A Fluorescence-Based Synthetic LPS Sensor
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Abstract: For the detection of bioanalytes, there is an ongoing search for synthetic sensors to replace enzyme-based assays which are sensitive to contaminants or suboptimal storage conditions. Lipopolysaccharide (LPS), a bacteria-borne endotoxin that may lead to life-threatening conditions such as septic shock, is one such case. Fluorescently labeled analogues of two peptide variants derived from the putative ligand-binding domain of the LPS-binding protein CD14 were developed that detect and discriminate LPS and lipids down to the submicromolar concentration range. Peptides are terminally labeled with carboxyfluorescein and tetramethylrhodamine. For one given peptide, sensitivity and specificity for the detection of LPS and discrimination from other lipids are achieved by spectral signatures that combine changes in the fluorescence resonance energy transfer (FRET) between both dyes and the total emission of tetramethylrhodamine. Alternatively, specificity is obtained by combining the FRET efficiencies of both peptide variants. In comparison to published synthetic LPS sensors, the CD14-derived sensors yield an increase in sensitivity by about 3 orders of magnitude and exhibit specificity for analytes for which the design of synthetic recognition elements is a challenging task. Moreover, one of the sensors enabled the detection of LPS in the presence of up to 50% fetal calf serum, thereby demonstrating the feasibility of this peptide-based approach for clinically relevant samples.

Introduction
In the Western world, sepsis and septic shock are major causes of death, accounting for about 150 000 casualties each year alone in the United States. These conditions can be initiated by the massive release of lipopolysaccharide (LPS) from the cell wall of gram-negative bacteria, which then inappropriately activates the innate immune system.1 Due to the high toxicity of LPS, continuous effort is directed toward the development of specific LPS detection systems that are able to trace the smallest amounts of LPS. Currently, the enzymatic Limulus amebocyte lysate (LAL) assay is used most widely to detect and quantify endotoxin in clinical and nonclinical samples. However, there are several limitations to this procedure. The LAL assay, as with most enzymatic tests, is very sensitive but highly susceptible to changes in temperature and pH. Moreover, carbohydrate derivatives other than LPS, such as β-glucans, also react positively in this assay.2 A variety of alternatives to the LAL assay have been developed, but none of them has been introduced into praxis because of poor sensitivity or inconvenience.3 Synthetic small molecule sensors have been developed for a number of analytical purposes.4–6 Such sensors enable a modification of sensitivity and specificity by chemical approaches. To this point, however, a synthetic LPS sensor with sensitivity in the submicromolar range has not been presented.

Ideally, a biomedical assay should enable the highly sensitive and specific detection of the analyte and be robust to changes in the molecular environment and prolonged storage. Sensors based on small synthetic receptors are considered a highly promising strategy for this purpose. Ion-selective fluorophores that change their spectral characteristic upon binding of analyte are one classical example for synthetic sensors.5 Aptamers, short RNA or DNA oligonucleotides, have been generated for the detection of small analytes and proteins.4,6 As for natural biological receptors, in most cases, binding of analyte leads to a conformational change that is linked to a transducer that reports on the binding of the analyte by a physical readout. Fluorescence resonance energy transfer (FRET) provides a sensitive readout for recognition elements in which binding of the analyte induces a conformational change.7,8

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In addition to the oligonucleotide-based aptamers, peptide-based recognition elements have been exploited in a number of applications, typically using changes in FRET as the readout. In contrast to the aptamers, which are identified by screens of large collections of molecules, these recognition elements mostly incorporate known substrates or ligand-binding domains. For the detection of lipids, a sensor based on a recombinant fusion protein in which the lipid-binding peptide derived from ActA was flanked by cyan and yellow fluorescent proteins at both ends was recently presented. Binding of polyphosphorylated phosphoinositides induces a random coil to helix transition that increases the separation of the terminal fluorophores, leading to a decrease in FRET.

For LPS, there has only been very limited progress in the development of small molecule-based biosensors. Lipid-functionalized polydiacetylene liposomes were applied to the discrimination of LPS from different species and of unrelated lipids based on a colorimetric change. However, LPS concentrations at around 100 µM were employed. Recently, we identified an LPS-binding 20 amino acid peptide amide from the LPS-binding domain of the LPS receptor CD14. CD14 is a 55 kDa glycoprotein that directly binds to LPS and is required for the LPS-dependent activation of Toll-like receptor (TLR) 4, the transmembrane molecule that initiates the intracellular signaling cascade in response to LPS. For LPS at a concentration of about 2 nM, the peptide amide CD14 (81–100) exhibited an LPS-neutralizing capacity at submicromolar concentrations. Three leucines at positions 7, 11, and 14 were found to be essential for activity. Exchange of amino acid residues in positions 5, 9, 13, and 17 of the native peptide CD14 (81–100) for lysine residues and proline residue 3 for an alanine residue led to a more active analogue with higher solubility in aqueous buffers. In the LAL test, this analogue had an IC_{50} value of about 10 µM.

Evidence was obtained that binding of LPS to the 20 amino acid peptide induces a random coil to helix transition. In aqueous buffers, the peptide and its analogue were present as a random coil. Upon addition of TFE, a membrane-mimicking organic solvent, the peptides assumed an α-helical conformation corresponding to the conformation of the corresponding segment in the whole protein. Such a random coil to helix transition has also been described for the well-known LPS-binding peptide magainin when binding to LPS.

Given the high LPS-neutralizing activity and the potential LPS-induced conformational change, we considered CD14 (81–100) and its analogue as promising candidates for LPS sensors using FRET for the detection of binding.

In this study, the synthesis and functional characterization of carboxytetramethylrhodamine (Tamra)/fluorescein-labeled LPS sensors based on the CD14-derived peptide are described. Tamra was attached to the N-terminus and carboxyfluorescein to the C-terminus. It has been reported previously that, for this combination of fluorophores, when attached to a conformationally unconstrained peptide, the fluorescence of both fluorophores is quenched by a mechanism involving dimer formation. Any conformational change of the peptide that induces the separation of the fluorophores decreases the quenching so that FRET may be detected. In the presence of LPS, the CD14-derived fluorescent peptides alter their spectral characteristics with respect to both FRET and the fluorescence intensity of Tamra. Selectivity of these sensing elements was established by probing the response to a series of lipids and lipopeptides. Detection of LPS was achieved for as little as 150 nM, thereby exceeding previously published synthetic LPS sensors by about 3 orders of magnitude. Finally, for the sensor in which several amino acids were exchanged for cationic residues, the detection of LPS was achieved in the presence of fetal calf serum (FCS), a condition mimicking highly complex samples of clinical relevance.

**Experimental Section**

**Reagents.** Lipopolysaccharide from *Escherichia coli* (phenol-extracted and purified by ion-exchange chromatography) was from Sigma (Taufkirchen, Germany). The molecular weight of commercial LPS varies between 3 and 20 kDa. In our calculations, we assumed a molecular weight of 10 kDa. The water-soluble S-[2,3-bis(palmitoyloxy)propyl]-N-palmitoyl-(R)-cysteinyl-(S)-seryltetrasyaline (PamCS-SK), S-[2,3-bis(palmitoyloxy)propyl]-N-palmitoyl-(R)-cysteinyl-(VPGVG)\_4 VPGKG-NH\_2 (PamCys-(VPGVG)\_4 VPGKG-NH\_2) (BCI-009), and the diacylated lipopeptide S-[2,3-bis(palmitoyloxy)propyl]-(R)-cysteinyl-GNDESNISPFEK (PamCys-GNDESNISPFEK) with the sequence of MALP2-22 were synthesized as described. Chemicals were from Fluka (Deisenhofen, Germany) and Merck (Darmstadt, Germany) with the exception of Fmoc amino acids (Novabiochem, Lautelfingen, Switzerland; Sem Chemicals, Dielsdorf, Switzerland, and Orpegen Pharma, Heidelberg, Germany). 5(6)-Carboxyfluorescein (Fluo) and 5(6)-carboxytetramethylrhodamine (Tamra)-N-succinimidyl ester were from Fluuka. 1-Palmitoyl-2-oleoylphosphatidylcholine and 1-palmitoyl-2-oleoyl-phosphatidylethanol were from Avanti Polar Lipids (Alabaster, AL).

**Solid-Phase Synthesis of Tamra-CD14 (81–100)-Fluo.** The doubly labeled peptides Tamra-QVPAQLLVLGALRLVSLRKLK-(Fluo)-NH\_2 (sensor 1) and Tamra-QVAAKLVLKVLRLKLYRKLK-(αFluo)-NH\_2 (sensor 2) were synthesized using a previously described (Fluo-Tru)-Lys-Rink amide resin. The carboxyfluorescein is attached to the N\_\_amino group of the resin-bound lysine, and the peptide was assembled on the N\_\_amino group of the lysine residue. The 20 amino acid peptide amides were assembled on this resin in a 15 µmol scale according to published protocols for the automated solid-phase synthesis.

of peptide. A sensor 1 analogue lacking the C-terminal fluorescein QVPAQLLVGLAVLRSKLL-NH₂ was synthesized on Rink amide resin. The resin-bound and side-chain-protected peptides (5 μmol) were then reacted with 5(6)-carboxytetramethylrhodamine-N-succinimidyl ester (10 μmol, 5.3 mg) in N,N′-dimethylformamide (200 μL) containing N,N′-disopropylethylamine (26 μmol, 4.3 μL). After 16 h, the resin was thoroughly washed. Peptides were cleaved off the resin and side chains deprotected by treatment with trifluoroacetic acid/triisopropylsilane/H₂O (92:5:2.5:5, v/v/v/v) for 4 h. Crude peptides were precipitated by adding cold diethyl ether (−20 °C). The precipitated peptides were collected by centrifugation and resuspended in cold diethyl ether. This procedure was repeated twice. Finally, the peptides were dissolved in acetonitrile/water, lyophilized, and analyzed by HPLC and MALDI-MS. Tamra-QVPAQLLVGLAVLRSKLLK(α(Fluo))NH₂: purity 80% (HPLC, 214 nm), calcd [M+H]⁺ = 3094.7 Da, expt [M+H]⁺ = 3094.6 Da. Tamra-QVAAKLLVKALRKLAYKRLK(α(Fluo))NH₂: purity 80% (HPLC, 214 nm), calcd [M+H]⁺ = 3209.5 Da, expt [M+H]⁺ = 3209.5 Da. Tamra-QVPAQLLVGLAVLRSKLLK(α(Fluo))NH₂: purity 80% (HPLC, 214 nm), calcd [M+H]⁺ = 2608.2 Da, expt 2608.8 Da. The fluorescent peptide amides were then purified by preparative HPLC to purities >95%.

**Peptide Stock Solutions.** Doubly labeled peptides were dissolved in DMSO to concentrations of 10 mM. Peptide concentrations were determined by U/V-vis spectroscopy of a further dilution (1:100) in methanol. Absorptions were measured at a wavelength of 540 nm (ε = 95 000 l/(mol·cm)).

**Preparation of Lipids and Fluorescence Spectroscopy.** Lipopolysaccharides were dissolved in DMSO (10 mM), lipopolysaccharides in water (5 mg/mL), and phospholipids in chloroform (32 mM). The phospholipids were further diluted in DMSO (500 μM). All lipids were sonicated at 40 W using a Sonorex Super RK 510H sonicator (Bandelin, Berlin, Germany) until solubilization as indicated by a clear solution. Doubly labeled peptides were diluted in PBS to a concentration of 100 nM. The solution of doubly labeled peptide (400 μL) was transferred to an Eppendorf reaction tube, and lipids were added to a final concentration of 2 μM. Titration experiments were performed in 96-well round-bottom microtiter plates. The samples were incubated for 10 min on ice and subsequently for 45 min on a shaker at 37 °C. After incubation, probes were subjected to fluorescence spectroscopy performed in quartz cuvettes using an LS50B luminescence spectrometer (Perkin-Elmer, Norwalk, CT). The excitation wavelength was set to 492 nm; emission spectra of the samples were recorded immediately.

**Measurements in FCS-Containing Buffers.** Fetal calf serum was diluted in PBS as indicated, and consecutively sensors 1 or 2 (100 nM) and LPS (2 μM) were added to the respective solutions. The samples were incubated for 10 min on ice and for at least 45 min at 37 °C on a shaker. Fluorescence spectroscopy was performed as described above. Furthermore, a mixture of LPS (40 μM) and FCS (100%) was diluted in PBS as indicated, and subsequently sensor 2 (100 nM) was added to the samples. The incubation was performed for 10 min on ice and for at least 45 min at 37 °C on a shaker. Probes were subjected to fluorescence spectroscopy as described above.

**Results**

The native sequences of the CD14-derived peptide and an analogue with higher solubility were selected as recognition elements for small molecule LPS sensors (Figure 1). The native sequence (sensor 1) corresponds to amino acids 81–100 of human CD14. In the analogue, residues 5, 9, 13, and 17 of the native peptide were replaced by lysine residues, and residue 3 was replaced for an alanine residue. It was shown by an alanine scan that these residues were not required for the LPS-neutralizing activity. We reasoned that the increased solubility would be beneficial for its application as a sensor. Furthermore, we were interested to learn whether both molecules exhibited different sensitivity for LPS and possibly also different specificity for related amphiphilic ligands (Figure 1) due to the enhanced positive net charge of sensor 2.

In buffer, both molecules displayed a two-peak fluorescence spectrum upon excitation at 492 nm (excitation of fluorescein) (Figure 2A,B). Upon addition of lipopolysaccharide (E. coli, serotype O111:B4), both sensors altered their spectral characteristics leading to a reduced fluorescence emission at 520 nm and a significantly enhanced emission at 588 nm, indicating an increase in FRET. For the lipid sensor based on ActA, the fluorophore separation due to ligand binding resulted in a decrease in FRET.

For our sensors, the spectral changes in the presence of LPS should therefore also be a consequence of dimer quenching due to spatial separation of fluorophores. To assess to which degree environment-dependent changes in fluorescence quantum yield due to interaction of the fluorophores with the amphiphilic ligand also contributed to the spectral changes, a sensor 1 analogue (sensor 3) was synthesized that lacked the fluorescein moiety (Figure 2C). Despite the absence of a potential energy donor, in the ligand-free state sensor 3 exhibited a stronger rhodamine emission than sensor 1, demonstrating that the rhodamine fluorescence was nearly completely quenched due to dimer formation. The presence of environment-dependent effects on the quantum yield of rhodamine was confirmed by the 3.5-fold increase in sensor 3 fluorescence upon addition of LPS.

To determine the maximal loss of quenching of fluorescein fluorescence and to obtain further information on the dimer-induced quenching of rhodamine fluorescence, sensor 1 was digested by proteinase K (10 μg/mL). Proteinolysis induced a strong increase in the intensity of the fluorescein emission at 520 nm (Figure 2D) and of the Tamra emission at 588 nm (Figure 2E) when the fluorophores were excited at 492 and 541 nm, respectively. These results demonstrate that, in the LPS-free state, fluorescence of both fluorophores is nearly completely quenched.

For the single labeled peptide, binding of LPS already led to an environment-dependent increase in fluorescence quantum yield. However, the increase of intensity alone is little resistant to changes in the concentration of a sensor containing only one fluorophore. In contrast, for doubly labeled peptides, the ratio of emission intensities is independent of concentration. For each of the functional states of the sensors (ligand-free, LPS-bound, digested), a specific ratiometric value was obtained (Figure 2F).

Interestingly, the emission ratios for both sensors in the LPS-free as well as in the LPS-bound state also differed significantly. For sensor 2, the ratio was larger than for sensor 1.

Next, we tested the concentration dependence of the LPS-dependent increase in emission ratios (Figure 3). For sensor 1, the ratio increased linearly with concentration over a concentration range of up to 1.5 μM LPS. Saturation was observed at concentrations higher than 2 μM. The detection limit of LPS

was about 150 nM. In contrast, sensor 2 exhibited a steeper increase in the ratiometric values at concentrations between 0.5 and 1.5 \( \mu \text{M} \), a larger maximum ratiometric value, but also a higher detection limit (about 0.5 \( \mu \text{M} \)) in comparison to sensor 1 containing the native sequence (Figure 3C).

In addition to sensitivity, a sensor is characterized by its specificity. To determine whether the sensors were able to discriminate LPS from other molecules with similar physicochemical characteristics, their fluorescence emission spectra were recorded for increasing concentrations of several amphipathic lipids. These included LPS from *E. coli*, the neutral and negatively charged phospholipids POPC and POPG, and di- and triacylated lipopeptides. The latter comprised the bacterial lipopeptide MALP-2\(^{22}\) and the synthetic lipopeptides Pam\(_3\)Cys-SK\(_4\),\(^{23}\) and BCI-009, which only differed in their peptide moiety (Figure 1).

Since both sensors exhibit characteristic differences in the recognition of varying concentrations of LPS (Figure 3), we hypothesized that the sensors also differentially recognize these other lipids. For both sensors, the concentration dependence of the fluorescence ratios was determined (Figure 4A,B). The steepest increase and the highest ratiometric values were obtained in the presence of LPS and the lipopeptide Pam\(_3\)Cys-SK\(_4\). In contrast, the maximum values evoked by BCI-009, MALP-2, and the phospholipid POPC were significantly lower (3, 4, and 1, respectively), and the curves exhibited a flatter increase. Pam\(_3\)Cys-SK\(_4\) with a positively charged peptide moiety...
induced a stronger change than BCI-009 with a hydrophobic moiety containing only one lysine residue. Interestingly, POPG was differentially recognized by the sensors. Whereas sensor 2 exhibited a steep increase of the ratiometric values and a high maximum value, sensor 1 exhibited a curve comparable to that for MALP-2. In contrast, the neutral phospholipid POPC was detected neither by sensor 1 nor by sensor 2. However, with these different sensitivities for the panel of ligands alone, it would be impossible to discriminate a high concentration of, for example, POPG from a low concentration of LPS.

Therefore, we next plotted the ratiometric values as a function of the fluorescence emission at 588 nm (Figure 4C,D) to assess whether the concentration-dependent alterations of the ratio and the corresponding fluorescence emission at 588 nm behaved in a distinctive manner for the different analytes. This was indeed the case: For sensor 1, LPS evoked the steepest increase in the emission ratios in relation to the increase of emission at 588 nm in comparison to the other lipids, resulting in a separation of its curves from those of POPC, POPG, MALP-2, and BCI-009. For lipopeptide BCI-009, the curve was separated from those of all other lipids, due to a significant weaker increase in the emission ratio in relation to the emission at 588 nm. For LPS and Pam3 Cys-SK4, the curves intersected, indicating that the sensor does not discriminate both analytes over the whole concentration range. At high Pam3 Cys-SK4 concentrations, high emission ratios were accompanied by significantly higher emission intensities at 588 nm. The two triacylated lipopeptide analogues Pam3Cys-SK4 and BCI-009 were discriminated for the major part of the concentration range. For sensor 1, consideration of the two-dimensional spectral signature (ratio and emission at 588 nm) significantly improved the discrimination of analytes. In contrast, for sensor 2 the spectral signatures were less well separated. Most remarkably for BCI-009, a much smaller increase in emission intensity at 588 nm was observed.

Finally, we tested to which degree a combination of the emission ratios for both sensors enabled a discrimination of ligands (Figure 4E). For most lipids, both sensors exhibited comparable ratiometric signals. Only for POPG the sensitivity of sensor 2 was significantly enhanced compared to that of sensor 1 reflected by a steeper increase of the fluorescence ratio at POPG concentrations below 5 μM. The separation of the curves was comparable to the one obtained by combining the spectral signatures for one sensor alone.

The results presented above had shown that the spectral characteristics for the sensors in the presence of LPS differ from the ones of other lipids over a wide concentration range. However, these experiments did not enable us to predict whether the sensors possess this discriminatory ability also in the presence of a complex mixture of lipids. We therefore asked whether LPS is also detectable under more complex and clinically relevant conditions (e.g., in the presence of FCS). Moreover, proteolytic degradation was also a concern.

In a first experiment, the sensors were incubated with various concentrations of serum, and finally LPS was added at a
The lipopolysaccharide was premixed with the serum. In this case, interactions of LPS with serum lipids could compromise the detection by our peptide. LPS (2 μM) in 6.25% FCS corresponds to 32 μM in 100% FCS, FCS was premixed with 40 μM LPS. This mixture was diluted, and sensor 2 was added to the samples. For this condition, LPS could already be detected in a 1:2 dilution, that is, 20 μM LPS in 50% FCS (Figure 5B). At FCS concentrations higher than 25%, the detection again benefited from a parallel consideration of the ratio of fluorescence at 588 versus 520 nm and the emission intensity at 588 nm. Considering the emission ratios alone, a detection had only been possible for FCS concentration lower than about 15%.

**Discussion**

The peptide corresponding to amino acids 81 to 100 of human CD14-labeled N-terminally with Tamra and C-terminally with fluorescein and its more soluble analogue exhibited significant changes in FRET in the presence of submicromolar to lower micromolar LPS concentrations. The spectral characteristics of the sensors differed for LPS and a set of lipids and lipoids. Moreover, sensor 2 enabled the detection of LPS in the presence of 50% FCS, a highly complex mixture of lipids. These results demonstrate the successful realization of a synthetic LPS sensor based on an LPS-neutralizing peptide.

The high LPS-neutralizing activity of these 20 amino acid peptide amides matched their sensitivities as LPS sensors. Rather unexpectedly for the sensing of amphiphilic ligands by a peptide that may assume an amphipathic α-helical conformation upon ligand binding, the sensors readily discriminated LPS from the lipopeptide (MALP-2) derived from mycoplasmic lipoprotein, and from further synthetic lipopeptide analogues and lipids. Interestingly, not only the ratio of fluorescence emissions at 588 and 520 nm depended on the presence of ligand but also the emission intensity at 588 nm. The discrimination of the set of lipophilic compounds over the major part of the tested concentration range was achieved either by combination of emission ratios and emission at 588 nm for any given sensor or by combination of emission ratios for both sensors. For instance, when the ratio of the emission was plotted against the concentration discrimination between LPS and the lipopeptide Pam3Cys-SK4 was not possible. However, the lipopeptide evoked a significantly enhanced fluorescence emission at 588 nm compared to LPS, which could be consulted for discrimination between the two molecules.

The sensors possessed remarkable discriminatory ability for structurally similar ligands. The zwitterionic phospholipid POPC was not detectable by either sensor. In contrast, the spectral characteristics of both sensors were strongly affected by the negatively charged POPG, with sensor 2 being more sensitive than sensor 1. This enhanced sensitivity of sensor 2 might be due to additional electrostatic interactions between the positively charged sensor and the negatively charged phospholipid. Similarly, the triacylated lipopeptides Pam3Cys-SK4 and BCI-009 and the diacylated lipopeptide MALP-2 were readily discriminated.

In contrast to the rather subtle differences in their response to different ligands when present in isolation, both sensors showed striking differences in the detection of LPS in the presence of fetal calf serum. Both sensors showed an increase in fluorescence emission at 588 nm. However, only for sensor 2 an increase was observed in emission ratios and emission at
588 nm observed. Interestingly, even though containing a complex mixture of lipids, FCS induced a particular response profile, that is, an increase in fluorescence at 588 nm in the presence of constant emission ratios, different from the one of all defined lipids included in our study. Only for sensor 1 was the response profile similar to the one obtained for the lipopeptide BCI-009.

Compared to synthetic LPS sensors published before, the CD14-derived peptides constitute an improvement in sensitivity by 3 orders of magnitude. The remaining gap in sensitivity and specificity between synthetic peptide-based LPS recognition elements and enzymatic assays could now be closed by the combination of two approaches. First, our comparison of the CD14-derived peptide and its analogue has demonstrated already
that sequence variation allows a tailoring of the sensitivity and specificity profile. Therefore, further improvements are expected through sequence variations. In correspondence to the increase in specificity afforded by the combination of emission ratios for our pair of sensors, the combination of readouts for even more sensor variants, in the form of sensor arrays, should further increase the specificity.

Finally, instead of using changes in the spectral characteristics, conformational changes of a sensing element may be coupled to highly powerful signal amplification mechanisms. The exploitation of the recognition of LPS provided by nature in the form of the CD14-derived LPS-binding domain therefore provides a promising starting point for the further development of alternatives for the enzymatic LPS assays. The successful detection of LPS in the presence of serum demonstrated that proteolytic degradation that may limit the application of a peptide-based recognition element in complex biological materials was not a concern.

Conclusions

In summary, we show that an LPS-binding and -neutralizing domain of the LPS receptor CD14 constitutes a highly sensitive and specific recognition element for a fluorescence-based LPS sensor. Combination of two spectral readouts, either from one sensor or from two sensor variants, enables the discrimination of different lipophilic analytes for which the development of synthetic receptors is a challenging task. Sequence variations change the dose–response profile as well as the specificity profile of the sensors, demonstrating a tailoring of the response characteristics through structural modifications. In comparison to previous synthetic LPS sensors, these sensors, derived from a natural LPS receptor, constitute an increase in sensitivity by 3 orders of magnitude. In addition to discriminating between lipophilic analytes when present in isolation, one of the sensors also enabled the detection of LPS in the presence of high concentrations of fetal calf serum, a condition mimicking samples with clinical relevance.

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