will be obtained. The solubilisation of the cake should be developed for 15 min to allow a homogeneous protein solution, considering that part of the cake can be on the glass-walls of the container. Upon reconstitution, leave the solution at least 15 min homogenizing with a mild agitation at 4°C. Avoid vigorous shaking that can cause foaming and protein denaturation. With this reconstitution, the protein will be maintained at pH 6.5. It is recommended that the users carry out their absorbance determinations to avoid concentration variabilities due to the equipment used, mainly in reproducibility analysis.

2. TOTAL QUANTITY PER ALIQUOT: 1 mg

3. SUGGESTED TITER BY ELISA: up to 1:1,280, which corresponds to 1 μ g/ml of protein concentration in plates for IgG detection.

4. STORAGE: protein is shipped at room temperature. Upon arrival, it should be stored at 4°C or -20°C. Once reconstituted, it should be aliquoted in order to avoid repeated freezing and thawing cycles and stored at -20°C to -80°C.

5. TESTED APPLICATIONS: ELISA.

6. POSIBLE APPLICATIONS: WB, DB, Indirect ELISA, positive control in direct ELISA, CLIA, lateral-flow. Where this product has not been tested for use in a particular technique, this does not necessarily exclude its use in such procedures. Suggested working dilutions are given as a guide only. It is recommended that the user titrates.

7. OBSERVATIONS: proteins should be maintained frozen at high concentrations. The dilution to be performed for ELISA assays should be made with a small quantity of protein, the same day of the experiment. Once reconstituted and in order to defrost the protein, maintain the aliquot at 25°C without shaking to avoid aggregation. Prior making test dilutions and after defrosting the protein, is recommended to remove possible protein aggregates by centrifuging the stock solution, avoiding alterations in the immobilization of the biomolecule to the solid surface.

Recombinant antigen OMP for Salmonella typhi



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RELATED PRODUCTS:

Flagellin.

BIBLIOGRAPHY:

Parkhill et al. Complete genome sequence of a multiple drug resistant Salmonella enterica serovar Typhi CT18. 2001, Nature 413:848-52.

Gill SC, von Hippel PH. Calculation of protein extinction coefficients from amino acid sequence data. *Anal Biochem.* 1989 Nov 1;182(2):319-26.

Important Notes: During shipment, small volumes of product will occasionally become entrapped in the seal of the product vial. For products with volumes of 200 μ l or less, we recommend gently tapping the vial on a hard surface or briefly centrifuging the vial in a tabletop centrifuge to dislodge any liquid in the containers cap.

Although recombinant antigens are expressed in non-pathogenic *E. coli* and bacterial integrity is destroyed during purification, the antigen preparation should be handled as potentially infectious.

FOR RESEARCH AND COMMERCIAL USE IN VITRO: not for human in vivo or therapeutic use.



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