

## On-line supporting information

### A universal, template-free approach to Porous oxide and Polymer film processing

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Materials and methods

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Movie S1 and Movie S2

## Materials and Methods

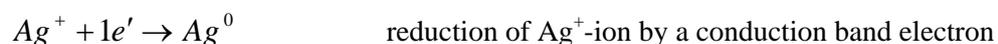
### Film preparation

#### a) Preparation of porous TiO<sub>2</sub> films

An ethanolic 0.15M Ti(IV) butoxide (Aldrich, 97% ) solution is first hydrolyzed with acidified 96% ethanol. After thorough mixing for 30 minutes, silver nitrate (Roth) or ammonium nitrate (Roth) is added and allowed to dissolve for 45 minutes. The molar ratios are 1Ti: 4H<sub>2</sub>O: 0.1 HNO<sub>3</sub>: 0,735 AgNO<sub>3</sub>/NH<sub>4</sub>NO<sub>3</sub> : 106 EtOH. Finally the sol is filtered through a 0.2 μm nylon filter.

Oxidized silicon or mirror finished Al 99.5% was used as substrate after thorough sonication in ethanol. The films were processed either by spin coating (Two steps: 800 rpm/8s and 6000 rpm/10s) or dip-coating at a withdrawing speed of 0.1mm/s. Drying was achieved at 200 °C for 30 minutes. An annealing treatment at 500 °C was eventually performed in order to drive crystallisation of the films. Silver containing samples were irradiated for 45 minutes with a

UV lamp ( wave length range from 240nm to 400nm, with maximum at 360 nm) in order to drive silver reduction following the well known reactions below.



#### b) Preparation of porous polymers

The materials used in this study were commercially available Polyvinylidene fluoride (PVDF) and PVDF-TrFE (trifluoroethylene) (Goodfellow, Germany),  $NH_4NO_3$  (Roth, Germany) and N,N-Dimethylformamide (DMF, Fluka, Germany) as solvent. First a polymer solution was prepared by dissolving the PVDF in DMF. Subsequently an appropriate amount of  $NH_4NO_3$  was dissolved in this solution by stirring and heating. A typical solution contains 15wt% PVDF and 15wt% of  $NH_4NO_3$ . Coatings were prepared on different substrates (steel, glass and polymer) by dip-coating. As the solution tends to gel after a certain time, the coating was done within 3 hours after dissolving the  $NH_4NO_3$ . Samples were dried at 120°C in a small perpendicular tube furnace for 15 min, followed by annealing at 130 (for PVDF-TrFE) and 150°C (for PVDF) for 12 hours. Afterwards, samples were cleaned and washed with deionized water and ethanol in an ultrasonic bath.

Functionalization of the porous matrix was performed by dip coating the samples in a dispersion of nanoparticles in Acetylacetone (Sigma-Aldrich, Germany). Acetylacetone leads to partial dissolution and quelling of the polymer surface, so that the polymer surface becomes solidly incrusted with the particles. The materials used were Hydroxylapatite (HAP, Riedel de Haen, Germany) and commercial  $TiO_2$  powder (P25, Degussa-Evonik).

#### Characterization

Particle size in the  $TiO_2$  precursor solutions were determined using Beckman-Coulter particle analyzer (Delsa NanoC). Morphological characterization was conducted on a high resolution scanning electron microscope (Ultra Plus, Zeiss, Germany) and structural characterization on

a Raman microscope (SENTERRA, Brucker, Germany) using a laser wave length of 532 nm and a laser power of 2mW.

### **Biocompatibility studies**

Prior to use in cell culture experiments the samples are rinsed with absolute ethanol (Merck) and dried in air.

Commercially available osteoblasts of the SAOS-2 cell line from human osteogenic sarcoma (ACC 243, Deutsche Sammlung von Mikroorganismen und Zellkulturen GMBH) are used for in vitro biocompatibility testing. SAOS cells are cultured in DMEM (4,5g glucose/l, PAA) supplemented with 10% FCS (PAA), 2.5µg/ml AmphotericinB, 100U/ml Penicillin, 100µg/ml Streptomycin (PAA) and 40µg/l Dexamethason (Serva).

Medium is changed every 72 hours. At 80-90% confluence cell monolayers are removed by addition of Trypsin/EDTA (0.05/0.02%, PAA) for 5 minutes at 37°C and 5% CO<sub>2</sub>.

For morphology characterisation cells are seeded in 6-well plates at a concentration of  $2.4 \times 10^5$  per well. Prior to cell seeding coated samples are added to plates. Negative control (NC) testing is carried out on coverslips (Thermanox, Nunc). After an incubation period of 24h, cells are either stained with Giemsa stain (PAA) or investigated by light microscopy, or critical point dried and characterised by SEM.

Viability testing is performed in the presence of sample eluates. For this purpose samples were covered with DMEM and incubated for 24h at 37°C/5%CO<sub>2</sub> in 6-well plates at a concentration of 0,6 ml/cm<sup>2</sup> sample surface. Negative controls (NC) are run with pure medium under the same incubation conditions.

For viability testing via XTT (Sodium3,3'-{-(Phenylamino)carbonyl]-3,4-Tetrazolium}-Bis(4-methoxy-6-nitro)benzenesulfonic acid hydrate)  $10^4$  cells/100µl/well are seeded in 96-well plates. After 1.5h, during which cells adhere, medium is substituted for the sample eluates or simple medium (NC) and a further incubation period of 24h at 37°C/5%CO<sub>2</sub> follows. Subsequently 25µl of a XTT/PMS (1.4 mM XTT (Serva), 0.04 mM PMS

(Methylphenazinium methylsulfate, Serva), PBS (PAA)) mixture in PBS is added and incubation at 37°C/5%CO<sub>2</sub> is prolonged for further 3h. Finally the optical density is measured at 470nm/750nm. Blanks consist of cell free medium.

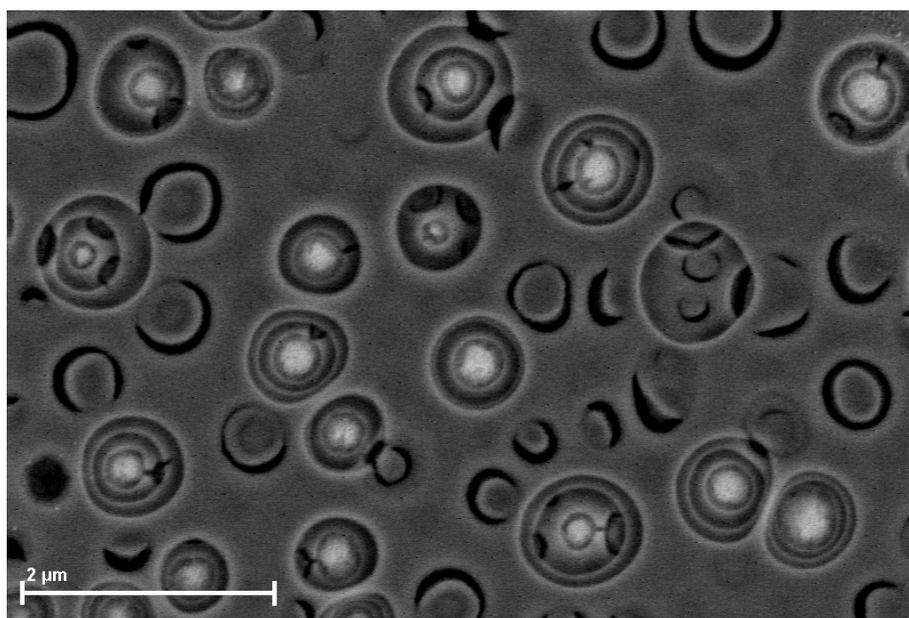


Fig.1S: Backscattered Electron micrograph of a hierarchical porous TiO<sub>2</sub> film, where the open nature of the pores is evident; one can “see” through the pores where the substrate is, conspicuous via its bright contrast. The film was spin-coated on a stainless steel substrate.

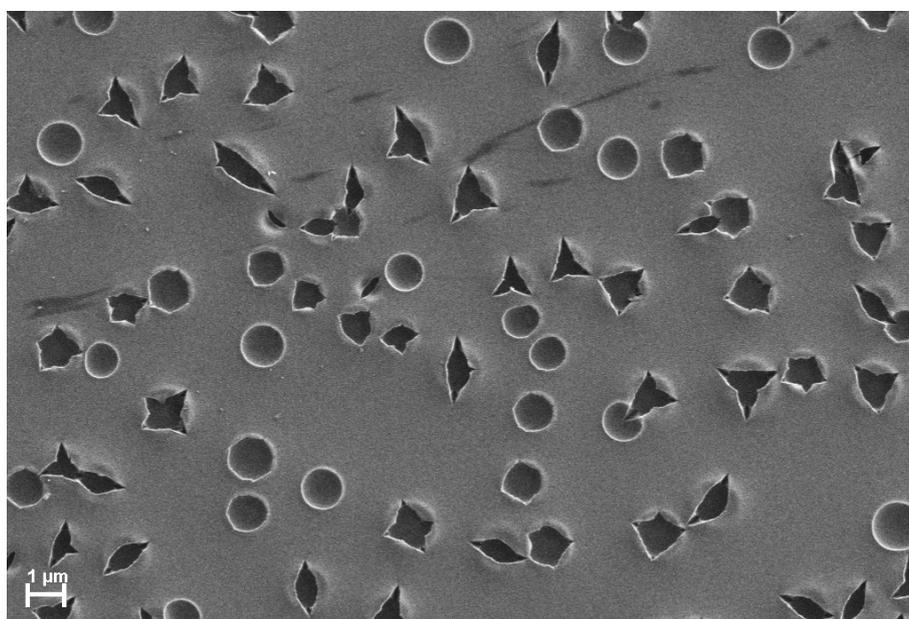


Fig.2S: Secondary electron micrograph of a TiO<sub>2</sub>-layer that was processed using a lesser amount of the NH<sub>4</sub>NO<sub>3</sub> than the optimum one. The sluggish nature of the reaction is evident from the irregular morphology of the pores, though the reaction zone was circular. One can also see that the released energy was not sufficient to open many pores.

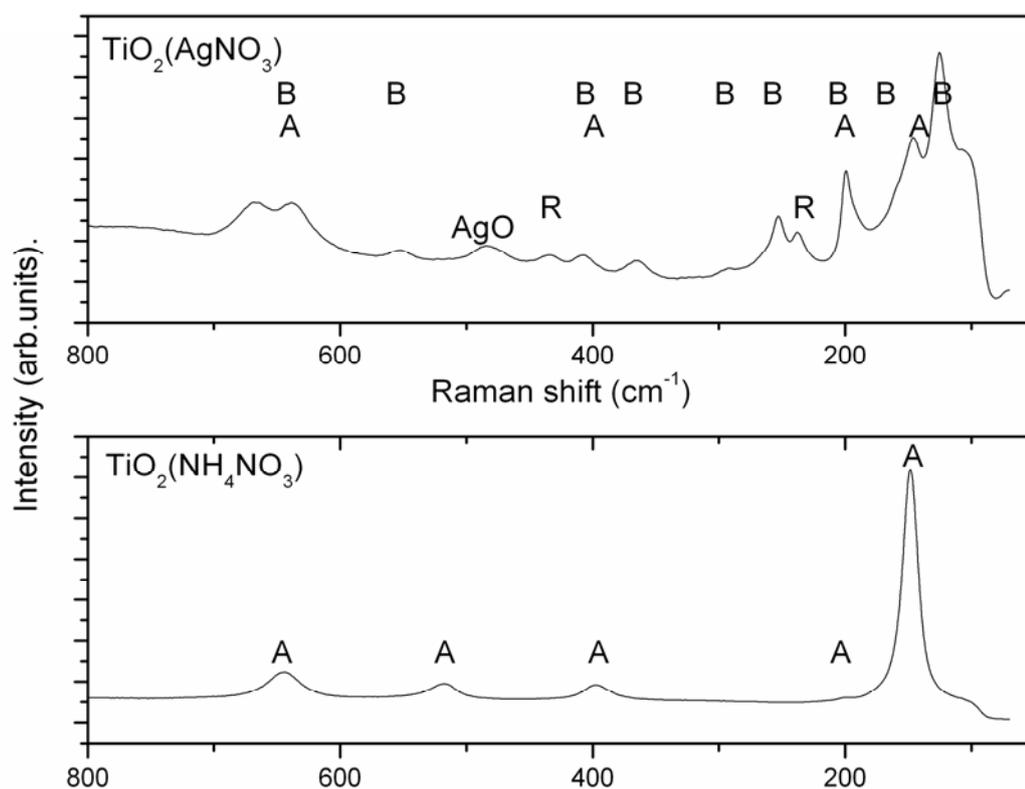


Fig.3S: Raman spectra of porous films made with NH<sub>4</sub>NO<sub>3</sub> (lower patterns) and AgNO<sub>3</sub> (upper patterns). The microstructures obtained are depicted in Figs. 2C and D. The film made with NH<sub>4</sub>NO<sub>3</sub> consists of pure anatase, A, as indicated by characteristic bands. The film made with AgNO<sub>3</sub> shows a complex microstructure that consists of a mixture of brookite, anatase and small amounts of rutile. Some of AgO is also present. The bands at 148.5, 398, 518 and 644 cm<sup>-1</sup> are attributed to anatase (space group *I4<sub>1</sub>/amd*)<sup>1S</sup>; in the upper spectrum the bands at 125, 195, 253, 291, 366, 407, 552 and 639 cm<sup>-1</sup> are assigned to brookite (space group *Pbca*). The bands at 238 and 434 cm<sup>-1</sup> may be assigned to rutile (space group *P4<sub>2</sub>/mmm*). We attribute the band at 484 cm<sup>-1</sup> to AgO, but characteristic bands for this compound are missing.

Further, there are bands at 105 and 670  $\text{cm}^{-1}$  that cannot be assigned to any of Ti-oxide or silver oxide structures.

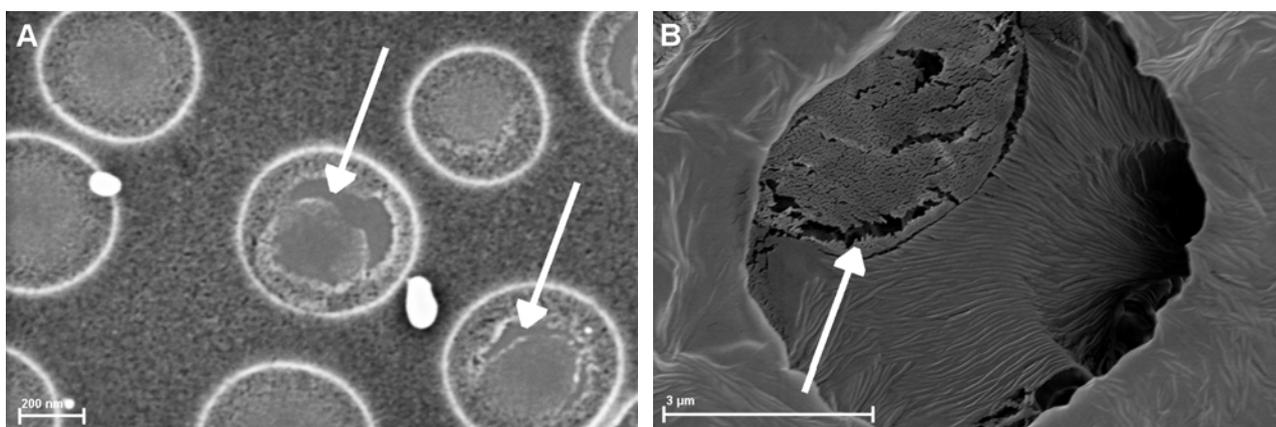


Fig. 4S: (A) Porous  $\text{TiO}_2$  dried at room temperature. Arrows show skins that are still attached to the pore wall because of a sluggish reaction. (B) Incomplete removal of the “skin” in a PVDF-TrFE film.

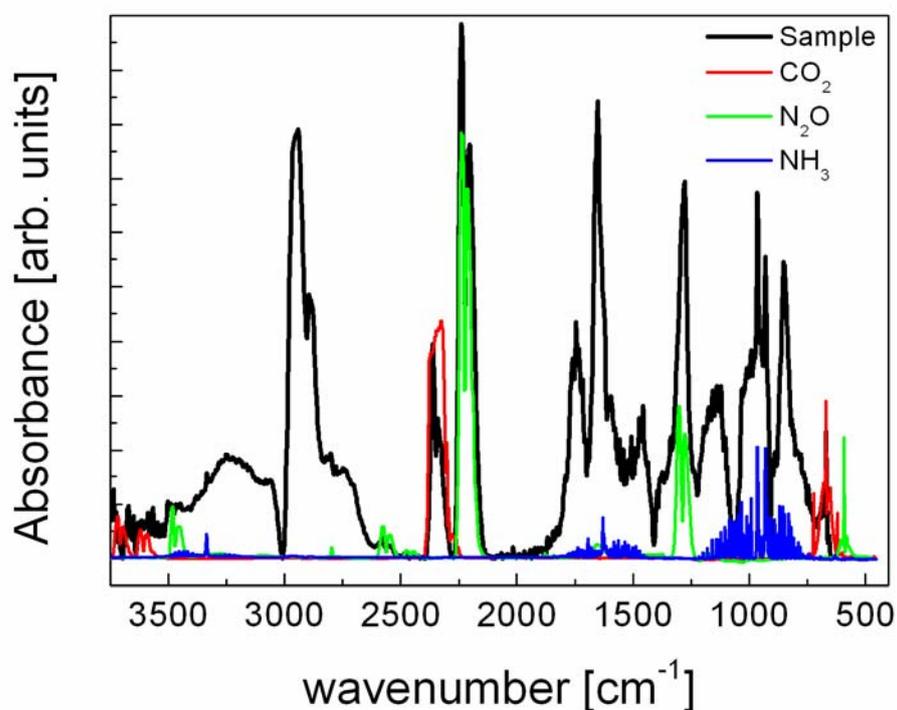


Fig. 5S: FTIR-spectra of evolving gas at 199°C, during a TG-experiment that was conducted on a precursor solution containing  $\text{AgNO}_3$  as oxidant. The spectra of the sample are superimposed on reference spectra of  $\text{CO}_2$ ,  $\text{N}_2\text{O}$  and  $\text{NH}_3$ .

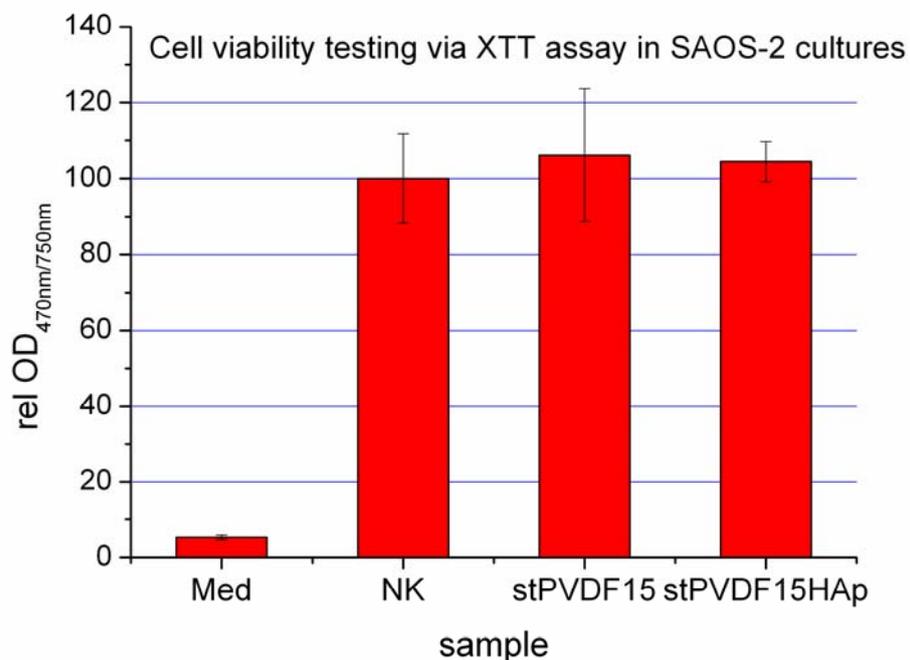


Fig. 6S: XTT assay results of viability and proliferation in the presence of 24h-eluates of the porous PVDF and PVDF/Hap coatings in SAOS-2 cultures after 24h. The results assign excellent biocompatibility to both types of coating. These findings are confirmed by morphological studies using light and scanning electron microscopy. Med.: cell free medium; NK: negative control (cells without eluates); stPVDF15 and stPVDF15HAP are porous PVDF samples without and with HAP, respectively.

### Movie captions

Movie S1. Video-track of pore formation in a thick PVDF-TrFE copolymer film obtained using an optical microscope (20x objective lens) equipped with a high definition video camera. At the beginning one can see “dimple” formation that suggests self-organization of droplets; this process may last several hours. A fast reaction (when the reaction front had reached the location under view, see Video S2) is seen at the end of the video, where the dimples grew larger and subsequently pore formation takes place by gas-expansion. The dark color originates from light absorption by gaseous species, e.g. nitrogen oxide, and light scattering in voids. One hints also that micro-detonations take place at different levels of film thickness.

Movie S2. Macro-video track of advancing reaction front (opaque, bright front) in the gel-like film.

## References

- 1S. G.A. Tompsett, G.A. Bowmaker, R.P. Cooney, J.B. Metson, K.A. Rodgers, J.M. Seakins,  
*J. Raman Spectrosc.* **1995**, 26, 57-62.