

YERSINIA PESTIS

A MONOCLONAL ANTIBODY THAT BINDS SURFACE NON-FRACTION 1 ANTIGENS OF *YERSINIA PESTIS*

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Most available diagnostic approaches for *Yersinia pestis* are based on the detection of fraction-1 antigen (F1), which is the dominant surface antigen expressed by the pathogen when it is grown at 37°C. A monoclonal antibody was developed based on the detection of non-F-1 surface antigen. F-1 deficient *Y. pestis* cells were induced by growing cultures in brain heart infusion broth adjusted to pH 5.5 with lactic acid and at 15°C for 7 days with 30 continuous weekly transfers under the same growth conditions. The loss of expression of F-1 by these cells was confirmed by an immunoblot of OMPs using a monoclonal antibody that specifically recognizes F-1. The F-1-deficient *Y. pestis* cells were used to immunize BALB/c mice from which a monoclonal antibody (IgG1) that specifically recognizes *Y. pestis*, with or without F-1, was obtained. This monoclonal antibody (6B5) did not cross-react with enteric bacteria, including *Y. enterocolitica*, *Salmonella* Typhimurium, *S. Enteritidis*, *Citrobacter freundii*, *Listeria monocytogenes*, *L. innocua*, *E. coli*, *E. coli* O157:H7, *E. coli* O111:NM, *E. coli* O26:H11, *Serratia marcescens*, *Vibrio cholerae*, *Klebsiella pneumoniae*, *K. oxytoca*, *Aerococcus viridans*, *Hafnia alvei*, and *Proteus hauseri*. ELISA results revealed that MAb 6B5 is specific for *Y. pestis*, with the exception of a minor cross-reaction with *Y. pseudotuberculosis*. Western immuno-blot analysis revealed that MAb 6B5 recognizes a *Y. pestis* OMP of ca. 30 Kb.

