MONOCLONAL ANTI-OCHRATOXIN A ANTIBODY APPLIED IN DEVELOPMENT OF AFFI-GEL 10 BASED IMMUNOAFFINITY COLUMN

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ABSTRACT

The immunoaffinity column (IAC) based on monoclonal antibody (mAb) immobilized in gel provides simply and high specific tool for clean-up, as well as analyte concentration from complex matrix. This research proposed the optimization of IAC developed in-house applying anti-ochratoxin (OTA) mAb produced in different stages of Hybridoma OTA.1 cultivation, with modification and adaptation viabilized for local condition. The hybridoma was cultivated in RPMI 1640 medium supplemented with fetal bovine serum and L-glutamine, which was gradually replaced by Hybridoma-Serum Free Medium (H-SFM). Six experimental IACs were confectioned by performing the assay with 5 mg of mAb produced in following cultured condition: 75 % H-SFM (one IAC); mix of 75 and 100 % H-SFM (1:1; three IACs) and 100 % H-SFM (two IACs). Such mAbs were immobilized in one mL of Affi-gel 10, which was used as support. The immobilization rate ranged from 77.43±6.39 to 97.55±0.24 %. The IAC prepared with mAb produced in 75 % H-SFM showed higher performance concerning anti-OTA activity evaluated by indirect ELISA. This data indicated improving of previous IAC column prepared by our group, which showed immobilization rate of 86.43±1.65 %, and OTA retention rate of 72.08±1.41 %; this IAC was developed mixing mAb produced in 75 and 100 % H-SFM (1:1). The developed IAC could be a suitable tool destined for tracking OTA in food quality control, assuring the safety of Brazilian agricultural products in globalized world.

Key words: hybridoma, monoclonal antibody, ochratoxin, immunoaffinity column

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