P.9 Field-Portable Immunoassay Instruments and Reagents to Measure Ionic Uranium and Related Radionuclide Wastes

Robert Blake II
rblake@xula.edu; Tel. 504-483-7489
Xavier University of Louisiana
1 Drexel Drive
New Orleans, LA 70125

Diane A. Blake
blake@tulane.edu; Tel. 504-584-2478
Tulane University School of Medicine
1430 Tulane Avenue
New Orleans, LA 70112

Abstract

The development of sensors that use antibodies as the recognition element appeals to a large number of potential end-users, since these devices may be used to monitor a very broad range of analytes. The variety of molecules that can be quantified by immunosensors is virtually unlimited, and depends primarily upon the bnding affinities and specificities of the antibodies incorporated into the devices. The high sensitivity and selectivity of such sensors makes them attractive for situations where both speed and accuracy are required. Previous studies from our laboratories have demonstrated the feasibility of isolating monoclonal antibodies that recognize specific metal ions. The goals during the current grant period are: (1) to isolate and characterize antibodies that recognize the most mobile form of uranium, UO_2^{2+}; (2) to assemble, test, and validate a new field-portable immunosensor based on these antibodies and a hand-held flow fluorimeter; and (3) prepare new monoclonal antibodies to the primary uranium chelators (EDTA and DTPA) found in DoE wastes.

Three hybridoma cell lines were generated that synthesize and secrete monoclonal antibodies that bind tightly and specifically to UO_2^{2+} complexed to 2,9-dicarboxy-1,10-phenanthroline (DCP). Cloning and sequencing of the cDNAs that code for the light and heavy chain variable regions of these antibodies demonstrated that all three have distinct binding sites. Two antibodies (designated 8A11 and 12F6) were selected for further studies, based upon their affinity for the UO_2^{2+}-DCP complex and their resistance to changes in pH and ionic strength.

A prototype competitive immunoassay for UO_2^{2+} was developed that accurately monitored UO_2^{2+} at concentrations from 10 to 120 nM in buffers amended with 12.5 micromolar DCP. These assays were conducted on the KinExA, a semi-automated flow fluorimeter designed for sophisticated studies in the laboratory. As part of our effort to adapt these and related immunoassays for use in the field, an *alpha* version of a field-portable KinExA flow fluorimeter was developed for experimental purposes. Issues currently under investigation using this *alpha* unit include: (i) the effects of flow rates of liquid reagents on the accuracy and sensitivity of the immunoassay; (ii) the necessity for pre-treatment of the sample prior to analysis; (iii) practical limits on the volume of sample to be introduced into the field instrument; and (iv) the effects of lyophilization and reconstitution of disposable reagents on the performance characteristics of the assay. It is anticipated that these studies could generate a useful marketable product, a collection of portable field tests for uranium and related wastes that could be exploited both in government and private sectors.