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Development of Hybridoma for the Production of Monoclonal Antibody against Sulfamethazine

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Fifteen monoclonal antibodies(MAbs) against the widely used antibiotic sulfamethazine (SMZ) were produced by hybridomas from two fusion of myeloma cells and spleen cells isolated from BALB/c mice immunized with hapten(SMZ-hemisuccinate, SMZ-hemiglutarate conjugated with keyhole limpet hemocyanin(KLH) and soy bean trypsin inhibitor(STI). To evaluate sensitivity of MAbs, competitive indirect ELISA was used. MAb 1-H11-5 exhibited the highest sensitivity and selectivity toward sulfamethazine. A direct competitive ELISA was established with SMZ coupled to horseradish peroxidase as the labeled antigen. This assay was very sensitive and had a linear range from 0.1-10ng SMZ per ml sample. The developed antibody only showed 1.68% cross-reactivity to sulfamerazine but not with other analogues. Recoveries of SMZ added to milk samples at 1, 10, 100 ug/ml from 96 to 119%. This ELISA provided a valuable method to quantify SMZ in various food samples including milk.