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Enhancement of a Catalysis-Based Fluorometric Detection Method for Palladium through Rational Fine-Tuning of the Palladium Species

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Abstract: Metal analyses in chemistry, materials science, and environmental science are currently performed using techniques such as inductively coupled plasma mass spectrometry and X-ray fluorescence, which require expensive instrumentation and are not high-throughput. Although fluorescent probes are known for their sensitivity and specificity and are amenable to high-throughput analyses, the robustness of such analyses are typically limited due to their binding-based nature. Herein we report an improvement of our previously reported catalysis-based fluorescent probe for palladium by rationally fine-tuning the redox and coordination chemistries of the palladium species involved in the *O*-deallylation reaction. This method now rivals current analytical methods with respect to sensitivity. We demonstrate palladium detection in various active pharmaceutical ingredients, spent catalytic converter materials, and a metal scavenger resin. Thus, fluorescent methods may have the potential for substituting the current instrument-intensive techniques.

Introduction

Many chemosensors noncovalently bind to an analyte to produce a fluorescence signal (Figure 1a) and have become powerful tools in cell biology.[1] For example, the first calcium sensor[2] and Zinpyr-1[3] were developed on the basis of binding-induced fluorescence off−on switches and have had significant impact. In contrast, applications of fluorescent probes in crude samples are rare in the literature. Instead, for nonliving samples, inductively coupled plasma mass spectrometry (ICP-MS) continues to be a widely used method because of its superior sensitivity and robustness.

Such dominance is particularly prominent in palladium analysis in chemistry,[4] materials science,[5] and environmental science.[6] In chemistry, Pd-catalyzed reactions represent powerful transformations for the synthesis of complex molecules, including many drugs (refecoxib, eniluracil, cefprozil).[4] Despite the frequent and fruitful use of such reactions, one major setback

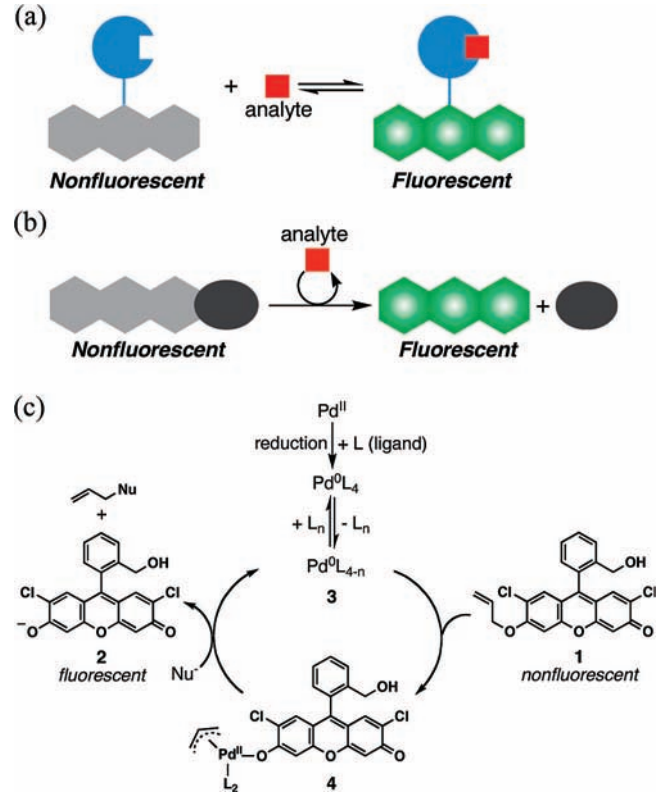

Figure 1. Binding- versus catalysis-based fluorescent probes. (a) Binding-based sensing approach. (b) Catalysis-based detection approach. (c) Catalytic mechanism of Pd detection method.

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is the high level of palladium in the resultant compounds.[7,8] The metal has the potential to bind to proteins, DNA, and other biomolecules and thus may be a health hazard.[9] As a result, the proposed maximum dietary intake of palladium is <1.5−15 μg/day per person, and its threshold in drugs is 5−10 ppm.[7] Because compounds synthesized by means of Pd-catalyzed reactions often contain much more palladium than this threshold limit (typically 300−2000 ppm), extensive purification is required, starting with medium- to large-scale screening of scavenging methods to remove these impurities.[10,11] During this process, it is critical to quickly evaluate and prioritize these palladium-scavenging techniques.

Although highly sensitive,[12] ICP-MS analyses suffer from the high cost of the instrument, isotope effects, and spectral and nonspectral interferences due to matrix effects.[13,14] Additionally, use of the instrument requires highly trained individuals, and reliability and interpretation of results is dependent upon the operator.[13] The assessment of many samples is time-consuming, especially since scrupulous washings must be carried out between samples to minimize cross-contamination.[15] Thus, the ICP-MS approach is not high-throughput. These shortcomings warrant the development of alternative methods for metal analyses.

One approach for developing a sensitive fluorometric detection method is to amplify the fluorescence signal (Figure 1b). In this approach, the analyte catalyzes a chemical transformation that produces a fluorescence signal. Due to the catalytic nature of the analyte, the signal will be amplified. This has been widely used in biology (e.g., reporter gene assays,[16] enzyme assays,[17] and cat-ELISA[18]) but rarely in chemistry. Recently, our group reported a catalysis-based fluorometric method for palladium[19,20] based on the Tsuji−Trost allylic oxidative insertion mechanism (Figure 1c).[21,22] Key to this method is the initial formation of a coordinatively unsaturated palladium(0) species **3**, which inserts into the nonfluorescent allylic ether **1** to form the putative π-allylpalladium complex **4**. This complex then reacts with a nucleophile to form the fluorescent compound **2** and an allylated nucleophile. This may be the first example where a metal analyte amplifies the fluorescence signal directly through a catalytic process.[23]

Despite the conceptual novelty of palladium detection, the turnover frequency (TOF) in the previous method was only ∼1.8 h^{-1}. In other words, one molecule of palladium affords 1.8 molecules of the fluorescent compound **2** per hour under the reaction conditions, not fully exploiting the potential of the catalytic system. In this method, Ph_3P was used both as a reducing agent and as a palladium ligand. We speculated that the use of electron-rich Ph_3P might account for the low TOF because Ph_3P presumably stabilizes the catalytically inactive, coordinatively saturated palladium species Pd^0L_4 (Figure 1c) and may be detrimental for the generation of coordinatively unsaturated, catalytically active palladium species Pd^0L_{4-n}.[22,24] Another major problem was that the previous method was incapable of detecting palladium in the presence of sulfides, a commonly used functional group in drugs (e.g., penicillin) and drug synthesis.[4b] This is presumably because sulfides are a better ligand than phosphines for palladium, prompting us to consider a more competitive ligand against sulfur that does not interfere with the oxidative insertion of the metal into the allylic ether bond. Here we report solutions to both of these limitations through the use of an electron-poor phosphine ligand to better generate the coordinatively unsaturated palladium species.[25,26] In this new method, $NaBH_4$ reduces Pd^{II} to Pd^0 more rapidly than Ph_3P and competes favorably against sulfides since hydrides are among the best ligands for palladium.

Results and Discussion

Phosphine/$NaBH_4$ Screening. In order to improve our palladium detection method,[27] we first screened phosphine and phosphite ligands in the absence and in the presence of $NaBH_4$.[28] Screening in the presence of $NaBH_4$ was particularly illuminating because we could interpret the effects of phosphine reagents solely as ligands and not as reducing agents. $NaBH_4$ was chosen as a reducing agent because of its known ability to efficiently reduce Pd^{II} to Pd^0 (Figure 1c),[25,29] which may enhance turnover. As Figure 2a shows, our screening efforts identified two phosphine ligands, JohnPhos and tri-2-furylphosphine (TFP), that significantly enhanced the Pd-catalyzed deallylation of **1** relative to Ph_3P.[19] Of particular interest was the combination

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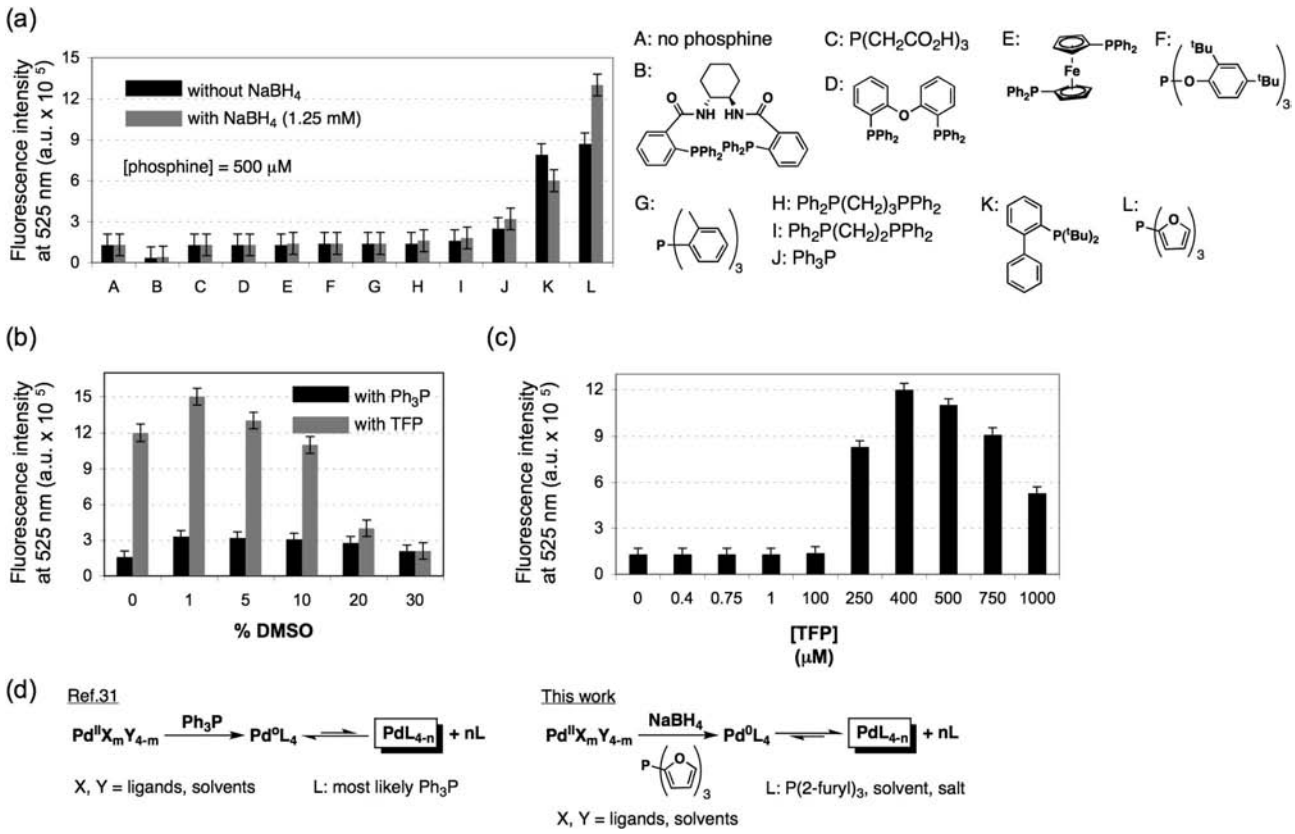

Figure 2. Phosphine screening and TFP optimization. [**1**] = 12.5 μM and [Pd] = 100 nM, and the assays were performed for 1 h at 24 °C in DMSO/pH 7 buffer (5:95 for (a) and (c)) before fluorescence measurement. (a) Phosphine screening. (b) Effect of % DMSO on phosphines with $NaBH_4$. (c) Correlation between fluorescence intensity and [TFP]. (d) Conversion of all Pd^{II} species to a uniform Pd^0 species.

of $NaBH_4$ and electron-poor TFP that gave a much greater fluorescence signal than the combination of Ph_3P and $NaBH_4$ and than Ph_3P alone, thus supporting our hypothesis.

TFP is a widely used ligand in the Stille reaction because its low electron density[30] enhances the rate of transmetalation via facile dissociation.[31] However, TFP has generally been reported to be a poor ligand in Pd-catalyzed allylic alkylation chemistry,[32] especially when phosphine concentrations are low (<6 equiv relative to palladium) in THF. This observation was attributed to a decreased reactivity of $Pd(TFP)_2$ compared to $Pd(PPh_3)_2$ for the oxidative addition.[33] However, in the presence of large excesses of phosphine (>6 equiv relative to palladium) in THF or DMF, the concentration of $Pd(TFP)_2$ was considerably higher than that of $Pd(PPh_3)_2$, resulting in a higher rate of allylic alkylation chemistry.[33] Amatore and co-workers speculate that the rate enhancement by TFP is likely due to faster nucleophilic attack in THF and the combination of faster oxidative addition and nucleophilic attack in DMF.[33] Although it is unclear how TFP accelerates the Pd-catalyzed Tsuji−Trost-type reaction in aqueous solution, the role of TFP in our method is likely similar (Figure 2d), facilitating dissociation from the metal center to allow available d-orbitals for the oxidative insertion and promoting nucleophilic turnover of the π-allylpalladium species.[34] This result also implies potential improvement in the Pd-catalyzed allylation in aqueous media.[35]

Using TFP, we next examined the compatibility of the method with DMSO, which is crucial because DMSO readily dissolves most organic compounds. Figure 2b shows that the Pd-catalyzed deallylation using TFP and $NaBH_4$ is most effective in the 0−10% DMSO range. We also found an optimal TFP concentration to be in the 400−500 μM range (Figure 2c). Higher concentrations of TFP were detrimental, presumably because the equilibrium shifted from PdL_{4-n} to PdL_4 (Figure 2d), decreasing the concentration of the reactive palladium species and thereby retarding the deallylation of **1**.

We next set out to further study the Pd-catalyzed deallylation reaction under the TFP/$NaBH_4$ conditions. As Figure 3a shows, this method can be used in the pH 5−10 range.[36,37] The initial rate under these conditions was 5 times faster than that under our previous conditions (Figure 3b).[19,37] The fluorescence intensity correlated to the concentration of palladium in the 1.0−50 nM (0.1−5.3 ppb) range after 1−4 h at 24 °C (Figure 3c). We observed a signal-to-background (S/B) ratio of 3 at 42 nM (4.5 ppb) after 1 h and at 24 nM (2.6 ppb) after 4 h, which is an 8-fold enhancement compared to our previous method.[19,37]

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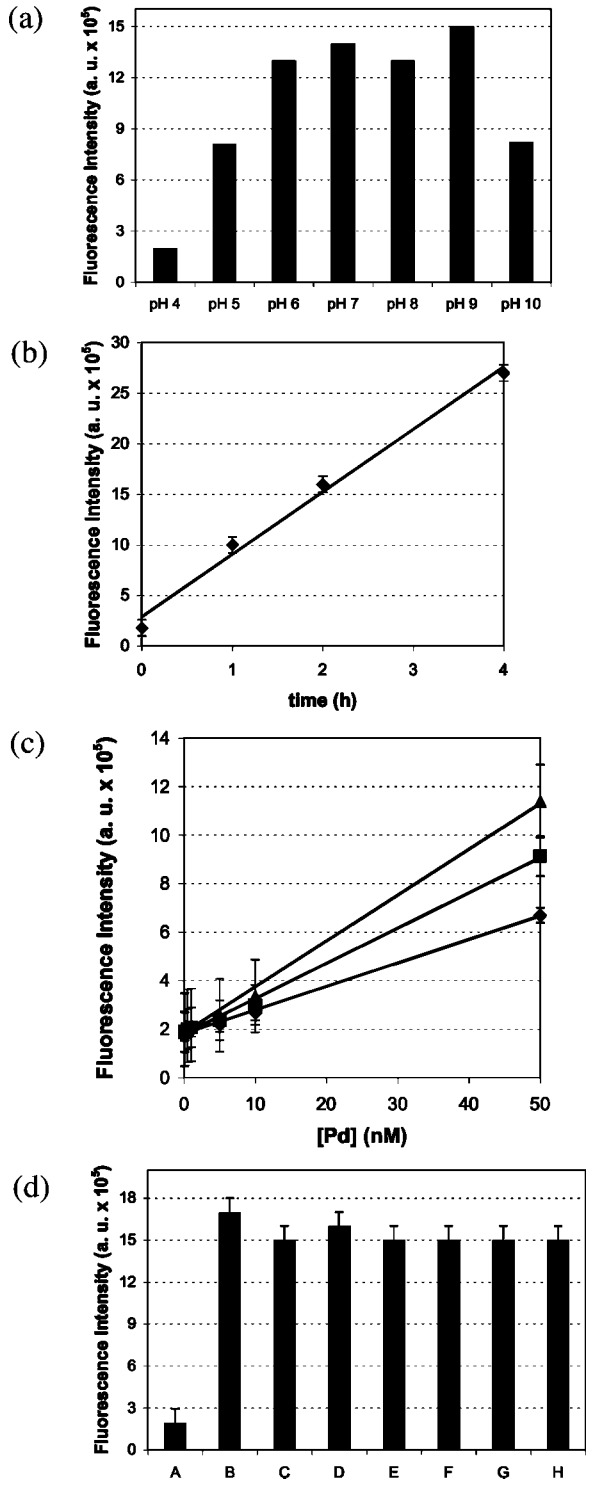

Figure 3. Catalytic Pd detection method with TFP/$NaBH_4$. [**1**] = 12.5 μM, [TFP] = 500 μM, and [$NaBH_4$] = 1.25 mM, and the assays were performed for 1 h at 24 °C in DMSO/pH 7 buffer (5:95) before fluorescence measurement (λ_{em} = 525 nm). (a) The pH-dependent deallylation of **1** in the presence of Pd (100 nM). (b) Initial rate analysis for deallylation of **1** in the presence of Pd (100 nM). $y = 6.18x + 2.88$; $R^2 = 0.991$. (c) Correlation between fluorescence intensity and [Pd]. ◆ = 1 h; $y = 0.0971x + 1.81$; $R^2 = 0.998$. ■ = 2 h; $y = 0.145x + 1.81$; $R^2 = 0.997$. ▲ = 4 h; $y = 0.189x + 1.86$; $R^2 = 0.996$. (d) Fluorescence induction by various Pd species and oxidation states (100 nM): A, no Pd; B, $PdCl_2$; C, $Pd(OAc)_2$; D, $Pd(acac)_2$; E, $Pd(PPh_3)_2Cl_2$; F, $Pd(MeCN)_2Cl_2$; G, $Pd_2(dba)_3$; H, Pd standard.

The TOF in this reaction was determined as 14.1 h^{-1}. This method was also found to be general for many palladium sources because they are efficiently reduced to Pd^0 by $NaBH_4$ and ligated with TFP regardless of the source (Figures 3d and 2d).

Palladium Detection in Functionalized Organic Compounds. Toward the goal of detecting palladium in the presence of functionalized organic compounds, ampicillin **22** (Figure 4c) was used as a model since it represents a highly functionalized drug. Here, a success can be measured by % recovery of signals as defined in Scheme 1. Because this method is reactivity-based, >100% recovery may be expected when a palladium-contaminated organic compound accelerates the deallylation of **1**. In order to minimize interference by the organic molecule, we tested different pretreatment methods (not shown) to disrupt putative ampicillin−palladium complexes. It was found that treatment of the Pd-spiked ampicillin in DMSO with 12 N HCl, followed by the deallylation of **1** in DMSO/pH 10 buffer containing TFP and $NaBH_4$ generated a fluorescence signal recovery of 76% (Scheme 1 and Figure 4a).[38]

We then applied this method for palladium detection in a variety of functionalized organic compounds as shown in Figure 4c−e. As Figure 4a,c shows, the procedure shown in Scheme 1 with Pd-spiked samples (Pd = 8.5 ppm in the compound) led to successful recovery of fluorescence signal for a variety of molecular structures. Importantly, we were able to recover the signal in the presence of chelating atoms found in many privileged structures and drug-like compounds, potentially reactive compounds (**8**, **9**, **19**), and other fluorophores (**20**). Compounds **5**, **11**, and **12** appeared to enhance the rate of deallylation, which may be linked to the ability of these compounds to behave as nucleophiles to aid in catalyst turnover.[39] Nonetheless, these semiquantitative recoveries in the presence of such a large excess of compound imply limited interferences from differences in the sample matrix.

One problem with this method, however, became apparent: because amines such as **13**, **14**, **15**, and **17** are largely unprotonated at pH 10, they may chelate to palladium more strongly under basic conditions. As such, DMSO solutions of Pd-spiked samples were diluted with pH 7 buffer to ensure overall pH 7 in the final solution containing TFP, $NaBH_4$, and **1**.[40] At pH 7, these compounds remain protonated, which should prevent palladium chelation. As Figure 4b,c shows, this method efficiently recovered fluorescence signal, even in the presence of these amines. Interestingly, under these conditions, alkyne **8** and bromobenzene **9** were found to accelerate the reaction. In the case of **8**,[41] the internal alkyne may serve as a weak ligand, thereby promoting formation of a coordinatively unsaturated palladium species. Compounds **6**, **10**, and **12** accelerated the deallylation of **1**, possibly due to their role as nucleophiles. While compounds containing carboxylate moieties (**19**, **20**, **22**) appear to be interfering, this was not found to be general.[42] Thus, through judicious choice from our two methods, palladium can be detected in functionalized small molecules.

(38) In ICP-MS, these requirements are achieved via sample preparation with concentrated nitric acid and in some cases microwave digestion. In fact, this step is necessary because ICP-MS is sensitive to the chemistry within the coordination sphere of the metal (see ref 13).
(39) Mackenzie, P. B.; Whelan, J.; Bosnich, B. *J. Am. Chem. Soc.* **1985**, *107*, 2046.
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(42) Acrylic acid does not interfere with the deallylation reaction.

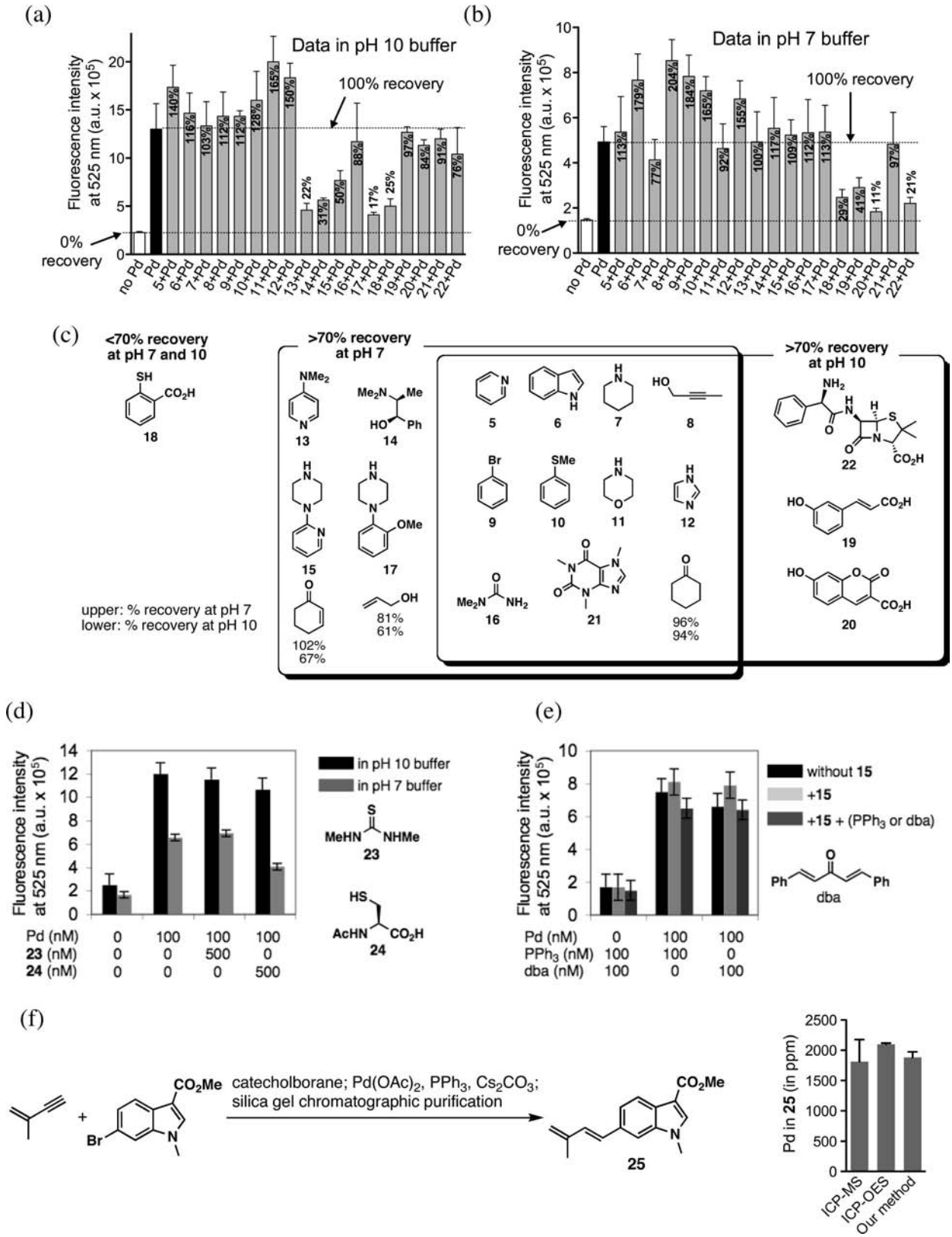
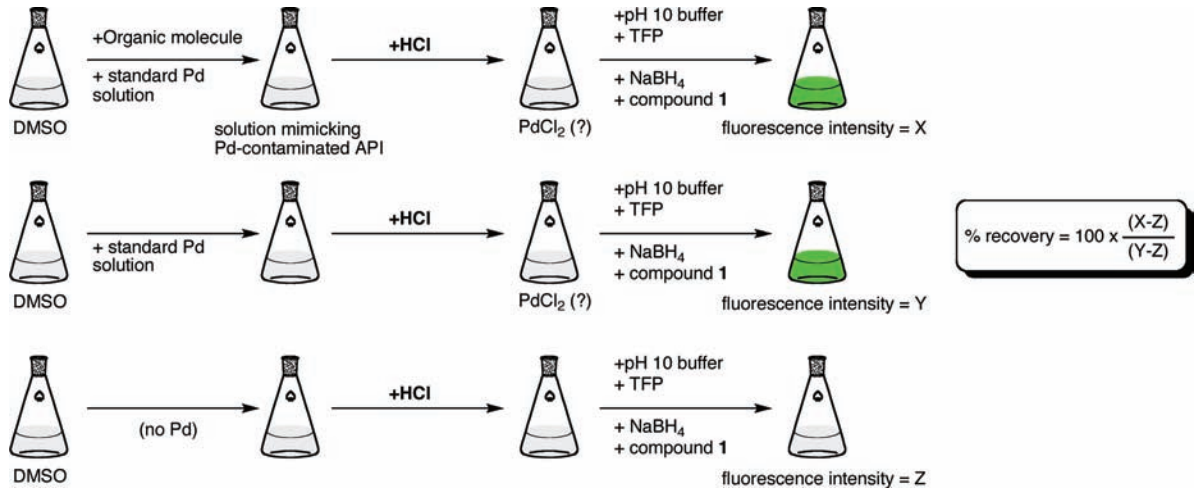


Figure 4. Pd detection in functionalized organic compound samples. [**1**] = 12.5 μM, [TFP] = 500 μM, and [$NaBH_4$] = 1.25 mM, and the assays were performed for 1 h at 24 °C before fluorescence measurement. (a) Detection in DMSO/pH 10 buffer (5:95) after pretreatment with concentrated HCl (60 mM) compared to a positive control (Pd). (b) Detection in DMSO/pH 7 buffer (150 mM) (5:95) after pretreatment with concentrated HCl (60 mM) compared to a positive control (Pd). (c) Structures of Pd-spiked compounds and summary of % recoveries. (d) Detection of Pd in the presence of **23** or **24**. (e) Detection of Pd in **15** in the presence of Ph_3P or dba. (f) Comparison of our method with other analytical methods. Compound **25** was prepared by a Pd-catalyzed reaction.

Another potential problem with the pH 10 method was observed with compounds containing α,β-unsaturated ketones (67% recovery of fluorescence signal, not shown). Because allyl alcohol interferes with deallylation (61% recovery of fluorescence signal; Figure 4c), we speculated that excess $NaBH_4$ had reduced such enone functionalities to the corresponding allylic

Scheme 1



alcohol and that the allylic alcohol interfered. A model study comparing cyclohexanone and cyclohexenone, simple examples of a saturated ketone and an α,β-unsaturated ketone, was performed in DMSO/pH 7 buffer in the presence and in the absence of $NaBH_4$ and confirmed our hypothesis (not shown).[43] Because allylic hydroxy groups are rarely present in drugs due to their susceptibility to biological oxidation, this may not be a severe limitation to our method. Figure 4c summarizes these results and the results shown in Figure 4a,b.

In addition to functionalized organic compounds, palladium must be detected in the presence of residual sulfur-based reagents,[7] phosphines, and organic ligands as they are used as scavengers or part of palladium catalysts. In the presence of *N*,*N*′-dimethylthiourea (**23**), *N*-acetylcysteine (**24**), Ph_3P, and dibenzylideneacetone (dba) in realistic quantities, palladium could be detected with good percent recovery (Figure 4d,e).[44] These results indicate that palladium quantification by our fluorescent method will not be skewed by the presence of these strong Pd ligands, presumably due to the use of $NaBH_4$.

Palladium Detection in Synthetic Samples. To further validate our method, we compared our fluorescent method to ICP-MS analysis. Specifically, we analyzed the indole derivative **25** that was prepared according to Figure 4f. For fair comparison, the pretreatment of the sample was analogous to that of typical ICP-MS analysis in this study. Comparison of the fluorescence signals and a standard curve under the modified conditions indicated that the palladium content in **25** was 1884 ± 169 ppm (mean $\pm$ standard deviation). Two independent ICP-MS analyses of this sample at two institutes revealed that the palladium content of this sample was 1813 ± 316 and 2097 ± 108 ppm, respectively. This double-blind cross-examination of palladium content in **25** supports the validity of our fluorescence-based quantification of palladium in synthetic compounds.

Palladium Detection in Used Resin Scavengers. Adsorption via resin-based thiol and amine scavengers is frequently employed to remove palladium.[7,11] Despite the high cost of these resins, it may not be cost-effective to recycle used resins because the metal analysis is currently time-consuming and expensive. To determine whether our palladium detection method can aid such recycling efforts, a sample of a thio-functionalized polysiloxane-based scavenger was first used for palladium removal.[45] Following scavenging, the resin was dried and treated with TFP, $NaBH_4$, and **1** in DMSO/pH 7 buffer. As Figure 5a shows, palladium could be visually detected within 15 min with a hand-held laser pen or long-range UV lamp. Thus, our palladium detection method may provide a useful means of determining whether resins contain palladium even in the presence of sulfur atoms. In addition, this method may be extended to facilitate studies on palladium materials, including colloid- and polymer-bound catalysts.[5]

Palladium Black Detection. During Pd-catalyzed cross-coupling reactions, a common catalyst deactivation pathway is the formation of palladium black. Because this process likely occurs to some extent during palladium-catalyzed reactions, palladium black could be a component of the palladium contamination found in the final products. As such, it is crucial that our method be able to detect even this insoluble form of palladium. A sample of palladium black (<2.25 μmol/mL) was treated with TFP, $NaBH_4$, and **1** in DMSO/pH 7 buffer. As Figure 5b shows, our fluorescent method can be used for insoluble palladium.

Palladium Detection in Spent Catalytic Converter Materials. X-ray fluorescence (XRF) is also a common method in palladium analysis, particularly in the catalytic converter industry.[46,47] In light of the current palladium and platinum shortages, recycling of catalytic converters is an important objective.[48] Although XRF analysis typically requires little sample preparation (only grinding in the case of catalytic converters), this method suffers from sample inhomogeneity and lacks sensitivity.[47] As such, we wanted to apply our fluorescent method for palladium detection in spent catalytic converters. Samples of Mg−Al silicate spiked with Pd, Pt, and Rh at various concentrations, which are mimics of catalytic converters, were treated with TFP, $NaBH_4$, and **1** for 30 s. To ensure uniformity across many samples that were analyzed at the same time, we

(43) Little reaction occurs in pH 10 buffer in the absence of $NaBH_4$; see Figure 3a for pH data.
(44) *N*-Acetylcysteine and 85 000 equiv of *N*,*N*′-dimethylthiourea were found to completely halt the Pd-catalyzed deallylation reaction.
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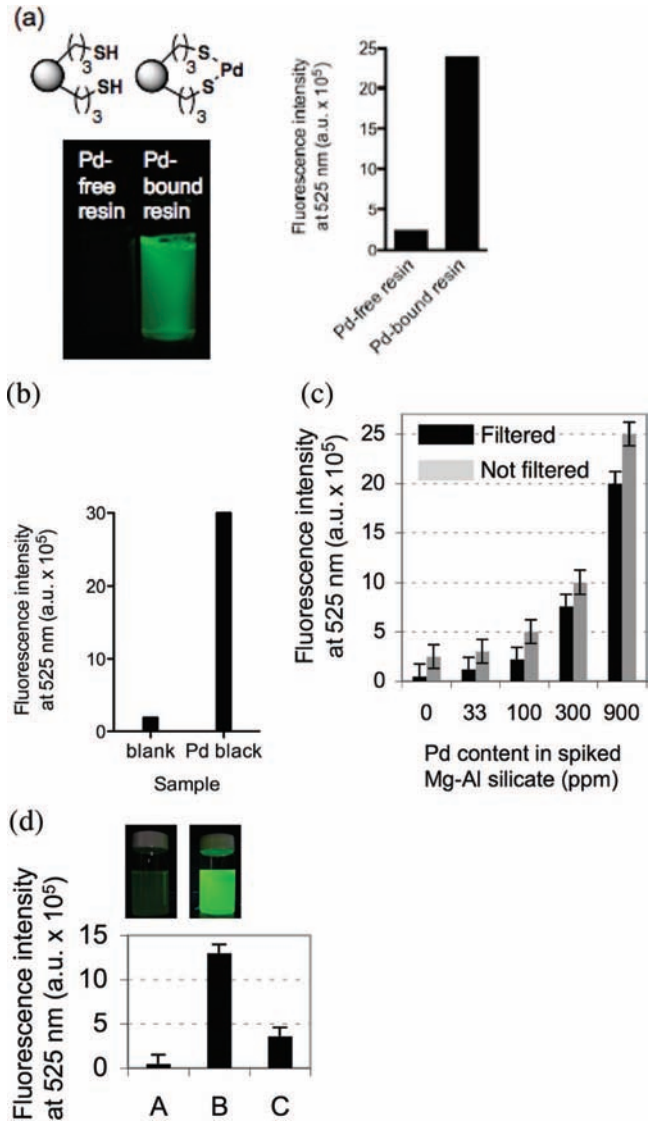

Figure 5. Pd detection in heterogeneous samples. [**1**] = 12.5 μM, [TFP] = 500 μM, and [$NaBH_4$] = 1.25 mM, and the assays were performed at 24 °C before fluorescence measurement. (a) Scavenger resin (Deloxan MP metal scavenger) and its Pd-bound form were treated with a solution of **1**, $NaBH_4$, and TFP in pH 7 buffer. (b) Detection in the presence of Pd black (<2.25 μmol/mL). (c) Detection in spiked Mg−Al silicate (Pd/Pt/Rh 2:2:1). Each reaction was quenched by the addition of *N*-acetylcysteine in large excess after 30 s and filtered before fluorescence measurement. Filtering of the samples, however, is not necessary for detection. (d) Detection in spent catalytic converter sample. A, Pd-free Mg−Al silicate; B, industry spent catalyst (alumina; [Pd] = 2345 ppm from ICP-MS); C, spent catalyst (cordierite; [Pd] = 1062 ppm, [Pt] = 827 ppm, [Rh] = 224 ppm from ICP-MS). Each reaction was quenched by the addition of *N*-acetylcysteine after 2 min and filtered before fluorescence measurement. Photos of samples A and B are shown above the graph.

used an excess of *N*-acetylcysteine to halt the metal-catalyzed deallylation.[44] As Figure 5c shows, this method can easily detect Pd/Pt down to 100 ppm, even in such heterogeneous mixtures.

We then applied this method for palladium detection in actual spent catalytic converter samples. Figure 5d shows that this method is capable of detecting palladium in these samples at concentrations of 1000−2500 ppm in various sample matrices (silicate, alumina, cordierite). Because these materials are often exposed to high temperatures, the metal particles tend to agglomerate and form palladium aggregates.[49] Thus, successful detection in these real catalytic converter samples indicates that aggregates may not present problems for this method. Currently recycle/no-recycle decisions are made at 900 ppm; therefore, our fluorescent method offers fast identification that does not require the preparation of samples and can give results using a hand-held fluorometer as the sole instrument.

Conclusion

We have demonstrated that the combination of compound **1**, TFP, and $NaBH_4$ represents a catalytically active fluorometric detection system for palladium. Importantly, this new method overcame problems associated with our previous palladium detection method, namely the compatibility with functionalized organic molecules and sensitivity. We envision that our method may be used in conjunction with ICP-MS for palladium detection in synthetic intermediates and final products. ICP-MS should be used to determine initial and final palladium contents. However, our method can be used to monitor relative palladium contents in a compound to prioritize scavenging methods in a high-throughput manner. We also demonstrated the utility of our method for detection of residual palladium remaining in sulfur-based resin scavengers. The streamlining of palladium analyses using our high-throughput method could greatly aid in the synthesis and analysis of drugs. It should be noted that this new method detected palladium residue in a functionalized organic compound at a pharmaceutical company that an ICP-MS analysis did not (not shown).

We also envision that our method may be used to detect palladium in recycling efforts. Due to the widespread usage of palladium (dentistry, electronics, fuel cells, catalytic converters, jewelry) and the current mining problems,[48] recycling of this precious metal is crucial for sustaining world supplies. One of the largest markets for palladium recycling is in the spent catalytic converter industry, and it is critical to this field to rapidly identify catalytic converters that contain viable amounts of palladium. Our method can readily identify Pd-rich catalytic converters in <2 min regardless of the sample matrix and without pretreatment, indicating potential on-site applications for making recycle/no-recycle decisions.

Experimental Section

General Information. Pd standard was purchased from High-Purity Standard (catalog no. 100038-1, lot no. 632601) and used as received. $PdCl_2$ and $Pd(PPh_3)_2Cl_2$ were purchased from Alfa Aesar and used as received. $Pd(PPh_3)_4$, $Pd(acac)_2$, and $Pd(MeCN)_2Cl_2$ were purchased from Strem and used as received. $Pd(OAc)_2$ was purchased from TCI and used as received. Tri-2-furylphosphine was purchased from Strem and used as received. Sodium borohydride was purchased from Acros and used as received. Buffers were purchased from J. T. Baker (pH 7, catalog no. 5608-01; pH 10, catalog no. 5609-01) and used as received. pH 7 buffer concentrate was purchased from Fisher Scientific (catalog no. SB109-500) and used as received. Pure water is Aristar ULTRA water. Deloxan MP metal scavenger (>2.0 mmol/g; effective capacity = 0.3−0.5 mequiv/mL) is a product of Strem sold in collaboration with Evonik (>2.0 mmol/g; effective capacity = 0.3−0.5 mequiv/mL). Pd black (surface area = 40−60 m^2/g) was purchased from Aldrich and used as received. Magnesium aluminum silicate was purchased from Spectrum and used as received. *N*-Acetyl-L-cysteine was purchased from TCI and used as received.

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Stock Solution Preparation and Notes. A 10.0 mM stock solution of **1** (42.7 mg; 0.10 mmol; solution A) was prepared in DMSO (10 mL). This solution was stored in the dark as a precautionary measure. A 100 mM stock solution of TFP (232.2 mg; 1.00 mmol; solution B) was prepared in DMSO (10 mL). This solution was freshly prepared every 2 weeks as a precautionary measure due to phosphine oxidation. A 2.5 M stock solution of $NaBH_4$ (943 mg; 25 mmol) was prepared in 10 M NaOH (10 mL). Further dilutions of this solution with pH 9 borate buffer were performed to prepare a 100 mM stock solution (solution C). Solutions of Pd standard were prepared by diluting a commercial Pd standard solution (1000 ± 3 μg/mL in 10% HNO_3) with 1% HNO_3. All solutions were stored at 24 °C.

Fluorescence Spectroscopy. Fluorescence spectra were recorded in a 1 × 1-cm disposable cuvette (VWR; catalog no. 58017-880) on a Jobin Yvon FluoroMax-3 spectrometer under the control of a Windows-based PC running FluorEssence software. The samples were excited at 497 nm, and the emission intensities were collected at 525 nm. All spectra were corrected for emission intensity using the manufacturer-supplied photomultiplier curves.

Phosphine Screening. Pd standard (40.0 μL of 10.0 μM stock, $[Pd]_{final}$ = 100 nM), phosphines (20.0 μL, $[phosphine]_{final}$ = 500 μM), and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM) were added to a mixture of DMSO/pH 7 buffer (5:95) (4.0 mL). After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was added, and the samples were incubated for 1 h at 24 °C before fluorescence measurement.

Palladium: Buffer Screening. Pd standard (40.0 μL of 10.0 μM stock, $[Pd]_{final}$ = 100 nM), solution B (20.0 μL, $[TFP]_{final}$ = 500 μM), and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM) were added to a mixture of DMSO/pH *X* buffer (5:95) (*X* = 4.0−10.0) (4.0 mL). After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was added, and the samples were incubated for 1 h at 24 °C before fluorescence measurement.

Palladium: TOF. To determine the TOF, a solution containing Pd standard ($[Pd]_{final}$ = 100 nM), solution B (20.0 μL, $[TFP]_{final}$ = 500 μM), solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM), and solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was prepared in DMSO/pH 7.0 buffer (5:95). The intensity of this sample was compared to a standard solution containing **2** ($[\mathbf{2}]_{final}$ = 100 nM) in DMSO/pH 7.0 buffer (5:95). The intensity of the Pd^{2+}-containing sample was 1.2×10^6 after 1 h at 24 °C, and the intensity of the standard solution of **2** was 8.2×10^4. The turnover frequency is therefore $(1.2 \times 10^6 / 8.2 \times 10^4)/1\ h = 14.1\ h^{-1}$.

Palladium: Concentration Dependence/Initial Rate. Varying amounts of Pd solution, solution B (20.0 μL, $[TFP]_{final}$ = 500 μM), and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM) were added to a mixture of DMSO/pH 7.0 buffer (5:95) (4.0 mL). After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was added, and the samples were incubated for 4 h at 24 °C before fluorescence measurement. The concentration dependence data over time were plotted to obtain the initial rate.

Dependence on Palladium Reagents. Pd solutions (40.0 μL of 10 μM stock, $[Pd]_{final}$ = 100 nM), solution B (20.0 μL, $[TFP]_{final}$ = 250 μM), and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM) were added to a mixture of DMSO/pH 7.0 buffer (5:95) (4.0 mL). After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was added, and the samples were incubated for 1 h at 24 °C before fluorescence measurement.

Palladium Detection in Compounds: pH 10 Method. DMSO (200 μL) and Pd standard solution (40 μL of 10 μM solution in 1% HNO_3; $[Pd]_{sample}$ = 8.5 ppm) were added to each of the compounds shown in Figure 4c (5.0 mg), and the resulting sample was allowed to sit on a bench for 30 min. Following this period, the sample was treated with 12 N HCl (20 μL, $[HCl]_{final}$ = 60 mM) and incubated at 24 °C for 10 min. The sample solution was then diluted with pH 10 buffer ($[K^+]$ = 114 mM; 3.8 mL) and treated with solution B (20.0 μL, $[TFP]_{final}$ = 250 μM) and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM). After shaking and 5 min incubation at 24 °C, solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was added, and the samples were incubated for 1 h at 24 °C before fluorescence measurement.

Palladium Detection in Compounds: pH 7 Method. DMSO (200 μL) and Pd standard solution (40 μL of 10 μM solution in 1% HNO_3; $[Pd]_{sample}$ = 8.5 ppm) were added to each of the compounds shown in Figure 4c (5.0 mg), and the resulting sample was allowed to sit on a bench for 30 min. Following this period, the sample was treated with 12 N HCl (20 μL, $[HCl]_{final}$ = 60 mM) and incubated at 24 °C for 10 min. The sample solution was then diluted with pH 7 buffer ($[PO_4^{3-}]$ = 150 mM; prepared by diluting pH 7 buffer concentrate ($[PO_4^{3-}]$ = 1.25 M) with pure water; 3.8 mL) and treated with solution B (20.0 μL, $[TFP]_{final}$ = 250 μM), and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM). After shaking and 5 min incubation at 24 °C, solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was added, and the samples were incubated for 1 h at 24 °C before fluorescence measurement.

Pd Detection with Other Ligands. 2-Pyridylpiperazine, Pd standard (40.0 μL of 10.0 μM stock, $[Pd]_{final}$ = 100 nM), and Ph_3P/dba (40.0 μL of 10.0 μM stock, $[Ph_3P/dba]_{final}$ = 100 nM) were added to a mixture of DMSO/pH 10 buffer (5:95) (4.0 mL). Solution B (20.0 μL, $[TFP]_{final}$ = 500 μM) and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM) were then added. After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was added, and the samples were incubated for 1 h at 24 °C before fluorescence measurement.

Aqua Regia. Aqua regia was prepared by mixing concentrated HNO_3 (ultrahigh purity with trace metals) and concentrated HCl (ultrahigh purity with trace metals) in a 1:3 ratio immediately prior to use.

Preparation of Compound 25. Catecholborane (1.70 g, 14.3 mmol) and 2-methylbut-1-en-3-yne (2.36 g, 36.0 mmol) were added to a 25-mL sealed tube at 24 °C that was purged with nitrogen, and the resulting mixture was stirred at 80 °C for 2 h. In a 50-mL round-bottom flask at 24 °C that was purged with nitrogen, $Pd(OAc)_2$ (47.0 mg, 0.05 mmol), PPh_3 (110 mg, 0.10 mmol), and THF (40 mL) were stirred at 24 °C for 30 min and then transferred via syringe to a 250-mL round-bottom flask that was purged with nitrogen. The mixture of catecholborane and 2-methylbut-1-en-3-yne, MeOH (15 mL), THF (38 mL), methyl 6-bromo-1-methyl-1*H*-indole-3-carboxylate (1.10 g), and Cs_2CO_3 (4.00 g, 12.3 mmol) was then added to the 250-mL round-bottom flask at 24 °C, and the resulting reaction mixture was stirred at 70 °C for 12 h. The crude mixture was then filtered through a plug of silica gel (70 mL), rinsed with EtOAc (100 mL), and concentrated *in vacuo*. The crude residue was purified by flash chromatography (10 to 40% EtOAc in hexanes) on silica gel (100 mL) to afford **25** (1.0 g).

Detection of Pd in Compound 25. Varying amounts of Pd standard solution were added to 1-dram vials and treated with aqua regia (70 μL). The resulting mixtures were capped, mixed by vortex for ∼1 s, and incubated at 24 °C for 5 min. The sample solutions were then diluted with pH 7 buffer concentrate (2.5 mL, $[PO_4^{3-}]$ = 1.25 M) and treated with TFP (200 μL, 1.2 mM in DMSO, $[TFP]_{final}$ = 80 μM), $NaBH_4$ (100 μL, 30 mM in 0.12 N NaOH, freshly prepared, $[NaBH_4]_{final}$ = 1.0 mM) and **1** (100 μL, 375 μM in DMSO, $[\mathbf{1}]_{final}$ = 12.5 μM). The samples were vortexed for 2 s and incubated for 1 h at 24 °C before fluorescence measurement. Synthetic compound **25** (30 μL, 100 μg/mL in DMSO) was analyzed using the above protocol in triplicate.

Pd Detection in Used Resin Scavengers. A mixture of DMSO/pH 7 buffer (5:95) (4.0 mL) was added to the used Deloxan MP metal scavenger (20 mg). Solution B (20.0 μL, $[TFP]_{final}$ = 500 μM) and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM) were then added. After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final}$ = 12.5 μM) was added, and the samples were incubated for 20 min at 24 °C before fluorescence measurement. Visible detection with a laser pen was noticed after 15 min. A negative control was performed using unused Deloxan MP metal scavenger.

Pd Black Detection. Pd black ($[Pd\ black]_{final}$ < 2.25 μmol/mL), solution B (20.0 μL, $[TFP]_{final}$ = 250 μM), and solution C (50.0 μL, $[NaBH_4]_{final}$ = 1.25 mM) were added to a mixture of DMSO/

pH 7.0 buffer (5:95) (4.0 mL). After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final} = 12.5$ μM) was added, and the samples were incubated for 1 h at 24 °C before fluorescence measurement.

Pd/Pt Detection in Spiked Mg−Al Silicate. A solution of Pd, Pt, and Rh (2:2:1) was prepared by dissolving PtO_2 (10 mg) and $Rh(Ph_3P)_3Cl$ (5 mg) in a commercially available Pd standard solution in 10% HNO_3 (20 mL). Varying amounts of Pd/Pt/Rh solution and water (50 mL) were added to Mg−Al silicate (10 g), and the resulting samples were heated to dryness on a hotplate.

pH 7 buffer (5 mL) was added to a 100-mg portion of Pd/Pt/Rh-treated Mg−Al silicate. The slurry was then treated with solution B (20.0 μL, $[TFP]_{final} = 400$ μM) and solution C (50.0 μL, $[NaBH_4]_{final} = 1.0$ mM). After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final} = 10$ μM) was added. After 30 s at 24 °C, *N*-acetylcysteine was added (∼5 mg) to halt the reaction, and the solution was filtered through a syringe filter before fluorescence measurement.

Pd Detection in Spent Catalytic Converter Samples. pH 7 buffer (5 mL) was added to a 100-mg portion of catalytic converter sample in a 10-mL plastic syringe that was capped at the tip. The slurry was then treated with solution B (20.0 μL, $[TFP]_{final} = 400$ μM) and solution C (50.0 μL, $[NaBH_4]_{final} = 1.0$ mM). After shaking, solution A (5.0 μL, $[\mathbf{1}]_{final} = 10$ μM) was added. After 2 min at 24 °C, the cap was removed, and the solution was filtered through a syringe filter into a disposable cuvette containing *N*-acetylcysteine (∼5 mg) before fluorescence measurement.

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