Applied Surface Science
Room 210A - Session AS-MoA

SIMS II - Biological and Organic

Moderator: G. Gillen, National Institute of Standards and Technology

2:00pm **AS-MoA1 Single Photon Ionization of a Derivatized Peptide Covalently Bound to a Surface**, *P.D. Edirisinghe, S.S. Lateef, C.A. Crot,* ***L. Hanley,*** University of Illinois at Chicago*; J.F. Moore, W.F. Calaway, M.J. Pellin,* Argonne National Laboratory

Covalently bound peptides, proteins, and other biomolecules are widely used for microarrays, microfluidic channels, cell growth surfaces, and biosensors. Detection of these surface bound species by matrix assisted laser desorption ionization or secondary ion mass spectrometry (SIMS) is often complicated by low ionization yields and/or high fragmentation. Single photon ionization is one method that shows great promise for enhancing ionization yields with a minimum of fragmentation. The fluorine excimer laser is an intense laboratory source of vacuum ultraviolet radiation, but the 7.87 eV photons it generates are lower in energy than the ionization potential of many target species. A method is described here whereby derivatization of peptides with the Fmoc group allows efficient fluorine laser single photon ionization of the entire labeled peptide. Various Fmoc labeled peptides are covalently bound to oxidized Si(100) wafers via maleimide coupling to surface bound aminopropyltriethoxysilane. Physisorbed films of Fmoc labeled peptides, unlabelled peptides, and various amino acids are prepared by drying a solution thereof onto the silicon wafer. Both covalently bound and physisorbed peptides are analyzed by laser desorption photoionization (LDPI) mass spectrometry. Only the Fmoc labelled peptides form large ions identified as common peptide fragments bound to either Fmoc or the surface linker. Unlabelled peptides and amino acids do not form large representative ions. Electronic structure calculations performed with Gaussian 98 indicate the Fmoc label is behaving as an ionization tag for the entire peptide, lowering the ionization potential of the complex below the 7.87 eV photon energy. This method should allow detection of many molecular species covalently or electrostatically bound to surfaces.

2:20pm **AS-MoA2 ToF-SIMS Applications in the Analysis of DNA Microarrays**, ***K.K. Soni,*** Corning Incorporated

DNA arrays are typically prepared by microcontact printing of DNA on a glass surface that has been modified by an aminosilane. The DNA immobilization on the modified glass surface relies on direct contact between DNA and amine groups supplied by the silane layer. The presence of surfactants such as Triton even at trace level can completely inhibit attachment of DNA. Given the tendency of this molecule to segregate at interfaces, its detection by surface sensitive techniques such as ToF-SIMS is facilitated; in other words, the sample preparation method and ToF-SIMS analysis constitute a very sensitive analytical procedure to detect trace levels of surfactant molecules in DNA preparations. To further corroborate the impact of surfactants, pure DNA solutions were intentionally contaminated with Triton X-100 in varying concentrations. The resulting mixtures were used for printing on modified glass slides. It was demonstrated that the presence of Triton reduces the amount of immobilized DNA and above a certain concentration (10-100 ppm by volume) can completely inhibit DNA printing. In another application of ToF-SIMS, we demonstrate the ability to study fluid flow and evaporation dynamics in a drying droplet in microarrays. Evaporation behavior of the drop strongly influences the solute transport and hence the uniformity of the dot. We have utilized ToF-SIMS analysis of printed DNA dots to determined the final distribution of the solute by mapping the sodium concentration in the drop. Three different kinds of effects were observed: the first kind having higher concentration of solute on the outer edge of the drop (rim effect); the second kind having higher concentration at the center of the drop (pinprick) and the third having uniform distribution.

2:40pm **AS-MoA3 Cell Imaging, DNA Diagnostics, Protein Analysis: Mass Spectrometric Characterization of Biological Surfaces**, ***H.F. Arlinghaus,*** Physikalisches Institut der Universität Münster, Germany **INVITED**

We have used time-of-flight secondary ion mass spectrometry (TOF-SIMS) and laser postionization secondary neutral mass spectrometry (Laser-SNMS) to analyze various biological surfaces. Both techniques use a focused energetic primary ion beam for bombarding a solid sample, and a mass spectrometer for analysis. But unlike SIMS, which analyzes only the sputtered secondary ions, Laser-SNMS uses laser beams to either resonantly or non-resonantly ionize the majority of sputtered neutral particles. In our presentation, we will compare and discuss the salient characteristics of the TOF-SIMS and Laser-SNMS techniques and will show applications in the following fields: (a) imaging and quantifying target-specific drug delivery systems as well as intrinsic elements and molecules in single cells with subcellular resolution in vitro, i.e. in cell cultures, and in vivo, i.e. in tissues, (b) investigation of the immobilization process of PNA and the influence of length and type of spacer molecules on the efficiency of hybridizing DNA to PNA biosensor chips and investigation of its use for DNA diagnostics with unlabeled DNA, (c) detection of proteins in cells, and (d) investigation of yield behavior and fragmentation patterns using different primary ions (Ar@super +@, Xe@super +@, SF@sub 5@@super +@, Au@super +@, Au@sub n@@super +@) for increasing efficiency and sensitivity in cell and DNA diagnostics. Furthermore, we will discuss current instrumental developments, particularly in regard to 3D molecular imaging with nanometer-scale resolution. We will show that TOF-SIMS and Laser-SNMS are well suited for imaging and quantifying trace element and molecule concentrations in biological materials with very high efficiency and nanometer-scale resolution. In particular, TOF-SIMS has the potential for providing a new method for rapid unlabeled DNA diagnostics, and its high detection efficiency makes this technique especially useful for directly analyzing genomic DNA.

3:20pm **AS-MoA5 Molecular Depth Profiling of Polymer Multilayers using a Polyatomic Primary Ion Beam**, ***M.S. Wagner,*** National Institute of Standards and Technology

Obtaining characteristic molecular information during the secondary ion mass spectrometry (SIMS) depth profiling of polymers has been severely limited due to primary ion-induced sample damage when using monatomic primary ions. Polyatomic primary ions have shown promise for the molecular depth profiling of thin (< 250 nm) polymer films due to their low penetration depth and high sputter rates. In this study, dual-beam time-of-flight SIMS (sputter ion = 5 keV SF@sub 5@@super +@, analysis ion = 10 keV Ar@super +@) was used to depth profile spin-cast multilayers of poly(methyl methacrylate), PMMA, poly(hydroxyethyl methacrylate), PHEMA, and trifluoroacetic anhydride-derivatized PHEMA, TFAA-PHEMA, on silicon substrates. Despite extended SF@sub 5@@super +@ bombardment (> 5 x 10@super 14@ ions/cm@super 2@), characteristic pendant-group-related positive and negative secondary ions of the different polymer layers were observed as a function of depth during the depth profiles. The sputter rates of the polymers in the multilayers typically were lower than corresponding single layer films, with the ion-induced damage accumulation rate of the outermost polymer layer affecting the sputter rate of the underlying layers. Due to its higher ion-induced damage accumulation rate, PHEMA lowered the sputter rates for underlying PMMA or TFAA-PHEMA layers. Similarly, PMMA reduced the sputter rate for underlying TFAA-PHEMA layers. Typical interface widths between adjacent polymer layers were 10-15 nm for the bilayer polymer films; however, the layer order significantly impacted the interface widths for trilayer films. The interface widths in the trilayer films increased with depth to ~ 35 nm, showing the formation of sputter-induced surface roughness during depth profiling of these films. This study demonstrates the utility of polyatomic primary ions for molecular depth profiling and presents new opportunities for the characterization of thin polymer films.

3:40pm **AS-MoA6 Applications of Cluster SIMS for Molecular Depth Profiling in Biomaterial Systems**, ***C.M. Mahoney,*** National Institute of Standards and Technology*; J.-X. Yu, J.A. Gardella, Jr.,* State University of New York at Buffalo*; A.M. Johnson, R. Langer,* Massachusetts Institute of Technology

Polymeric biomaterials have numerous clinical applications including as surgical implants, absorbable sutures, tissue engineering scaffolds and drug delivery devices. Time-of-Flight Secondary Ion Mass Spectrometry (TOF-SIMS) has proven to be particularly useful in the surface analysis of these polymeric biomaterials@super 1@. However, much of this work has been done with monoatomic primary ion beams, which have a large amount of beam-induced damage associated with them. This precludes the ability to obtain in-depth information from organic samples. Compared to monoatomic ion bombardment, cluster primary ion beams, such as SF@sub 5@@super +@ have resulted in decreased subsurface damage and increased sputter rates in some cases, allowing the ability to depth profile in organic and polymeric materials for the first time@super 2-3@. This talk will briefly describe the ongoing research efforts at NIST to further develop cluster SIMS as a tool for biomaterials characterization. We have already shown the ability to depth profile in model polylactic acid (PLA) based drug delivery systems using cluster SIMS@super 3@. More recently, we have been able to successfully measure the extent of preferential segregation in polylactic acid / polyethylene glycol (PLA/PEG) blends as well as determine

the in-depth distribution of acetamidophenol doped PLA films as a function of increasing degradation time. We have also successfully obtained information as a function of depth in a novel drug delivery microchip. This work further demonstrates the increasing utility of cluster SIMS for biomaterials applications. @FootnoteText@ @footnote 1@ Lee, J.-W.; Gardella, J.A. Jr. Analytical Chemistry 75 (2003) 2950-2958.@footnote 2@ Gillen, G.; Roberson, S.; Rapid Commun. Mass Spectrom. 12 (1998) 1303.@footnote 3@ Mahoney, C.M.; Roberson, S.V.; Gillen, J.G. in "Depth Profiling of 4-Acetamidophenol Doped Poly(lactic Acid) Films Using Cluster SIMS"; Analytical Chemistry, Accepted March 2004.

4:00pm **AS-MoA7 Improved ToF-SIMS Ion Yields and Cationization of Water-Soluble Analytes by Polyelectrolyte Multilayers**, ***Y.-Y. Lua***, *C.A. Pew,* Brigham Young University*; A. Schnieders,* ION-TOF USA, Inc.*; P.B. Savage, R.C. Davis, M.R. Linford,* Brigham Young University

Arguably one of the most important issues that has faced time-of-flight secondary ion mass spectrometry (ToF-SIMS) since its inception more than 30 years ago is the need for improved ion yields from analytes. Here we describe an entirely new method for improving ion yields and cationizing analytes that is particularly effective for charged, water-soluble species. This approach takes advantage of the highly charged, ionic nature of polyelectrolytes and the ease with which they can be deposited onto surfaces by the well known layer-by-layer method. In particular, we show that after an ultrathin film (ca. 0.5 nm) of a polycation (polydiallyldimethylammonium chloride, PDADMAC) spontaneously adsorbs onto a silicon (oxide) surface, a mixture of a polyanion (poly(sodium 4-styrenesulfonate)) and a water-soluble analyte, which contains one or more basic nitrogen atoms, will adsorb to form a second layer (ca. 1.5 nm thick). ToF-SIMS of this bilayer shows a significant enhancement in quasi-molecular analyte ion yield (roughly a ten-fold increase in signal), compared to that of the pure compound, or the compound dissolved in dilute HCl and dried on a surface. This phenomenon is demonstrated for two large organic macrocycles (m/z 672 and 745), and a smaller aromatic compound (acridine, m/z 179). Similarly, a significant enhancement in the ion yield of the quasi-molecular ion of 9-anthracenecarboxylic acid (m/z 222) is observed when it spontaneously deposits with PDADMAC to form a ca. 0.5 nm film on silicon.

4:20pm **AS-MoA8 Three-Dimensional Reconstruction of Elemental Distributions from TOF-SIMS Image Depth Profiles**, ***S.R. Bryan***, *D.G. Watson,* Physical Electronics USA

Time-of-flight Secondary Ion Mass Spectrometery (TOF-SIMS) is a powerful techique for image the distribution of elements and organic molecules on surfaces with with spatial resolution down to 100 nm. When combined with sputter depth profiling, TOF-SIMS can characterize the 3-dimensional distribution of all elements in the near surface region of materials. Due to the parallel detection nature of the TOF-SIMS technique, the full 3-dimentional data can be acquired for all elements in a reasonable amount of time. The challenge is to effectively display the tremendous amount of information generated in an image depth profile. The use of one or two cross-section images does not adequetly display the 3-dimensional distributions. In this work, we have applied methods developed in the medical field for CT and MRI imaging to TOF-SIMS data. Through the use of isosurface reconstruction and translucent display, the full 3-dimensional distribution of multiple elements can be viewed simultaneously.

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