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STUDIES ON GUEST SELECTIVE MOLECULAR RECOGNITION ON AN OCTADECYL SILYLATED SILICON SURFACE USING ELLIPSOMETRY

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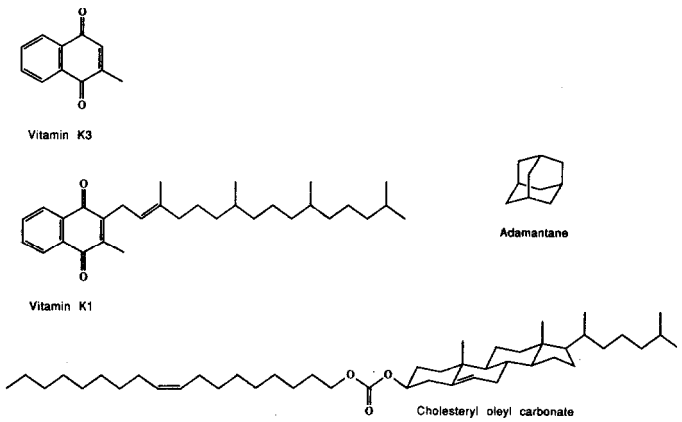
Application of the optical surface method ellipsometry to guest selective recognition is described. Detection of the affinity binding by guest recognition to an octadecylsilane derivatized silicon surface with vitamin K1 is exemplified with the technique.

The concept of guest selective recognition is particularly suitable for combination with sensor technology. In spite of this, relatively few efforts have been made to exploit the possibilities of using bioorganic molecular structures for construction of specific sensors in the same way as has been made with, for example, biosensors. To our knowledge only cyclic voltammetry has previously been studied[1]. This communication deals with ellipsometry, an optical surface method which permits determination of the thickness of thin organic layers covering a metallic surface by monitoring the change of light's polarization state when reflected upon the layers. An ellipsometer can thereby determine the mean thickness of a submonolayer in the range of 1-10 Å on an area of approximately 1 mm^2.

In order to obtain a selective surface, octadecylsilane was covalently bound to a hydrophilic silicon wafer in the presence of hexadecane as a guest template[2]. The wafer was soaked in a solution of 0.63 mM octadecyltrichlorosilane (ODS) dissolved in hexadecane/chloroform/tetrachloromethane (8: 0.8: 1.2 v/v/v) for 10 min[3]. Subsequently, the thickness of the formed ODS-hexadecane monolayer was estimated with a null-ellipsometer to be 29.4 ± 1.2 Å (Fig. 1)[4]. The estimation was made with an assumed refractive index (n_F) of the monolayer film of 1.450. This value has previously been obtained for thin Langmuir-films of C_{18}-fatty acids in other ellipsometric studies[5]. Minor deviations in the refractive index value in the range 1.40-1.60 caused by density variations, would not change the estimated thickness more than ± 2.4 Å. We therefore considered the assumed refractive index to be correct. Moreover, the experimental limitation of using a constant refractive index could easily be overcome by using a more sensitive null-ellipsometer together with a modified calculation model of the type described in reference 5.

The guest template, hexadecane, was removed by rinsing three times with chloroform, resulting in a reduction of the layer thickness to 15.2 ± 0.7 Å. This decrease can partly be ascribed to a lower refractive index due to a change in density of the film. The thickness value should therefore be regarded as a parameter of the number of molecules on the surface rather than physical thickness. When treating the bare silicon surface with an ODS-free hexadecane/$CHCl_3$/CCl_4-solution the increase in thickness was less than 1 Å. The removal of

hexadecane rendered the surface active for affinity binding of suitable molecular structures. The following guest molecules were tested for binding to the active surface:



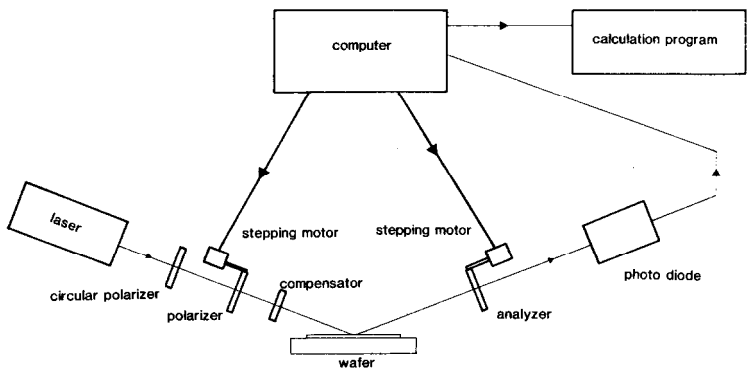


Fig 1. Ellipsometer configuration (See also reference 4).

Vitamin K1 and oleylcholesterol could both be expected to bind to the aliphatic octadecyl chain through their hydrophobic tails. A limited interaction with vitamin K3 and adamantane could possibly take place due to their general hydrophobic character, but much less pronounced than for the `tail´ binding molecules.

ODS-wafers were soaked in aqueous methanol solutions (2:3 v/v) of the four guest molecules at a concentration of 3.0 μM. After 10 min of treatment the thicknesses on the wafers were estimated from six measurements on three adjacent spots on each wafer. Table I shows the mean values obtained.

TABLE I. Mean Values of Guest Molecule Thickness on a Host Surface

Guest Molecule	Concentration (μM)	Δd_a (Å)	Δd_d (Å)
Vitamin K1	3.0	4.12 ± 0.9	5.40 ± 0.9
Vitamin K3	3.0	1.27 ± 0.8	0.78 ± 0.5
Adamantane	3.0	1.15 ± 0.6	0.97 ± 0.5
Oleylcholesterol	3.0	1.49 ± 0.5	1.20 ± 0.5

The guest molecules were removed from the ODS-surface by repeated chloroform rinsing whereby the initial layer thickness was restored. At a concentration of 3.0 μM only vitamin K1 showed a marked recognition ability. A control experiment with the MeOH/water solution resulted in an increase of 1.0 Å of the layer. Thus, the increase in thickness of approximately 1 Å obtained with the other guest molecules is probably caused by non-specific interaction with the aqueous methanol. In another control experiment a wafer was silanized for 20 min, resulting in a thicker ODS-layer (29.5 Å) with 70 % less capacity for binding of vitamin K1.

The dependence of the concentration of vitamin K1 on the thickness of the layer obtained was determined in the range 0.75 to 9 μM K1. Figure 2 shows the change in thickness with the concentration for both adsorption (Δd_a), and for desorption with chloroform (Δd_d)(n = 6). From the Langmuir adsorption isotherm the saturation level was calculated to be 11.8 Å. This value indicates a ratio of 0.61 vitamin K1 per hexadecane molecule at the surface. Thus, the ODS-layer appears to have a high degree of availability for vitamin K1. Calculation of the association constant (K) for vitamin K1 according to the Langmuir adsorption isotherm, $k_a C N (1-\theta) = k_d N \theta$, with the assumption of a total number of site of

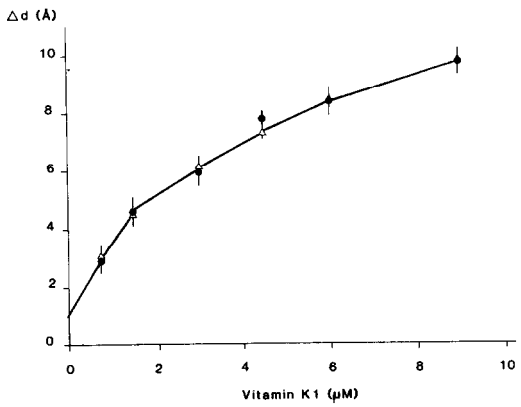


Fig. 2. Change in thickness versus concentration of vitamin K1

$N = d A \varsigma / \theta M$, gave a value of 2.8 10^5 dm^3/mole and 1.9 sites/100 $Å^2$. These values for K and N are of the same order as calculated by Tabushi et al.[1].

Ellipsometry, sometimes described as a semiquantitative method, seems here to be sufficiently reproducible to approach quantitative determination of the types of compounds investigated. The ellipsometric measurement principle therefore also appears to have potential as a sensor based on a guest selective surface in the same way as has been shown with voltammetry[1]. Moreover, the results show that silicon chemistry is suitable for formation of specific surfaces with host-guest recognition properties and is probably also applicable to other related optical methods such as reflectometry and internal reflection[6,7].

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REFERENCES AND NOTES

1. (a) K. Yamamura, H. Hatakeyama, K. Naka, I. Tabushi, K. Kurihara, J. Chem. Soc. Commun. 1092 (1988).
   (b) I. Tabushi, K. Kurihara, K. Naka, K. Yamamura, H. Hatakeyama, Tetrahedron Lett. 28, 4299 (1987).
2. Silicon wafer was made hydrophilic according to a procedure described by C.F. Mandenius, S. Welin, I. Lundström, K. Mosbach in Methods in Enzymology 137, p. 388 (1988), Academic Press, Inc.
3. J. Sagiv, Proc. Natl. Acad. Sci. (U.S.A.) 102, 92 (1980).
4. Measurements were carried out with null-ellipsometer from Rudolph Res. Fairfield, N.J. (Model Auto El II) and a computer program for ellipsometric calculation using a complex refractive index for the Si/SiO_2-surface of N = 3.86 - i0.1, a film index of 1.45, and an ambient index of 1.0.
5. P. Cuypers, J.W. Cosal, M.P. Janssen, J.M.M. Kop, W.T. Hermens, H.C. Hemker, J. Biol. Chem. 258, 2426 (1983).
6. H. Arwin, I. Lundström, Anal. Biochem. 145, 106 (1985).
7. M. Seifert, K. Tiefenthaler, K. Heuberger, W. Lukosz, K. Mosbach, Anal. Lett. 19, 205 (1986).

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