

GENERAL PHYSICS

I. MOLECULE MICROSCOPY

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1. SCANNING DESORPTION MOLECULE MICROSCOPY

National Institutes of Health (Grant 1 R01 GM23678-02)

Jeffrey G. Yorker, John G. King

The purpose of SDMM is to study the spatial variations in the adsorption characteristics of various small molecules on biological surfaces. These characteristics include the density of binding sites for molecules on the surfaces and the binding energy at these sites. By using different molecules, one can infer the local composition of a surface. The energies involved in adsorption and desorption are small, making SDMM a useful tool for nondestructive probing of biological surfaces in vacuum.

In our experiments, the sample is held at an appropriate low temperature and coated with a layer of molecules from a molecular beam. Molecules are then thermally desorbed from the samples with spatial resolution by an array of controlled microheaters on which the sample rests. The desorbed molecules are ionized by electron bombardment, filtered according to mass by an RF quadrupole mass filter, and amplified by an electron multiplier.

During 1979 we completed a prototype microheater array, or thermal desorption array (TDA). The TDA is an 8 x 8 array of 64 10-square micron npn transistors fabricated in the M.I.T. Microelectronics Laboratory. Preliminary tests show that we can melt spots of ice on the TDA surface as desired, and that the thermal time constants are in the right range ($\sim 10^{-4}$ sec). We have also completed a suitable vacuum system with a liquid nitrogen trap that surrounds the detector and reduces water background at mass 18 by a factor of 30. We have also developed a novel

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differentially pumped vacuum lock for moving the sample in and out of the high vacuum region without admitting air. This device consists of a large, flat stainless-steel plate which slides over a series of three concentric circular vacuum locks separated by teflon vacuum seals.

We are now starting tests of the entire system, the results of which will guide the design of a refined TDA with 10^4 $1\text{-}\mu$ elements to be developed elsewhere. Such a device, the analog of a glass slide for an optical microscope or a grid for an electron microscope, will make useful application of the SDMM possible.

Mr. R. Fastow (Course VIII senior), working in the new integrated Circuit Facility, has constructed a new TDA. It consists of blocks of chromium $2\ \mu\text{m}$ on a side and $0.5\ \mu\text{m}$ thick on a silicon wafer which is etched to $0.2\ \mu\text{m}$ in the sample region. The chromium blocks are heated separately in sequence by a focused electron beam, and the heat conducted through the silicon produces desorption from the sample. This method appears to be best adapted to obtaining high spatial resolution.

2. CELL-SURFACE STUDIES

Health Sciences Fund

National Institutes of Health (Grant 1 R01 GM23678-02)

Douglas J. Ely, John G. King

New sample holders consisting of 10 evaporated chromium thin-film heaters on a ground Vycor microscope slide have been constructed, and desorption of water from them observed. Recording equipment has been assembled, and both cell-desorption studies and computation of a catalog of stain and surface combinations can now be resumed with much greater efficiency.

3. SCANNING MICROPIPETTE MOLECULE MICROSCOPE

Health Sciences Fund

Joseph A. Jarrell, John G. King

The instrument has been modified to permit the detection of labelled water by the addition of an in-line uranium reduction furnace. Resolution has been demonstrated to be better than $3\ \mu\text{m}$ with a $2\text{-}\mu\text{m}$ probe tip, and with a $5\text{-}\mu\text{m}$ probe tip

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concentration differences of deuterated water of 0.1% can be detected with a signal-to-noise ratio of ~ 10 and a time constant of ~ 1 sec.

The instrument is currently being applied, in collaboration with Drs. John Mills and Alex Leaf at Massachusetts General Hospital, to the study of transepithelial water fluxes across frog skin sweat glands and toad urinary bladder, respectively, and model tissues for the study of cystic fibrosis and kidney function.

4. PATHWAYS OF TRANSEPITHELIAL WATER FLOW

National Institutes of Health (Grant 1 R01 AM25535-01)

Stanley J. Rosenthal, John G. King, Alvin Essig

Our collaborative project with Dr. A. Essig of the Department of Physiology at Boston University Medical Center began with the development of the molecule flux apparatus which made possible simultaneous measurement of CO_2 production and O_2 uptake without spatial resolution. After obtaining some preliminary results this crude apparatus (built from discarded apparatus) was shelved in favor of another SMMM not unlike that mentioned above (section 3) to be developed and applied to problems of water flow in tissue.

Despite intensive study, disagreement persists concerning the pathways of transepithelial water flow. This problem will be approached by use of mass spectrometry, adapted to provide spatial resolution. The high sensitivity provided by this technique should permit sampling of the concentration profile of water tracer in the "unstirred" layer adjacent to an epithelial membrane, allowing evaluation of the relative magnitude of transjunctional and transcellular water flux at the luminal surface.

"High-impedance" probes will be prepared from micropipettes of 2- μm outer diameter, plugged with dimethyl silicone to permit diffusive flow of water into the vacuum chamber of a mass spectrometer without tissue damage. The application of H_2O^{18} to the "serosal" surface of a membrane will establish a concentration profile of tracer water at the "luminal" surface, conforming to the pathways of transmembrane water flow. Initial studies employing Nuclepore, synthetic membranes with well-characterized pore size, will define the degree of spatial resolution available and

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optimal operating conditions. Subsequent experiments will be carried out in Necturus gall bladder, a well-studied loose epithelium with large cells and other desirable characteristics facilitating its study. Studies will be done in the presence and absence of isotonic fluid transport, during inward and outward osmotic flow, and in the presence of various agents influencing water transport.