

A Simple Method for the Production of anti-Fish IgM Monoclonal Antibodies

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Abstract

Immunoglobulin M (IgM) is the predominant antibody isotype in the serum of teleost fish and plays a central role in systemic immune responses. Monoclonal antibodies (mAbs) targeting fish IgM are essential tools for immunological research and disease management in aquaculture. However, conventional mAb production protocols require purified IgM, which is difficult to obtain due to the complexity of purification procedures in teleost species. This study introduces a simplified method for generating anti-IgM mAbs by immunizing mice with whole serum from rainbow trout (*Oncorhynchus mykiss*), thereby eliminating the need for IgM purification. To do this, hybridomas were screened using an indirect ELISA based on an unrelated antigen (e.g., maltose-binding protein) and trout sera containing antibodies against this antigen. This approach led to the identification of five stable hybridoma clones (1D4, 3D3, 3E3, 4B3, and 7F6). The resulting mAbs effectively recognized trout antibodies against *Infectious pancreatic necrosis virus*, *Lactococcus garvieae* bacterium and sheep red blood cells. Immunoblotting under reducing and non-reducing conditions confirmed the specificity of these mAbs for trout IgM. This method provides a practical and efficient alternative for producing functional monoclonal antibodies against fish IgM.

Key words: Monoclonal antibody, Trout, IgM, ELISA, Immunoblotting

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