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Sensor Array Composed of "Clicked" Individual Microcantilever Chips

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A simple technique is described to functionalize a small library of microcantilever (MC) chips presenting varied headgroups. A generic azide monolayer, bound to the MC surface, can be coupled with various alkynes using efficient "click" chemistry. This method is compatible with many functional groups, and novel headgroups are introduced on the MC surface by means of alkynes synthesized via a one-step reaction. The surface "click" reaction reduces greatly the effort that would be required to synthesize and purify the corresponding functional thiols. This technique represents a convenient complementary tool for Phase-Shifting Interferometric Microscopy (PSIM) read-out that has been developed in our group. The affinity of these surface coatings towards different solvents can be estimated by measuring the deflection of the cantilevers. A proof-of-concept sensor composed of four individual MC chips presenting different headgroups can unambiguously discriminate the fingerprint response of a nerve-gas simulant from other solvent vapors.

1. Introduction

The large variety of species that can be detected, on binding to receptor layers on the surface of microcantilevers by subsequent surface-stress-induced static deflections or adsorbate-mass-induced changes in the resonant vibrational frequency, demonstrates the versatility of MCs as chemical and biological sensors.[1] This technique is extremely sensitive and the detection of single-base mismatches,[2] absence of single hydrogen bonding sites,[3] ions,[4] hydrogen,[5] and volatile organic compounds (VOC)[6] have all been reported. Cantilever-based chemical sensors are limited, however, by the availability of coatings that interact exclusively and selectively with the analyte of interest. Unlike biomolecules, small organic compounds can lack a chemical "handle" that allows their selective recognition based on the lock-and-key principle. Therefore, some chemical sensors are susceptible to several compounds, resulting in possible false positives that cannot be tolerated for many applications. For example, microcantilevers with receptor layers presenting a copper-doped carboxylic acid headgroup have been proposed as potential candidates for nerve-gas sensing,[7] but these are also sensitive to exposure to solvents and changes of pH.

It has been proposed that an array of different receptors could be able to recognize certain molecules, even without highly selective interactions, in analogy to the olfaction mechanism in mammals.[8] For example, colorimetric arrays composed of metalloporphyrins and dyes have been used to differentiate volatile organic compounds (VOCs).[9] Also, solvent-vapor recognition has been recently achieved by principal component analysis of the response of an array of six microcantilevers functionalized with thin polymer films.[6f] Simultaneous monitoring of the cantilevers was achieved by means of a piezoelectric readout in that study.

The functionalization of individual microcantilevers in arrays with several distinct receptor coatings generally requires the use of micropipette or microfabrication tools that prevent the rapid screening of novel surface coatings. Therefore, most reports describe MC chips decorated with a single type of coating that gives redundant information and limits the miniaturization of the sensing devices. We have recently reported an optical method to determine simultaneously the entire bending profile of several cantilevers, belonging to independent chips, based on a Twyman–Green interferometer.[10] Phase-shifting interferometric microscopy (PSIM) uses a single light source and it is suitable for microcantilevers arrays. This readout system can be used in combination with an innovative low-volume measuring cell designed for up to four individual standard chips. In the present configuration, the system requires only minimum alignment and allows the rapid replacement of MC chips.

"Pick and Mix" arrays of cantilevers can therefore be built from individual chips. This has the advantage that each chip can be functionalized with a different monolayer simply by immersion using conventional laboratory glassware. Also, if a cantilever becomes damaged, or if its coating proves to be unsuitable for the application of interest, it is possible to replace this single MC chip while keeping the other sensor chips in place. This PSIM readout capability offers researchers without access to microfabrication tools the possibility to design complex

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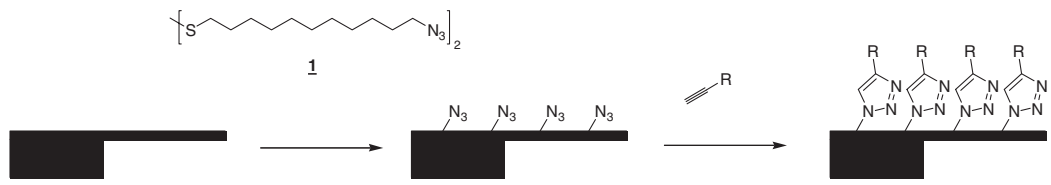

Scheme 1. Cartoon representation of the general functionalization of surface using "click" chemistry.

receptor-coated microcantilever arrays, and to test and optimize them rapidly.

The synthesis of compounds able to form monolayers is time consuming, resulting in a limited number of headgroups being readily available. Instead, we propose here to functionalize receptor coatings on microcantilever chips by "click" chemistry. The copper-catalyzed Huisgen cycloaddition between an azide and an alkyne (CuAAC)[11] is orthogonal to most functional groups, proceeds to high yield under mild conditions and without significant formation of side-products. This synthetic tool has been widely adopted by polymer and material chemists.[12] "click" reaction on gold surfaces (e.g., coating MCs) can be achieved by the formation of an azide monolayer followed by addition of an alkyne,[13] or by coupling of an alkyne surface with an organoazide.[14] A recent comparison of these methods suggested that the first route leads to greater coverage of the surface.[15] Also, it greatly reduces the handling of organic azides that must be considered as potentially hazardous, explosive and toxic compounds.[16] As in the case of solid-supported synthesis, performing the "click" reaction on the surface limits the purification effort that would be necessary if this was carried out before the monolayer formation.

In this report, we describe the preparation of a library of receptor-coated microcantilever chips by "click" reactions. Bis(11-azido undecanyl)disulfide **1** is used for the formation of azide monolayers on gold surfaces. Individual chips are functionalized by reactions with different alkynes, as shown in **Scheme 1**. Several simple alkynes are commercially available and two simple methods to synthesize novel alkyne derivatives are described. The alkynes used in this study were designed to present a wide variety of functional groups that could take part in the formation of non-covalent interactions, once immobilized on the surface of microcantilever chips. This technique reduces greatly the synthetic effort necessary to the preparation of such library of microcantilever chips. The affinity of these receptor surfaces for several solvents was evaluated by determining the bending response of the microcantilevers upon exposure to different solvent vapors. Combined with a PSIM read-out, the screening of different headgroups can be achieved rapidly and arrays of microcantilevers can be designed by combining individual chips without access to specialized microfabrication tools.

2. Results

2.1. Synthesis of Azide-Terminated Disulfide and Alkyne Compounds

The azide-terminated disulfide **1** was synthesized in three steps. Bis(11-hydroxyundecanyl)disulfide was obtained by oxidation of 11-mercaptoundecanol with *N*-chlorosuccinimide. It was converted to bis(11-methanesulfonate undecanyl)disulfide by reaction with mesyl chloride in the presence of triethylamine. Nucleophilic substitution of the mesylate groups by sodium azide gave bis(11-azidoundecanyl)disulfide **1**.

Novel alkynes (**Figure 1**) were synthesized in one step by reaction of alcohol derivatives with propargyl bromide in the presence of base, or by ester formation in the presence of EDC and DMAP. All compounds were obtained in good yield after purification by column chromatography, and their structures were confirmed by proton and ^{13}C NMR, FT–IR and elemental analysis.

2.2. "Click" Reaction of Simple Alkynes

Azide-terminated monolayers on gold-coated glass microscope slides were prepared by immersion overnight in 1 mM ethanol solution of **1**. Loosely bound molecules were removed by washing the substrate with ethanol and hexane. The "click"

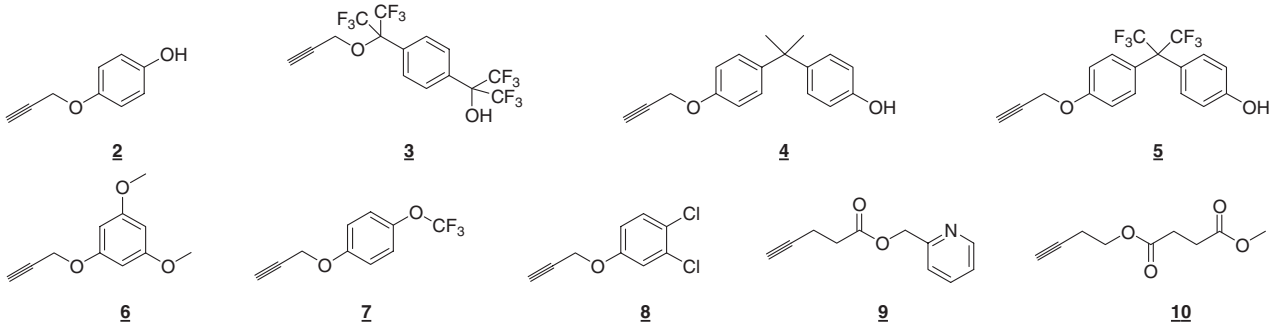

Figure 1. Alkyne derivatives synthesized in this study.

reactions of receptor layers on microcantilevers presenting azide groups with 4-pentynoic acid, N,N dimethylaminopropyne, 3-butynol and 9-decyne were performed in a mixture of isopropanol and water in the presence of copper sulfate and sodium ascorbate. After reaction, the catalyst and excess of alkyne were removed by washing the substrate extensively with ethanol, followed by water, ethanol and hexane. Sessile drop contact angles with water were determined. The surfaces presenting a polar head group were found to be hydrophilic (carboxylic acid = 43.8 ± 1.1°;. N,N dimethylamine = 27.1 ± 1.0°; alcohol = 36.8 ± 1.4°). The surface presenting a 'clicked' alkane was hydrophobic (83.1 ± 2.3°).

2.3. Surface Functionalization of Microcantilevers

The chips used in this study had two rectangular 500 μm long single-crystal Si cantilevers pre-coated with gold on the top side (Nanoworld, TL2Au). They were mounted on a custom-made clamp to minimize the risk of damaging the cantilever when manipulating the chips. Prior to surface functionalization, the cantilever chips were cleaned by successive washing with 1 N HCl, water, ethanol, hexane and ethanol. A library of chips was obtained by carrying out the "click" reaction with a set of different alkynes **2**–**10**. Another set of chips presenting a carboxylic acid head group was obtained by immobilization of 4-pentynoic acid, followed by immersion in a solution of copper sulfate. Finally, an alkane-coated chip was obtained by functionalization with octadecanethiol (ODT) without using "click" chemistry.

2.4. Response of Microcantilevers to Solvent Vapors

The microcantilevers' responses were characterized by their bending behavior when exposed to solvent vapors. Solvent vapors were generated by bubbling nitrogen gas through a bottle of solvent under constant gas flow. Chips were mounted in the PSIM read-out system and exposed to one-minute long injection of several vapors that were repeated three times. With the PSIM optical readout technique, the entire profiles of all monitored cantilevers are obtained simultaneously. In the present study, only the position of the tip of the cantilever is reported for clarity. The affinity of all the cantilevers was determined for toluene, ethanol and DMMP, and the bending responses are summarized in **Table 1**. The response of selected cantilevers was also determined for dichloromethane and acetone.

Table 1. Maximum amplitude of microcantilever bending upon injection of solvent vapors. The position of the tip of the cantilever is given in nm.

Surface coating	Toluene	Ethanol	DMMP
2	−336	−673	−302
3	−164	−197	324
4	−450	−360	−178
5	166	220	344
6	119	166	279
7	192	182	176
8	−89	185	289
9	7	−48	18
10	−136	42	216
$COOH/Cu^{2+}$	41	184	314
ODT	187	692	386

3. Discussion

Compounds that possess a thiol and an azide group at each end of a linear aliphatic chain have the ability to form azide-terminated monolayers on gold. In previous reports, the synthesis of 11-azidoundecane-1-thiol was carried out in four steps that all involved the handling of organic azides.[13a] This maximizes the risks associated with the handling of this class of compounds.[16] Also, organic azides are known to degrade with time and such compounds cannot be stored for prolonged periods of time. This limits the practical adoption of the "click" functionalization technique by material scientists that do not possess a chemical background. Disulfides can also form monolayers on gold and we have developed a simple method to synthesize bis(11-azido undecanyl)disulfide **1** to introduce the azide group in the last step. Firstly, commercially available 11-mercaptoundecanol was oxidized to avoid the competitive reaction between the thiol and alcohol in subsequent steps. Then, the alcohol was converted to a mesylate group. Large amounts of this intermediate compound can be prepared and stored for prolonged periods of time. Finally, the compound was reacted with sodium azide, which afforded **1** at 34% overall yield. When the last step of the synthesis was performed on a 50 mg scale, it was found that this compound could be purified easily by passing through a pad of silica gel in a Pasteur pipette. This results in enough material to functionalize several gold-coated substrates and ensures that freshly prepared **1** is systematically used.

The alkynes synthesized in this study were designed to confer a wide range of properties to the corresponding surfaces. To limit the effort dedicated to their synthesis, it was decided to prepare these compounds in a single step. A series of compounds was obtained in moderate to good yield by reaction of propargyl bromide with phenol derivatives (Figure 1). Alkynes presenting a phenol moiety (**2**, **3**, **4**, and **5**) are both hydrogen-bond acceptors and donors. Also, it has been reported that the hexafluoroisopropanol and 4-(hexafluoropropyl)phenol moiety found in **3** and **5** interacts selectively with organophosphates.[17] It was expected, therefore, that the corresponding surfaces could serve as nerve-gas sensors. Three electron-rich aromatic compounds that can take part in weak π–π stacking were also synthesized (**6**, **7** and **8**). Alternatively, the alkyne moiety can be introduced through the formation of an ester bond, using 4-pentynoic acid or 4-butynol. Reaction of 3-(hydroxymethyl)pyridine with 4-pentynoic acid in dichloromethane, in the presence of DMAP and EDC, gave **9** in excellent yield. While the formation of ether linkages is preferable as they are not susceptible to hydrolysis, this route can be preferred in case of incompatibility of the alcohol derivative with propargyl bromide. Finally, the aliphatic diester **10** was synthesized in excellent yield by esterification of 4-butynol with monomethyl succinate, under

similar conditions. This route can be applied to compounds presenting a carboxylic acid moiety, considerably expanding the scope of alkyne derivatives that can be synthesized in a single step.

Gold-coated microscope slides were functionalized with several commercially available alkynes and contact-angle measurements were performed. As expected, surfaces presenting a carboxylic acid, alcohol or amine headgroup were hydrophilic, while the substrate 'clicked' with the alkane was hydrophobic. This suggests, in a qualitative manner, that the surfaces were successfully modified. The efficiency of the surface "click" reaction has been demonstrated by several groups.[13,14] In particular, a comparative study confirmed the suitability of the condition chosen in our report.[15] Microcantilever chips were functionalized with alkynes **2-10** and 4-pentynoic acid using the "click" reaction and another chip was decorated with ODT.

The bending response of the four cantilevers belonging to two chips decorated with **3** upon repeated one-minute long exposures to toluene vapor is shown in **Figure 2**. The amplitude and direction of the bending of all four cantilevers was essentially identical over repeated exposures to toluene, demonstrating the reproducibility and uniformity of the "click" functionalization. The interaction of toluene molecules with the microcantilever surface resulted in a compressive stress and the maximum amplitude of the bending was reached in less than 10 seconds. When the first injection of toluene was completed, evaporation of the loosely bound molecules during an N_2 purge resulted in a fast partial recovery of the original position of the tip of the cantilevers. Toluene molecules involved in π–π stacking with the surface were desorbed in a second slow step, as demonstrated by the slow return of the cantilevers to their original position that took over a minute. It was found that ethanol, acetone and dichloromethane all gave rise to compressive stress, translated in downward bending (**Figure 3**). The variations of maximum amplitudes might arise from differences in the boiling point and vapor pressure of the solvents, and from their affinity for the microcantilever receptor surfaces through the formation of non-covalent interactions. After exposure to vapor, the rate of return of the cantilever to its original position was characteristic for each solvent. In the case of ethanol, it was difficult to determine the absolute position of the cantilever as a result of drift in the baseline. This was occasionally observed during our work and is suspected to be an artifact caused by gradual release of residual stress in the microcantilever surface coating. This baseline drift had no influence on the direction of the response of the cantilevers on exposure to a given solvent vapor. When the cantilever was exposed again to ethanol after 20 minutes, the amplitude of the downward bending was only half that of the first peak. This suggests that this solvent interacts strongly with the receptor surface through hydrogen bonding. Full recovery of the amplitude could be obtained by drying the chip under a stream of nitrogen overnight. After exposure to acetone, the cantilever returned to its original position in ca. 10 minutes. Ethanol has a greater affinity for the receptor surface as it is both a hydrogen-bond donor and acceptor, while acetone is a hydrogen-bond acceptor. Dichloromethane does not interact specifically with the receptor surface and desorption of loosely interacting molecules was rapid. Finally, dimethyl methylphosphonate (DMMP), a simulant for sarin, gave a distinctive tensile stress (upward bending) which could be readily identified from that of other solvents. Organophosphonates are capable of accepting multiple hydrogen bonds and it is suspected that a single molecule of DMMP interacts with several neighboring hexafluoroisopropanol sites.

The sensitivity of this technique is limited by the thickness of the monolayers and by the possible low density of functional groups. Indeed, the amplitude of the deflections observed in this study are smaller than in previously reported systems using thick polymeric coatings.[6e,6f] It has been shown that a granular surface morphology, obtained by plasma polymerization, greatly enhances the response and a MC coated with polyacrylonitrile showed a reversible response to naphthalene at 40 °C with a limit of detection of 1 ppb.[6e] The technique described in this study allowed the rapid screening of novel headgroups that could be adapted for functional polymer brushes or bulk polymer coatings.

In this study, the method used to produce the vapor of solvents did not allow control of the concentration of the analyte. For certain application such as nerve gas detection, it is less important to avoid false positives but determining the exact concentration

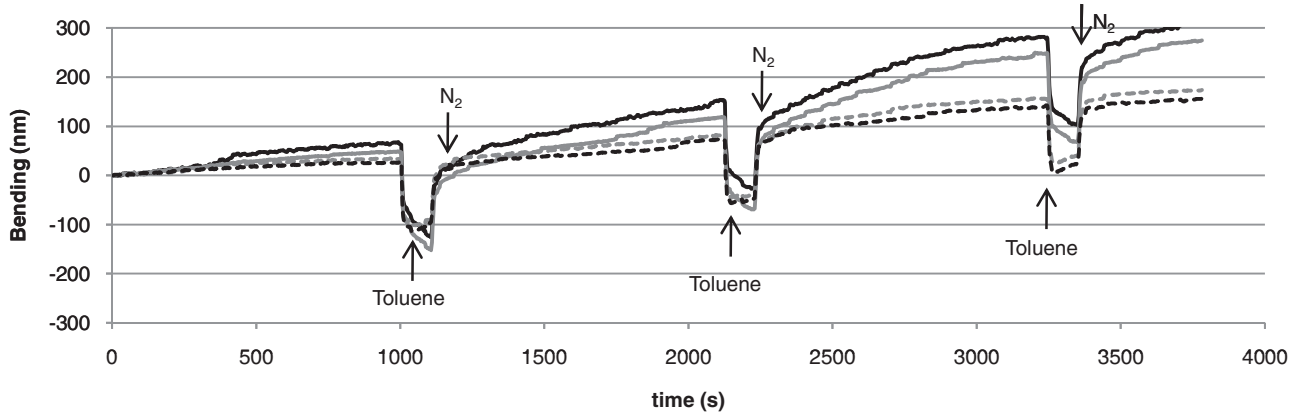

Figure 2. Response of four individual microcantilevers from two sensor chips, decorated with 3, upon repeated exposure to toluene vapor. Traces with solid lines are from different cantilevers from one sensor chip and traces with dashed lines are from different cantilevers from another sensor chip.

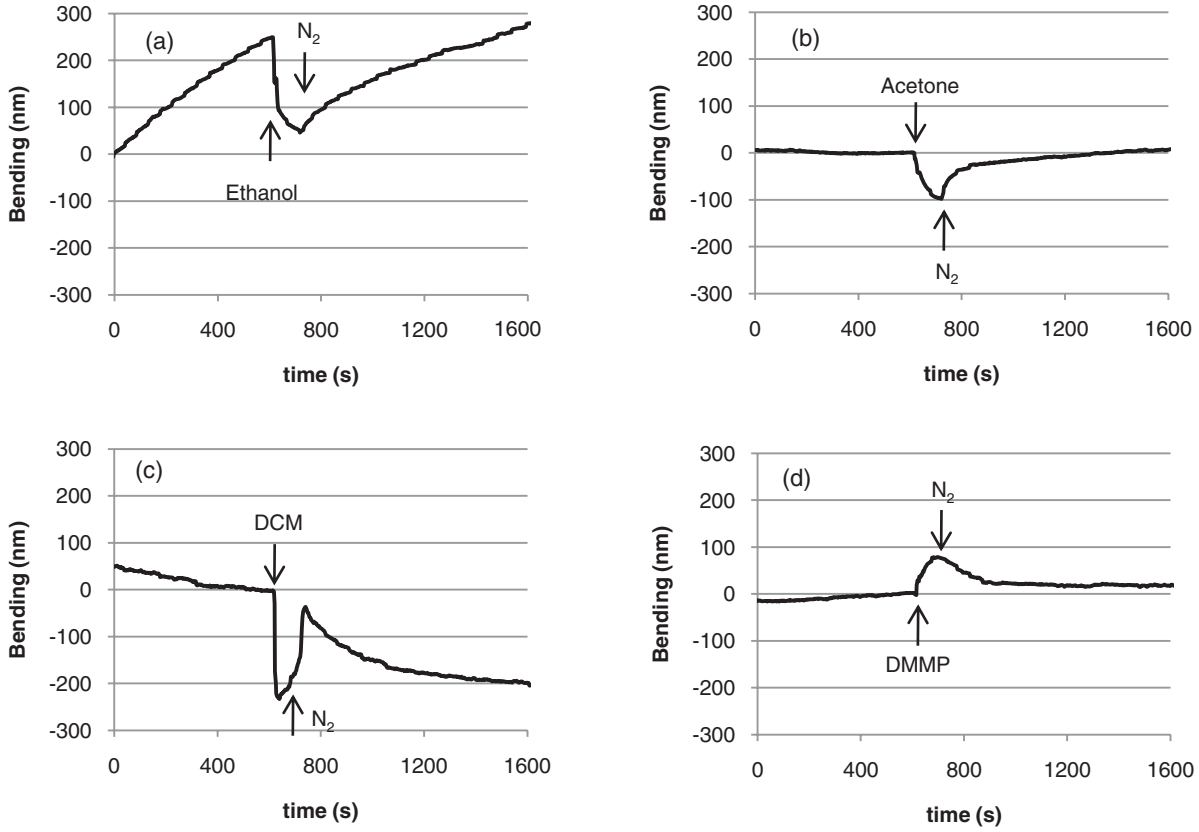

Figure 3. Response of a microcantilever functionalized with 3 upon exposure to (a) ethanol, (b) acetone, (c) dichloromethane and (d) dimethyl methylphosphonate vapors.

of extremely toxic substances is not essential. It was envisaged to group the coatings based exclusively on the bending direction (compressive or tensile) of their stress responses. The cantilevers decorated with ODT and those functionalized with pentynoic acid, **5**, **6** and **7** exhibited upward tensile bending when exposed to all solvents. The chip presenting the carboxylic acid headgroup and doped with Cu^{2+} ions exhibited the weakest response against toluene of this group, as this coating is not aromatic. Its response to DMMP was significantly stronger than to the other solvents, and it has been shown previously that this coating can interact with organophosphates.[7] However, the response is not exclusive and this microcantilever sensor alone is not sufficient to differentiate the nature of the vapors. This represents a major obstacle to the adoption of microcantilever sensors for security applications. For example, ethanol is one of the main components of perfumes, and it is present in food and drinks which would lead to repeated false positives that cannot be tolerated. The microcantilevers functionalized with a simple phenol group (**2** and **4**) gave a downward bending when exposed to all three solvents, suggesting that in the case of the microcantilever decorated with the receptor **5**, the headgroups are probably dominated by the hydrophobic moiety rather than by the hydroxyl group. Finally, for the chip functionalized with **8** and **10**, exposure to toluene gave rise to a tensile stress, while ethanol and DMMP induced compressive stress.

Based on the screening of individual chips, it would be possible to select four chips to build a microcantilever array capable of sensing and discriminating nerve-gas stimulants from common solvents. We have recently developed a 4-chip cell that could be used to build arrays. Using a dedicated measuring cell holding four chips, the fingerprint response of VOCs, could be identified using the PSIM readout. A preliminary experiment showed that an array composed of chips functionalized with **2**, **3**, **10** and ODT could recognize DMMP and ethanol unequivocally by monitoring the direction of bending of each cantilever (**Figure 4**). Toluene, acetone and dichloromethane could be

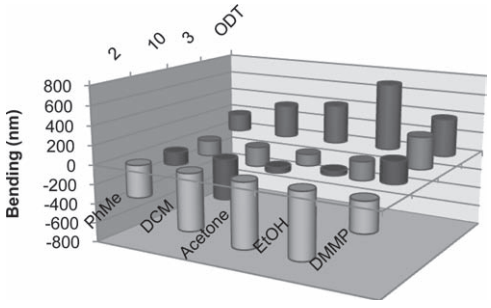

Figure 4. Bending response of four selected cantilevers when exposed to toluene, dichloromethane, acetone, ethanol and DMMP.

differentiated by the relative amplitudes of response. The cantilevers decorated with **2** and **10** had a strong response to the chlorinated solvent, whereas the amplitude of the response to acetone was significantly greater for **2** than for **10**. In the presence of toluene, the amplitude of the bending of cantilevers functionalized with **3** and **10** was about half of that observed for the cantilever decorated with **2**.

In future work, we plan to increase the number of simultaneously monitored cantilevers and will be using multivariate statistics techniques, such as principal component analysis, to increase the conclusiveness of our results. We also plan to compare accuracy and reproducibility of data obtained from limited time span exposure, as presented here, with saturation data.

4. Conclusions

The combination of "click" functionalization of receptor monolayers on microcantilevers and phase-shifting interferometric microscopy readout represents an excellent combination to rapidly screen the behavior of libraries of novel microcantilever-receptor coatings. New surface coatings can be designed using simple chemical reactions, expanding considerably the scope of functional groups that could be made available to material scientists. Also, sensor arrays can be built by combining individual 'clicked' chips prepared using exclusively standard laboratory glassware. The development of chemical sensors for point-of-care medical diagnosis or of microcantilever-based logic gates could be envisaged, as a result of the simplicity and versatility of this technique.

5. Experimental Section

General Methods and Instrumentation: Solvents were obtained from Fisher and used without purification. Chemicals were purchased from Aldrich or Acros and used as received. All reactions were carried out under a nitrogen atmosphere, unless otherwise noted. Attenuated total reflectance FT-IR spectra were acquired on a Perkin Elmer Spectrum One equipped with a Universal ATR accessory. NMR (^{1}H and ^{13}C) spectra were recorded on 400 MHz Bruker spectrometers. The chemical-shift data for each signal are given in units of δ (ppm) relative to tetramethylsilane (TMS) where δ (TMS) = 0, and referenced to the residual solvent resonances. Splitting patterns are denoted as s (singlet), d (doublet), t (triplet), q (quartet), m (multiplet), and br (broad).

Bis(11-Hydroxyundecanyl)disulfide: To a stirred solution of 11-mercaptoundecanol (1.022 g, 5.0 mmol) in dichloromethane (20 mL) was added *N*-chlorosuccinimide (0.334 g, 2.5 mmol) in dichloromethane (5 mL). A white precipitate was rapidly formed. After ten minutes, warm dichloromethane (50 mL) was added and the solution was washed with water (30 mL). The organic layer was washed with water (30 mL), dried with magnesium sulfate, filtered and evaporated under reduced pressure to yield bis(11-hydroxyundecanyl)disulfide (0.996 g, 98% yield) as a white powder, m.p. = 63-66 °C; FT-IR (neat) ν = 3347, 2917, 2850, 1469, 1346, 1055, 1038 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ = 1.25-1.39 (m, 28 H, CH_2), 1.52-1.73 (m, 8 H, CH_2), 2.71 (t, J = 7.4 Hz, 4 H, S-CH_2), 3.66 (t, J = 6.6 Hz, 4 H, O-CH_2); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 25.7, 28.5, 29.2, 29.4, 29.5, 29.6, 32.8, 39.2, 63.0; HR-MS (ESI): calculated for $C_{22}H_{46}O_2S_2$ [M+H]$^+$, 407.3012, found 407.3026.

Bis(11-Methanesulfonate Undecanyl)Disulfide: To a stirred solution of bis(11-hydroxyundecanyl)disulfide (0.407 g, 1.0 mmol) in dichloromethane (20 mL) and pyridine (0.2 mL) at 0 °C was added methanesulfonyl chloride (0.252 g, 2.2 mmol). After an hour, the mixture was stirred overnight at room temperature, and poured in water (30 mL) and dichloromethane (30 mL). The organic layer was washed with water (30 mL), dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded bis(11-methanesulfonate undecanyl)disulfide (0.355 g, 63%, R_f = 0.35 in CH_2Cl_2) as a white powder, m.p. = 70-72 °C; FT-IR (neat) ν = 2918, 2851, 1473, 1340, 1327, 1166, 1159 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ = 1.21-1.40 (m, 28 H, CH_2), 1.62-1.77 (m, 8 H, CH_2), 2.67 (t, J = 7.4 Hz, 4 H, S-CH_2), 2.99 (s, CH_3), 4.21 (t, J = 6.6 Hz, 4 H, O-CH_2); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 25.4, 28.5, 29.0, 29.1, 29.2, 29.4, 37.4, 39.2, 70.2; HR-MS (ESI): calculated for $C_{24}H_{50}O_6S_4$ [M+H]$^+$, 585.2382, found 585.2384.

Bis(11-Azido Undecanyl)Disulfide **1**: To a stirred solution of bis(11-methanesulfonate undecanyl)disulfide (0.056 g, 0.1 mmol) in dimethylformamide (2 mL) was added sodium azide (0.013 g, 0.2 mmol). After 48 hours, diethyl ether (40 mL) was added and washed twice with water. The organic layer was dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded bis(11-azido undecanyl)disulfide **1** (0.025 g, 55%, R_f = 0.71 in petroleum ether/ethyl acetate 9:1) as a colorless oil, FT-IR (neat) ν = 2924, 2853, 2091, 1464, 1259 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ = 1.21-1.45 (m, 28 H, CH_2), 1.55-1.69 (m, 8 H, CH_2), 2.67 (t, J = 7.4 Hz, 4 H, S-CH_2), 3.24 (t, J = 7.0 Hz, 4 H, N_3-CH_2); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 26.7, 28.5, 28.8, 29.1, 29.2, 29.5, 39.2, 51.5; HR-MS (ESI): calculated for $C_{22}H_{44}N_6S_2$ [M+H]$^+$, 457.3142, found 457.3123.

4-Propargyloxyphenol **2***:* To a stirred mixture of hydroquinone (0.220 g, 2.0 mmol) and potassium carbonate (0.138 g, 1.0 mmol) in dimethylformamide (5 mL) at 60 °C was added propargyl bromide (0.117 g, 1.0 mmol). After four hours, dichloromethane (50 mL) was added, the organic layer was washed with 10% HCl (30 mL), water (30 mL), dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded 4-propargyloxyphenol **2** (0.098 g, 66%, R_f = 0.15 in dichloromethane) as a yellow oil, FT-IR (neat) ν = 3370, 3289, 1702, 1506, 1146, 1375, 1199, 1027, 825 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ = 2.52 (t, J = 2.4 Hz, 1 H, CH), 4.64 (d, J = 2.4 Hz, 2 H, CH_2), 5.50 (br, 1 H, OH), 6.80 (m, AA'XX', 2 H, Ar), 6.88 (m, AA'XX', 2 H, Ar); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 56.7, 75.4, 78.8, 116.0, 116.3, 150.3, 151.5; Analysis calculated for $C_9H_8O_2$: C, 72.96; H, 5.44%. Found: C, 71.70; H, 5.40%.

1-(hydroxyhexafluoroisopropyl)-4-(propargyloxyhexafluoroisopropyl) benzene **3**: To a stirred mixture of 1,4-bis(2-hydroxyhexafluoroisopropyl) benzene (0.820 g, 2.0 mmol) and potassium carbonate (0.138 g, 1.0 mmol) in dimethylformamide (8 mL) at 60 °C was added propargyl bromide (0.117 g, 1.0 mmol). After three and a half hours, diethyl ether (75 mL) was added, the organic layer was washed with 10% HCl (50 mL), water (50 mL), dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded 1-(hydroxyhexafluoroisopropyl)-4-(propargyloxyhexafluoroisopropyl) benzene **3** (0.378 g, 84%, R_f = 0.52 in dichloromethane) as a white solid, m.p. = 58-60°C; FT-IR (neat) ν = 3486, 3317, 2963, 1258, 1171, 1154, 1100 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ = 2.59 (t, J = 2.4 Hz, 1 H, CH), 4.27 (d, J = 2.4 Hz, 2 H, CH_2), 7.75 (m, AA'XX', 2 H, Ar), 7.87 (m, AA'XX', 2 H, Ar); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 55.4, 76.0, 77.2, 82.8 (m), 122.0 (q, J = 290.0 Hz), 122.4 (q, J = 287.9 Hz), 127.3, 128.4, 129.9, 131.8; Analysis calculated for $C_{15}H_8F_{12}O_2$: C, 40.20; H, 1.80%. Found: C, 39.87; H, 1.79%.

2,2-(4-hydroxyphenyl)-(4′-propargyloxyphenyl)propane **4**: To a stirred mixture of bisphenol A (0.456 g, 2.0 mmol) and potassium carbonate (0.138 g, 1.0 mmol) in dimethylformamide (5 mL) at 60 °C was added propargyl bromide (0.117 g, 1.0 mmol). After two hours, diethyl ether (50 mL) was added, the organic layer was washed with 10% HCl (30 mL), water (30 mL), dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded 2,2-(4-hydroxyphenyl)-(4'-propargyloxyphenyl propane **4** (0.248 g, 50%, R_f = 0.29 in dichloromethane) as a colorless oil, FT-IR (neat) ν = 3378, 3286, 2968, 1608, 1508, 1447, 1364, 1297, 1218, 1178, 1027, 1014, 828 cm^{-1}; ^{1}H NMR ($CDCl_3$, 400 MHz) δ = 2.51 (t, J = 2.4 Hz, 1 H, CH), 4.66 (d, J = 2.4 Hz, 2 H, CH_2), 4.90 (br, 1 H, OH), 6.73 (m, AA'XX', 2 H, Ar), 6.88 (m, AA'XX', 2 H, Ar), 7.09 (m, AA'XX', 2 H, Ar), 7.15 (m, AA'XX', 2 H, Ar); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 31.1, 41.7, 55.8, 75.4, 78.8, 114.2, 114.7, 127.8, 128.0, 143.2, 144.1, 153.3, 155.4; Analysis calculated for $C_{18}H_{18}O_2$: C, 81.17; H, 6.81%. Found: C, 80.41; H, 6.87%.

2,2-(4-hydroxyphenyl)-(4′-propargyloxyphenyl)hexafluoropropane **5**: To a stirred mixture of 4,4′-(hexafluoroisopropylidene)diphenol (0.672 g, 2.0 mmol) and potassium carbonate (0.138 g, 1.0 mmol) in dimethylformamide (7 mL) at 60 °C was added propargyl bromide (0.117 g, 1.0 mmol). After three hours, diethyl ether (75 mL) was added, the organic layer was washed with 10% HCl (50 mL), water (50 mL), dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded 2,2-(4-hydroxyphenyl)-(4'-propargyloxyphenyl) hexafluoropropane **5** (0.348 g, 49%, R_f = 0.36 in dichloromethane) as a colorless oil, FT-IR (neat) ν = 3305, 2932,1613, 1514, 1238, 1205, 1132 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ = 2.55 (t, J = 2.4 Hz, 1 H, CH), 4.70 (d, J = 2.4 Hz, 2 H, CH_2), 5.92 (br, 1 H, OH), 6.83 (m, AA'XX', 2 H, Ar), 6.96 (m, AA'XX', 2 H, Ar), 7.25 (m, AA'XX', 2 H, Ar), 7.33 (m, AA'XX', 2 H, Ar); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 55.8, 63.5 (m), 75.9, 78.1, 114.3, 115.0, 124.3 (q, J = 284.7 Hz), 125.6, 126.4, 131.5, 131.7, 155.9, 157.7; Analysis calculated for $C_{18}H_{12}F_6O_2$: C, 57.76; H, 3.23%. Found: C, 57.73; H, 3.34%.

1,3-Dimethoxy-5-Propargyloxybenzene **6**: To a stirred mixture of 3,5-dimethoxyphenol (0.154 g, 1.0 mmol) and potassium carbonate (0.166 g, 1.2 mmol) in dimethylformamide (5 mL) at 60 °C was added propargyl bromide (0.143 g, 1.2 mmol). After four hours, diethyl ether (50 mL) was added, the organic layer was washed twice with water (30 mL), dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded 1,3-dimethoxy-5-propargyloxybenzene **6** (0.137 g, 71%, R_f = 0.62 in dichloromethane) as a white solid, m.p. = 42-43°C; FT-IR (neat) ν = 3238, 3014, 2945, 1594, 1477, 1459, 1377, 1198 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ = 2.52 (t, J = 2.4 Hz, 1 H, CH), 3.76 (s, 6 H, CH_3), 4.64 (d, J = 2.4 Hz, 2 H, CH_2), 6.12 (m, 3 H, Ar); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 55.4, 55.9, 75.6, 78.4, 93.8, 159.4, 161.5; Analysis calculated for $C_{11}H_{12}O_3$: C, 68.74; H, 6.29%. Found: C, 68.71; H, 6.25%.

4-(trifluoromethoxy)propargyloxybenzene **7**: To a stirred solution of 4-(trifluoromethoxy)-phenol (0.179 g, 1.0 mmol) and potassium carbonate (0.168 g, 1.2 mmol) in DMF (5 mL) at 60 °C was added propargyl bromide (0.143 g, 1.2 mmol). After four hours, the mixture was cooled down to room temperature, diluted with diethyl ether (50 mL) and washed with water (35 mL). The organic layer was washed with 10% hydrochloric acid (30 mL), twice with water (35 mL), dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded 4-(trifluoromethoxy)propargyloxybenzene **7** (0.177g, 82%, R_f = 0.76 in CH_2Cl_2) as yellow liquid; FT-IR (neat) ν = 3308, 1599, 1506, 1457, 1375, 1258, 1221, 1192, 1157, 1108, 1028 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ = 2.53(t, J = 2.4 Hz, 1 H, C≡C-H), 4.68 (d, J = 2.4 Hz, 2 H, O-CH_2), 6.94-7.17 (m, AA'XX', 4 H, Ar); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 56.2, 75.9, 78.1, 115.8, 121.8, 122.4, 143.4, 156.0; Analysis calculated for $C_{10}H_7F_3O_2$: C, 55.56; H, 3.26%. Found: C, 55.49; H, 3.28%.

3,4-dichloropropargyloxybenzene **8**: To a stirred solution of 3,4-dichlorophenol (0.160 g, 0.98 mmol) and potassium carbonate (0.167 g, 1.2 mmol) in DMF (5.0 mL) at 60 °C was added propargyl bromide (0.143 g, 1.2 mmol) within 10 minutes. After three and a half hours, the mixture was cooled down to room temperature, and diluted with diethyl ether (50 mL) and washed with water (35 mL). The organic layer was washed with 10% hydrochloric acid (30 mL), twice with water (35 mL), dried with magnesium sulfate, filtered and evaporated. Purification by column chromatography afforded 3,4-dichloropropargyloxybenzene **8** (0.139g, 69%, R_f = 0.77 in CH_2Cl_2) as a yellow oil; FT-IR (neat) ν = 3298, 2364, 2124, 1592, 1571, 1472, 1375, 1290, 1263, 1218, 1125, 1034, 1020 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ = 2.56 (t, J = 2.4 Hz, 1 H, C≡C-H), 4.69 (d, J = 2.4 Hz, 2 H, O-CH_2), 6.86 (dd, J_m = 2.9 Hz, J_o = 6.0 Hz, 1 H, Ar), 7.10 (d, J_m = 2.9 Hz, 1 H, Ar), 7.36 (d, J_o = 8.9 Hz, 1 H, Ar); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 56.3, 76.3, 77.6, 114.9, 117.0, 125.0, 130.7, 132.9, 156.5; Analysis calculated for $C_9H_6Cl_2O$: C, 53.77; H, 3.01%. Found: C, 53.97; H, 3.07%.

2-pyridinylmethyl 4-pentynoate **9**: To a stirred solution of 3-(hydroxymethyl)pyridine (0.109 g, 1.0 mmol) and 4-pentynoic acid (0.108 g, 1.1 mmol) in dichloromethane (20 mL) was added *N*-(3-dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride (0.211 g, 1.1 mmol) and dimethylaminopyridine (0.006 g, 0.05 mmol) at 0 °C. The solution was stirred for an hour at 0 °C, then overnight at room temperature. The organic layer was washed with saturated aqueous sodium bicarbonate, brine, dried with magnesium sulfate, filtered and evaporated. After purification by column chromatography, 2-pyridinylmethyl 4-pentynoate **9** (0.185 g, 98%, R_f = 0.31 in diethyl ether) as a clear oil, FT-IR (neat) ν = 3293, 2925, 1734, 1428, 1158 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ = 1.96 (t, J = 2.7 Hz, 1 H, CH), 2.51 (m, 2 H, CH_2), 2.60 (m, 2 H, CH_2), 5.16 (s, 2 H, O-CH_2), 7.29 (m, 1 H, Ar), 7.69 (m, 1 H, Ar), 8.57 (m, 1 H, Ar), 8.61 (m, 1 H, Ar); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 14.3, 33.2, 63.9, 69.3, 82.2, 123.5, 131.4, 136.1, 149.7, 171.5; Analysis calculated for $C_{11}H_{11}NO_2$: C, 69.83; H, 5.86; N, 7.40%. Found: C, 68.72; H, 5.98; N, 7.21%.

3-Butynyl Methyl Succinate **10**: To a stirred solution of monomethyl succinate (0.145 g, 1.1 mmol) and 3-butynol (0.070 g, 1.0 mmol) in dichloromethane (10 mL) was added a solution of *N*-(3-dimethylaminopropyl)-*N*′-ethylcarbodiimide hydrochloride (0.211 g, 1.1 mmol) and dimethylaminopyridine (0.012 g, 0.1 mmol) in dichloromethane (1 mL) at 0 °C. The solution was stirred for an hour at 0 °C, then overnight at room temperature. The organic layer was washed with saturated aqueous sodium bicarbonate, brine, dried with magnesium sulfate, filtered and evaporated. After purification by column chromatography, 3-butynyl methyl succinate **10** (0.155 g, 92%, R_f = 0.66 in ethyl acetate) as a clear oil, FT-IR (neat) ν = 3279, 2957, 1733, 1439, 1321, 1209, 1161 cm^{-1}; ^1H NMR ($CDCl_3$, 400 MHz) δ = 1.98 (t, J = 2.7 Hz, 1 H, CH), 50 (dt, J = 6.8 Hz, J = 2.7 Hz, 2 H, C-CH_2), 2.62 (m, 4 H, CO-CH_2), 3.66 (s, 2 H, O-CH_3), 4.17 (t, J = 6.8 Hz, 2 H, O-CH_2); ^{13}C NMR ($CDCl_3$, 100 MHz) δ = 18.9, 28.8, 29.0, 51.9, 62.3, 69.9, 79.9, 172.0, 172.7; Analysis calculated for $C_9H_{12}O_4$: C, 58.69; H, 6.57%. Found: C, 58.62; H, 6.50%.

"Click" Reaction on Microcantilevers: Two gold-coated microcantilever chips (Nanoworld, 10 nm chromium and 30 nm gold)) were mounted on a custom-made PEEK chip holder and washed successively by immersion in 1N HCl, water, ethanol, hexane and ethanol. After immersion in a bis(11-azido undecanyl)disulfide solution (1 mM in ethanol) overnight, they were rinsed with ethanol, hexane and dried under a stream of nitrogen. A solution of the desired alkyne (5 mM) in isopropanol and water (1:1) containing sodium ascorbate (15 mol%) and copper sulfate (1 mol%) was sonicated for a minute. The azide chips were immersed in this mixture overnight. After the "click" reaction, the substrates were rinsed with ethanol, water, ethanol, hexane and dried under a gentle stream of nitrogen.

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