

IVD_03 - Use of medium supplements to improve anti-MRSA mAb final concentration in hybridoma cell culture and reduce the cost production

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Introduction: MRSA (Methicillin Resistant *Staphylococcus aureus*) is one of the major causes of death by resistant bacteria in the whole world, due to the antibiotic's resistance mechanisms of *Staphylococcus aureus*, and its diagnostic is important to lay down a specific and the sooner the better treatment. A monoclonal antibody (mAb) anti-PBP2a, a MRSA surface protein, was developed in Bio-Manguinhos using the hybridoma technology. Bioprocesses could be extremely expensive when we talk those about monoclonal antibodies production. Those proteins are usually produced in cells, which needs properly conditions of microenvironment to act in an expression of these mAbs, as nutritional specific medium, e.g. A supplementation could be an alternative to make those cells produce more mAbs, showing an optimal cost benefit.

Objective: Introduce 6 different nutritional supplements in 3 different conditions, aim to evaluate viable cells/mL, cell viability and, mainly, the monoclonal antibody final concentration in 9 days of culturing.

Methodology: We performed 3 different experiments in order to enrich the medium and increase, mainly, the mAb amount, each one with a supplement concentration: 1g/L, 3,5g/L and 10g/L, solved in DMEM medium with 10% FBS (Fetal Bovine Serum) and 2mM L-Glutamine. These are 6 supplements, Cell Boosters (CB1, CB2, CB3, CB4, CB5 and CB6, from HyClone/Cytiva), which are chemically defined and free of animal proteins. The viable cells was determinate by trypan dye and the mAb concentration was quantified by Enzyme-Linked Immunosorbent Assay (ELISA).

Results: At the final of this study, we could demonstrate that our supplementation shows the increase of the anti-PBP2a mAb in the supernatant, expressed by our hybridoma cells. Without supplementation, the 9 days kinetics secreting anti-PBP2a shows an amount of 41,8ug/mL mAb. With a supplementation, the best one, with 3,5g/L of CB5, the 9 days kinetics shows an 130% increase of mAb concentration, with a final amount of 97,7ug/mL mAb.

Conclusion: We improve the mAb concentration with the supplements save by around, per liter, a considerable amount of R\$2.300,00 with a CB5 supplement. These experiments could be employed in the biopharmaceuticals production and reduce, significantly, the final cost of this manufacturing.

Keywords: Supplementation; MRSA; Monoclonal Antibodies