

Phosphonate molecular layers on TiO₂ surfaces

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Abstract. We investigated the deposition of (HO)₂-PO-S-(CH₂)₁₅-COOH on TiO₂. The phosphonate group has been chosen to serve as the anchoring moiety while the carboxylic group can be exploited for further coupling with antibacterial peptides. Native oxide-covered polished Ti samples were used as substrates for the molecular deposition. Previous to deposition, the substrates were plasma-cleaned in order to enrich their surfaces of hydroxyl groups, suitable for phosphonate coupling. The combined analysis of atomic force microscopy, X-ray photoemission spectroscopy and differential spectroscopic ellipsometry measurements indicates the formation of a submonolayer film made of irregular islands, one molecule thick.

1 Introduction

Titanium dioxide films are currently the object of intense investigation because of their wide range of applications, from photocatalysis, to photovoltaics and biomaterial development. In view of biomaterial development a crucial topic to be investigated is the interaction of the TiO₂ surface with organic and biological molecules. In particular, surface functionalization able to improve the antimicrobial and osteointegration properties of the TiO₂ surface is of large interest for implantology. Phosphonic acids [R-PO₃H₂] and their phosphonate ester derivatives [RPO₃R₂] are attractive anchoring groups for metal oxide surfaces [1-7].

In this work we investigated the deposition of a molecule bearing a phosphonate group at one end -the head- and a carboxylic group at the other end - the tail. We chose the carboxylic group as free tail since it can form amide ligation with the amine functions of biomolecules. In particular, we plan to couple antimicrobial peptides to the phosphonate layer in order to confer antibacterial properties to the functionalized TiO₂ surface.

2 Materials and Methods

Plates (10 mm x 10 mm x 1 mm) cut from a Ti sheet (99.6% purity, Advent Research Materials) were used as substrates. Before immersion in the phosphonate solution, the substrates were polished with SiC paper up to P4000 grit, then sonicated in acetone, ethanol and Milli-Q water.

Since surface bound-OH groups can serve as anchoring points for the formation of densely packed monolayers, some substrates were treated in an oxygen-plasma cleaner before immersion in the self-assembly solution in order to activate surface -OH groups. The properties of sonicated and plasma treated substrates were compared.

(HO)₂-PO-S-(CH₂)₁₅-COOH (ProChimia Surfaces, Poland), referred to as P-COOH in the following, was dissolved in 1,4-dioxane to a final concentration of 0.2 mg/ml. For molecular deposition, substrates were kept in the phosphonate solutions for 48 h at room temperature in the dark and subsequently rinsed in pure solvent.

Samples were characterized by Atomic Force Microscopy (AFM), X-ray Photoemission Spectroscopy (XPS) and Spectroscopic Ellipsometry (SE).

Tapping mode AFM measurements were performed using a Multimode/Nanoscope V system (Bruker) and Si cantilevers (OMCLAC160TS, Olympus).

The chemical analysis of the sample surfaces was carried out by XPS measurements using a 5600 Multi-Technique apparatus. An X-ray Al-monochromatized source (hν = 1486.6 eV) was used. The binding energy scale was referenced by setting the C1s of adventitious carbon at 284.8 eV. The data have been deconvoluted after subtraction of a Shirley background.

SE measurements were performed using a rotating compensator ellipsometer (M-2000, J.A. Woollam Co. Inc.). The variations induced in the Δ spectra by the formation of an ultrathin layer are usually small but they can be highlighted using the difference spectra (for brevity δ-spectra) formalism. In practice differences between film-covered sample spectra and substrate ones

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($\delta\Delta = \Delta_{\text{film}} - \Delta_{\text{substrate}}$) taken in the same ambient are considered [8,9].

3 Results and Discussion

As a first step we investigated the effect of plasma cleaning treatment on the surface chemical composition of TiO₂ polished substrates. XPS analysis indicates that oxygen plasma produces several changes in the surface composition of the TiO₂ substrates. The decrease in C1s signal intensity and the increase in Ti2p signal intensity (data not shown) indicate that oxygen plasma removes some adventitious carbon. Moreover, plasma treatment modifies the O1s signal line-shape. Even plasma cleaning duration affects the surface chemical composition. Fig.1 shows that increasing the plasma cleaning time results in an increase of the O1s components in the region 531-533 eV. This spectral range includes the contribution of the hydroxyl group [10] which should covalently bind the organophosphorous molecules. We therefore decided to treat TiO₂ substrates in a plasma cleaner for 45 min before depositing P-COOH.

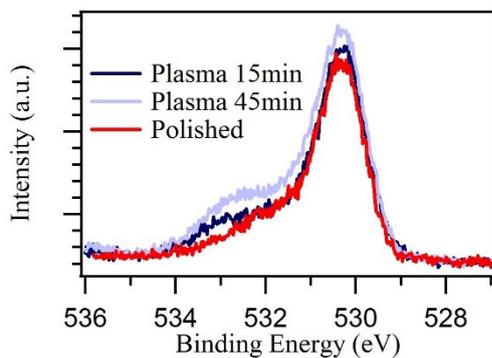


Fig. 1. XPS spectra of the O1s core level region of a polished TiO₂ surface as a function of the plasma cleaning time.

The analysis of AFM images indicates that oxygen plasma cleaning does not change the surface morphology significantly. At large scan size (several μm) polishing scratches can be observed (data not shown), but at smaller scales the surface morphology is relatively smooth and allows for monitoring morphological changes induced by molecular adsorption (see Fig.2).

AFM images taken after P-COOH deposition (Fig.2b) show significant changes compared to plasma cleaned substrates (Fig.2a). Irregular, partially coalesced islands form on the surface upon deposition. The average island height is around 1.5 nm, in agreement with the formation of monomolecular islands.

SE allows for a non-destructive fast check of the layer deposition. Fig.3a shows the Δ curves obtained before (blue line) and after (red line) P-COOH deposition. Preliminary simulations (not shown) suggest that the dip between 300-350 nm is related to the TiO₂ electronic gap and the position of the minimum depends also on the film thickness which can be estimated of the order of 10 nm. Simulations also indicate that the deposition of an

ultrathin transparent layer on TiO₂ induces a decrease of the Δ values; in particular the decrease of the Δ values in the infrared region is roughly proportional to the thickness of the deposited film. Assuming a reasonable value for the refraction index of ~ 1.5 , the decrease in Δ (see the $\delta\Delta$ spectrum in Fig.3b) fairly corresponds to the formation of a monomolecular layer. The tiny shift in the position of the Δ minimum upon deposition which produces the dip in the $\delta\Delta$ curve is also due to total film thickness increase upon P-COOH deposition. Such a dip must not be confused with a molecular absorption as observed, e.g., for cytochrome *c* monolayers on SiO₂ [11] or on gold substrates [9].

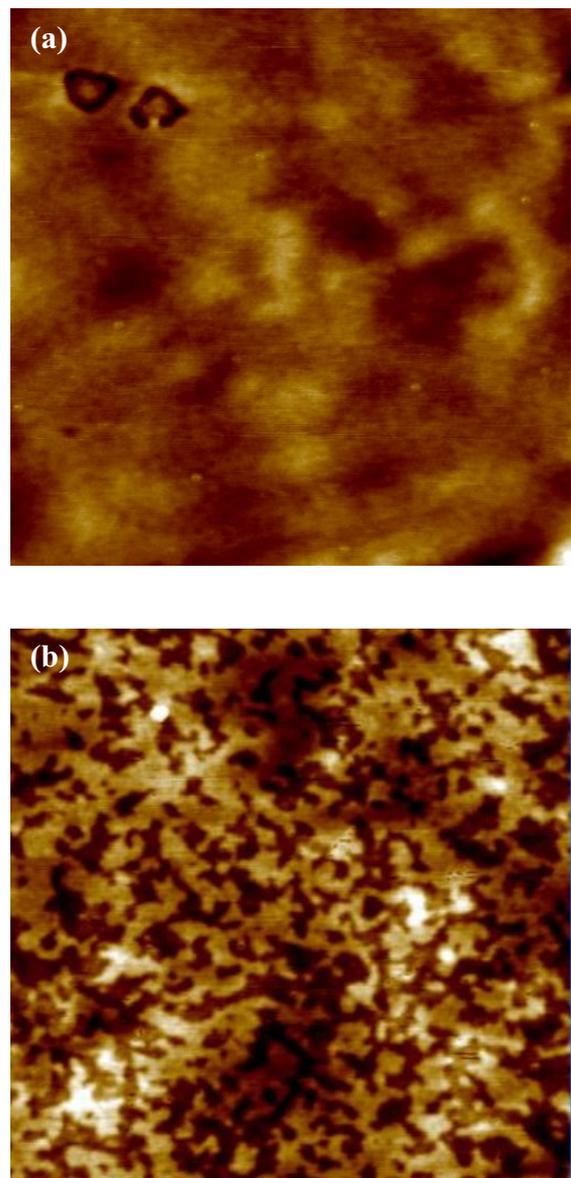


Fig. 2. Tapping mode AFM images of a plasma cleaned, polished TiO₂ surface, before (a) and after (b) P-COOH deposition. Scan size: 700 nm, z scale: 5 nm.

In order to gather more information about the nature of the islands, we employed XPS analysis to investigate the elemental composition of the surface upon P-COOH

deposition. Fig.4 shows the XPS spectra of the (a) P2p, (b) S2p, (c) C1s and (d) O1s core level regions of a P-COOH layer deposited on plasma cleaned TiO₂.

The P2p signal is modelled with a single 2p doublet using two Voigt functions with a branching ratio of 1:2 and a spin orbit splitting of 0.86 eV. The 2p_{3/2} component is found at (133.6±0.2) eV as expected for the phosphonate group [10].

In a similar way, the S2p spectrum can be deconvoluted with a single doublet, with a branching ratio of 1:2 and a spin orbit splitting of 1.2 eV. The BE of the 2p_{3/2} component is found at (163.5±0.2) eV, in agreement with the S signal of physisorbed organosulphur compounds [12]. The presence of the S atom in the alkylic chain provides a further chemical signature of the molecular deposition.

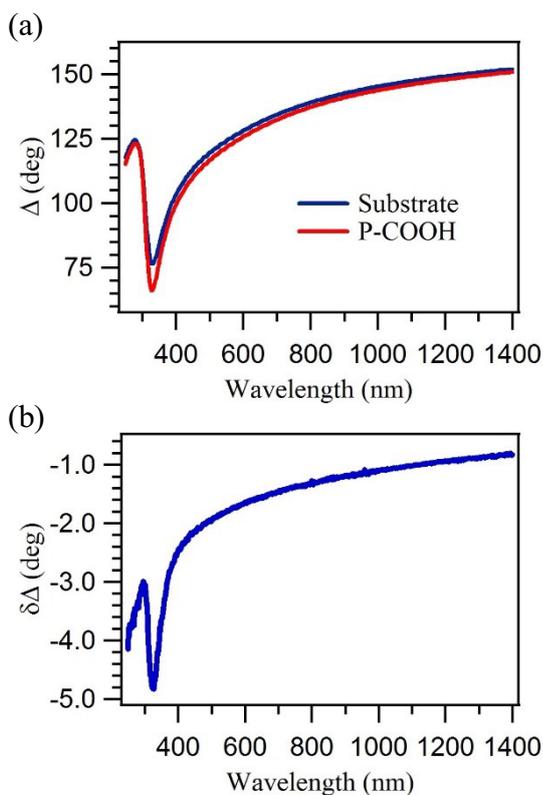


Fig. 3. (a) SE Δ spectra obtained on a TiO₂ plasma-cleaned substrate before (blue line) and after (red line) P-COOH deposition; (b) $\delta\Delta$ difference spectrum obtained as $\Delta_{\text{after}} - \Delta_{\text{before}}$

The C1s signal exhibits a main component with a shoulder on the high BE side and a second weak and broad signal at higher BEs. Three Voigt functions with the same FWHM have been fitted to the data. The main component, C1, at BE (284.8±0.2) eV is due to the alkylic chains and contains also the contribution of adventitious carbon. The second component, C2, at (286.5±0.2) eV can be attributed to C-S while the third component, C3, at (289.1±0.2) eV is related to the carboxylic group [3,10,13].

The O1s spectrum contains contributions from both the P-COOH molecule, the TiO₂ substrate and adventitious oxygen. The signal has a main component with a broad shoulder on the high BE side.

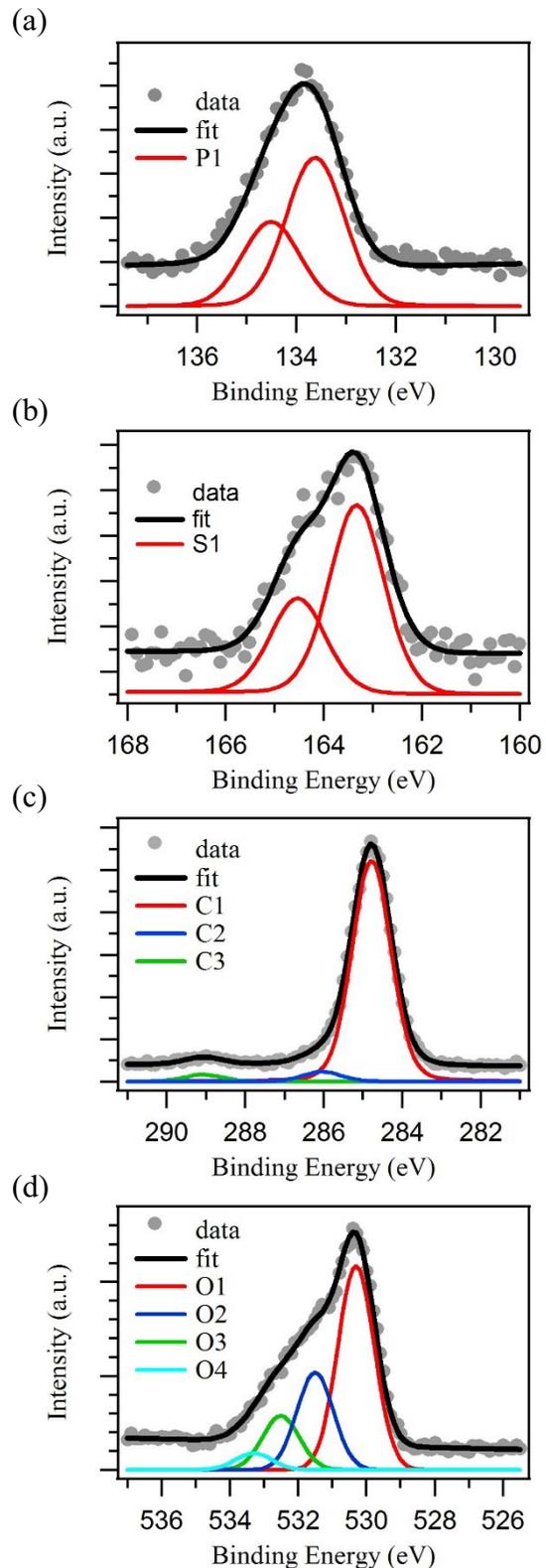


Fig. 4. XPS core level spectra of (a) P2p, (b) S2p, (c) O1s and (d) C1s regions of P-COOH layer deposited on plasma cleaned TiO₂ substrates

The data have been deconvoluted using four components with the same FWHM: O1 at (530.3±0.2) eV, O2 at (531.5±0.2) eV, O3 at (532.5±0.2) eV and O4 at

(533.3±0.2) eV. The O1 component can be assigned to the TiO₂ substrate [2,3,10,14]. Comparing with the expected BEs derived from literature, the O2 signal can be attributed to P-O-Ti and Ti-OH [10,14,15], the O3 component can be assigned to P=O [3,14,15] and C=O [3] while the O4 signal can be related to C-OH [3,14] and P-OH [3,15].

4 Conclusion

We investigated the adsorption on TiO₂ surfaces of an organophosphorous compound bearing a carboxylic end group (P-COOH) by combining AFM, SE and XPS. As concerns the substrate preparation, plasma cleaning was found to be useful to reduce adventitious carbon and to activate surface hydroxyl groups, suitable for anchoring phosphonates to the surface.

The AFM analysis indicates that P-COOH forms a sub-monolayer film on the TiO₂ surface with the formation of irregular islands on the surface. The analysis of the z profile of AFM images indicates the formation of islands of monomolecular height.

SE measurements show a decrease of the Δ values upon P-COOH deposition, further confirming the growth of the organic adlayer.

The analysis of the P2p, S2p, C1s and O1s XPS signals confirms the occurrence of molecular deposition as indicated by AFM and SE. However, at the present stage, we do not have conclusive evidence on the molecular binding mechanism and orientation on the surface. Indeed evidences have been reported in literature for the binding of carboxylic acid to TiO₂ surfaces [16,17]. On the other hand a previous study on the deposition of a ω -carboxyalkanephosphonic acid on TiO₂ reported, on the base of IR analysis, a preferential molecular binding through the phosphonate group [18]. Further analysis is therefore necessary to get a deeper insight in the molecule/surface binding mechanism. Ongoing angle-resolved XPS measurements will allow us to gain information on the molecular orientation within the layer, and therefore on the binding scheme.

A further aspect to be investigated will be the effect of post-deposition annealing on the film structure which has been reported to affect the binding as well [18].

Once clarified the anchoring mechanism, a step further will be the exploitation of unreacted carboxylic groups to anchor antimicrobial peptides through an amide bond in order to promote the antibacterial properties of the functionalized surface.

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