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# An amphiphilic pentaaryl cyclopentadienyl Ru(II) molecular rotor with a luminescent tag

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**Abstract:** This study presents a chemically modified molecular motor designed for interactions with lipid bilayers. By incorporating long alkyl chains into the anchoring fragment and a rhodamine luminescent tag onto the rotating subunit, we aim to monitor the position of the motor and allow for its real-time visualization during membrane perforation experiments. The synthesis of the functionalized tripodal ligand involved a seven-step process, culminating in the formation of a piano-stool Ru(II) complex that exhibits distinctive spectroscopic properties. The luminescence of the rhodamine tag, while slightly quenched upon incorporation in the Ru(II) complex, remains sufficiently strong to enable the single-molecule observation using a total internal reflection fluorescence (TIRF) microscope

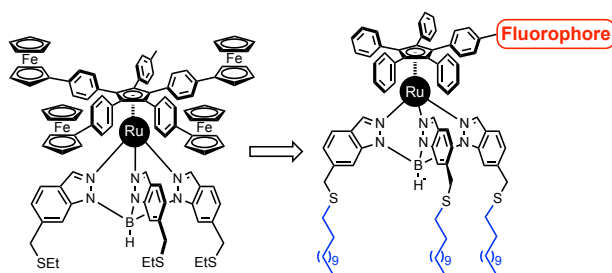
**Keywords** Molecular rotors, ruthenium, functionalization, fluorescent tag, amphiphile

Rotary molecular motors are fascinating nanoscale machines capable of converting chemical energy into rotational motion, their mechanism is often found often in nature and has been increasingly replicated in synthetic systems. Biological examples, such as ATP synthase<sup>2</sup> and bacterial flagellar motors,<sup>3</sup> demonstrate the efficiency of these systems in sustaining the processes essential for life, from ATP production to cellular motility. ATP synthase, for example, operates by rotating its components to synthesize the primary energy currency of the cell ATP, from ADP and inorganic phosphate, while bacterial flagella enable motility by a propeller like rotation to move cells.

Synthetic rotary motors, inspired by these biological examples, have been prepared in the last two decades<sup>4</sup> often with the goal of exploring the potential of such machines to achieve precise, directional movement, selectively targeting cells to induce biological processes. Rotary molecular motors offer an innovative approach to this challenge. For instance, it is now possible to exploit the molecular-mechanical action of a motor to alter or perforate a lipid bilayer;<sup>5</sup> either by physical rotation through membranes or by creating localized mechanical stresses that disrupt their structure, demonstrating their potential to deliver therapeutic agents across membranes or to kill cells by drilling holes in the membrane.<sup>6</sup> These results have radically transformed the idea of what can be classified as an active drug, with the emergence of a new type named mechanically-active drugs.

The use of molecular motors in nanomedicine is still in its infancy but they are uniquely suited for this task because of their precision, directionality, and energy efficiency in performing mechanical work on the nanoscale. However, one major challenge in applying rotary molecular motors to membrane perforation<sup>7</sup> is the ability to monitor and image their position and behavior once anchored to the membrane. For this reason, the incorporation of luminescent tags has become a vital strategy. By tagging the motors with fluorescent markers, it would be possible using advanced imaging techniques such as fluorescence microscopy to visualize and track their position and activity in real-time, ensuring that they target the correct cells and efficiently perforate the membrane.

We here present the chemical modification of a molecular motor we developed a few years ago (Figure 1) which is able to rotate unidirectionally on a gold surface.<sup>8</sup>



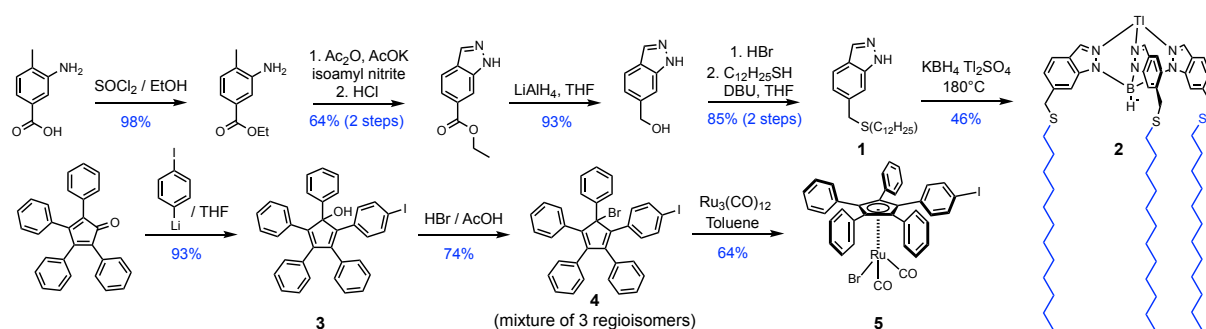
**Figure 1** Chemical modification of the molecular motor by addition of long alkyl chains on the anchoring ligand (in blue) and a fluorophore (in red).

Depending on which part of the rotor subunit is excited (tolyl or ferrocenyl subunits), the directionality of the rotation can be controlled, but thus far the molecule has been observed working only on metallic surfaces at low temperature. To move from the control of molecular motion to biological applications, studies in physiological media such as biomembranes, and the ability to operate at room temperature, are required. To achieve this, an updated molecular design derived from our already synthesized motor<sup>8</sup> was proposed with two key differences. First, it is required that the anchoring part (tripodal ligand) is functionalized with long alkyl chains (here a  $^{n}\text{C}_{12}\text{H}_{25}$  in blue) which are known to incorporate in the lipid bilayer. Second, a fluorescent tag (in red) is required on the rotating subunit to allow for direct visualization of these motors anchored on the membranes by a fluorescence microscope. To reach these goals, we selected a rhodamine B derivative<sup>9</sup> as a fluorescent tag which is well known for single molecule fluorescence. These chemical modifications will allow us to explore the molecules use in the expression of biological actions through both its interactions and the resulting structural change of the membranes.

The synthesis of the long chain functionalized tripodal ligand has been achieved by an adaptation of our previous strategy.<sup>10</sup> The preparation of the tripodal ligand **2** involves 7 steps starting from the commercially available 3-amino-4-methylbenzoic acid. Conversion to its ethyl ester derivative in the presence of excess thionyl chloride in refluxing ethanol gives the corresponding ethyl ester in an almost quantitative yield this is then engaged in a modified one-pot Jacobson procedure, using isoamyl nitrite as the nitrosating agent<sup>11</sup> to transform the *o*-toluidine derivative into the corresponding indazole. After acid hydrolysis and subsequent neutralization, the 6-(ethoxycarbonyl)indazole is obtained in 64% yield over two steps. Reduction of the ester using excess lithium aluminum hydride in tetrahydrofuran at 0 °C, affords the desired alcohol in 93% yield. After activation of the benzyl alcohol as a bromide derivative, subsequent conversion to the corresponding

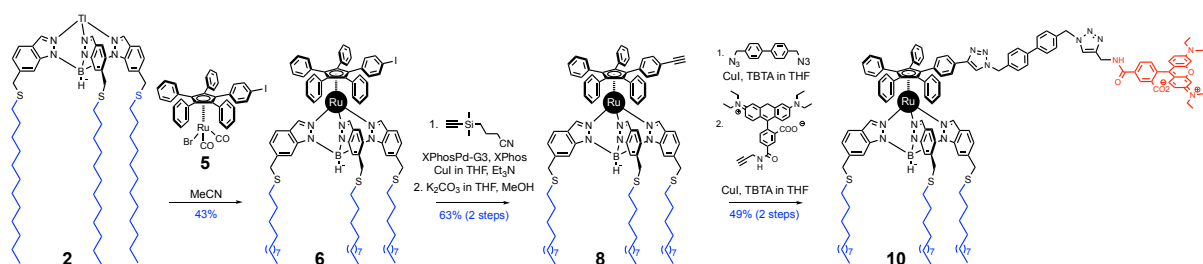
thioether **1** can be achieved. Then thallium(I) sulfate and 3.5 equivalents of 6-(dodecylsulfanyl)methylindazole **1** are heated without solvent to 180 °C until dihydrogen gas evolution ceased to give the desired thallium hydrotris(indazolyl)borate **2** in 46% yield after the unreacted indazole is removed by sublimation under reduced pressure.

As rotative subunit, the ligand **4**, a desymmetrized (1-(*p*-iodophenyl)-2,3,4,5-tetraphenyl)cyclopentadienyl bearing a bromine center to coordinate the ruthenium and a iodine to further incorporate a luminescent tag,<sup>12</sup> is synthesized in two steps from tetraphenylcyclopentadienone (Scheme 1). Ligand **4** is obtained as a mixture of three regioisomers due to the S<sub>N</sub>1 mechanism involved,<sup>13</sup> with the bromine present at different positions of the cyclopentadiene fragment. The presence of this mixture of regioisomers is not a problem since the next step, which consists of coordination to a ruthenium(II) center, leads to the same compound (**5**) for all three regioisomers. Due to the long alkyl chains, the dodecyl functionalized tripodal ligand **2** is not crystalline, however we can successfully coordinate it by reaction with the bromidodicarbonylruthenium(II) complex **5** to yield the piano-stool complex **6** in a 43% yield on gram scale.



**Scheme 1** Chemical synthesis of the modified tripodal ligand **2** and the precursor ruthenium(II) complex with one iodophenyl subunit to further incorporate a luminescent tag.

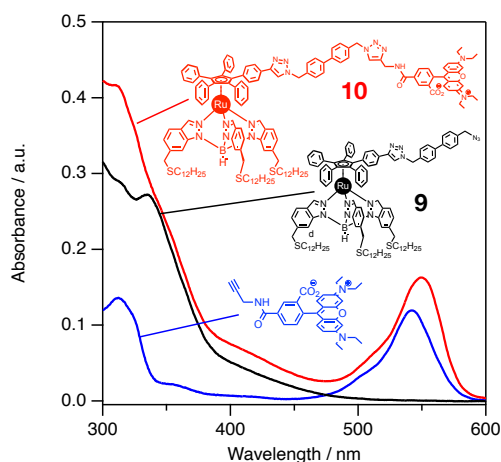
Taking advantage of the reactivity of the iodine center, we post-functionalize this ruthenium(II) complex with a derivative of rhodamine B. First, we introduce an alkyne function on the pentaphenylcyclopentadiene unit to obtain **8** in two steps. The use of a (3-cyanopropyl)dimethylsilyl protecting group, a polar analogue of trimethylsilyl,<sup>14</sup> allows for easy separation of complex **7** from the unreacted iodide derivative **6**. The alkyne-protected ruthenium(II) complex **8** is then subsequently reacted in two successive click reactions (Scheme 2). The 4,4'-bis(azidomethyl)-1,1'-biphenyl spacer is reacted first with **8** and then in a second click reaction with an alkyne-substituted rhodamine B fragment<sup>15</sup> to yield the desired amphiphilic motor **10** in four steps with an overall yield of 31% from the ruthenium(II) complex **6**. It must be noted contrary to the original molecular motor represented in Figure 1, this molecule **10** has no ferrocene. This is due to our recent finding that ferrocenes are not necessary to induce unidirectional rotation, only a desymmetrized cyclopentadienyl rotative ligand is needed.<sup>16</sup>



**Scheme 2** Chemical synthesis of the targeted rhodamine functionalized amphiphilic ruthenium(II) molecular motor.

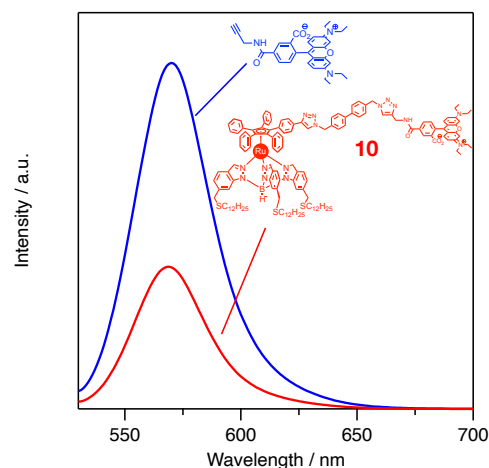
This new functionalized amphiphilic molecular motor with a rhodamine unit has been fully characterized by  $^1\text{H}$  and  $^{13}\text{C}$  NMR spectroscopy and HR-MS. We also measured the photophysical properties of **10** as well as the alkyne-substituted rhodamine B and the ruthenium complex without rhodamine **9**, as they constitute interesting model fragments. This piano-stool ruthenium(II) complex is already known to exhibit peculiar absorption features compared to its related ligands.<sup>17</sup> The analogous pentabiphenylcyclopentadiene ruthenium(II) complex with three ethylthioether groups shows a weak emission with a quantum yield of 0.27%, attributed to ruthenium-based charge transfer transitions involving the  $\pi^*$  orbitals of the phenyl substituents of the cyclopentadienyl ligand.

As shown in Figure 2, the absorption spectrum of the ruthenium(II) complex bearing a fluorescence tag corresponds to the sum of the two spectra of the separated subunits: the ruthenium(II) amphiphilic motor and the rhodamine fragment, illustrating the absence of any electronic effects.



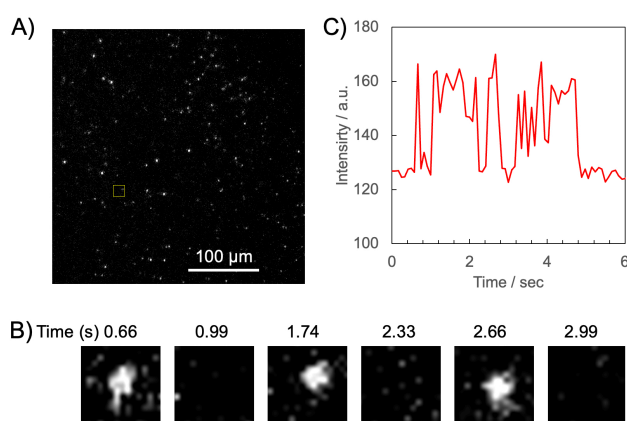
**Figure 2** UV-visible spectrum of **10** (red curve), model compounds **9** (black curve) and alkyne-substituted rhodamine B (blue curve) (conc. 10  $\mu\text{M}$  in chloroform).

The emission spectra of **10** was also measured. Despite some quenching of the luminescence of the rhodamine B fragment *via* a partial energy transfer from the rhodamine to the ruthenium fragment, the rhodamine fragment (Figure 3 in blue) is still fluorescent with an efficiency reduction of 41% once connected to the ruthenium motor (Figure 3 in red). This luminescence is reduced but still strong enough to allow for localization of the motor on the lipidic bilayer and facilitate following of the rotation and its effects.



**Figure 3** Emission spectrum of **10** (red curve) and model rhodamine compound (blue curve) ( $\lambda_{\text{exc}} = 520$  nm, conc.  $10 \mu\text{M}$  in chloroform).

To demonstrate the effectiveness of our molecular design for the fluorophore-modified molecular motor, we attempted the detection of single molecule fluorescence, which is known to be a powerful technique to evaluate the dynamics of membrane-associating molecules like membrane proteins.<sup>18</sup> TIRF microscopy was employed to examine the single-molecule fluorescence due to its high sensitivity and surface-selective detection of fluorescent signals.  $20 \mu\text{L}$  of a highly dilute chloroform solution ( $7.42 \times 10^{-12}$  M) of molecular motor **10** was deposited onto a coverslip to prepare the specimen. TIRF images show significant numbers of bright spots originating from the rhodamine emission attached to the molecular motor (Figure 4, Panel A). Time-lapse imaging reveals repeated blinking behavior, in some bright spots, suggesting single-molecule fluorescence (Figure 4, Panel B). The time-course fluorescence intensity of these spots displays a stepwise transition between dark and bright states (Figure 4, Panel C), further confirming that the fluorescence originates from individual molecular motors.



**Figure 4** TIRF microscopic image of **10** deposited on a glass surface (A), magnified fluorescence images of the blinking spot corresponding to the yellow square in panel A (B), and the time-course of the fluorescence intensity of the blinking spot (C).

We have synthesized and characterized<sup>19</sup> a molecular motor that has been chemically modified with long alkyl chains on its anchoring moiety for incorporation into lipid bilayers, and a rhodamine luminescent tag on its rotative part, enabling direct visualization during membrane perforation experiments. This amphiphilic motor functionalized with a luminescent tag presents suitable emission properties to allow

monitoring once incorporated in biomembranes. TIRF microscopic observation revealed that this rhodamine-tagged molecular motor can be applied to single molecule fluorescence observation. Work is now underway and we anticipate these chemical modifications to allow exploration into the use of fluorophore-labeled ruthenium(II) motor in the expression of biological actions through its interactions and resulting structural change of membranes.

This dual functionality, mechanical motion and luminescence, could greatly enhance the potential of such rotary motors for applications in nanomedicine, particularly in diagnostics, drug delivery, and cell-targeted therapeutics. While the field is still in its early stages, this combination of molecular motors with membrane perforation techniques has the potential to revolutionize this area. Our continued research will focus on enhancing the specificity and efficiency of these systems, optimizing their ability to interact with different types of membranes, and expanding their potential applications in medicine and biotechnology.

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- (19) **Experimental procedures and characterization for 10:** For other compounds, refer to the supporting information file.

A mixture composed of the compound **9** (295 mg, 160  $\mu\text{mol}$ ), alkyne-substituted rhodamine derivative<sup>[4]</sup> (102 mg, 195  $\mu\text{mol}$ ), CuI (19 mg, 100  $\mu\text{mol}$ ) and TBTA (44 mg, 82  $\mu\text{mol}$ ) were suspended in anhydrous tetrahydrofuran (17 mL). The resulting mixture was refluxed for 18 h. After removal of volatiles, the crude compound was purified by silica gel column chromatography (chloroform : methanol = 10 : 1) to give **10** as a brown solid with a yield of 71% (270 mg).

**<sup>1</sup>H NMR** (400 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 8.38 (br, 1H, NH), 8.11 (d,  $J$  = 0.8 Hz, 1H, H<sub>d</sub>), 7.93 (s, 3H, H<sub>d</sub>), 7.89 (s, 3H, H<sub>a</sub>), 7.63 (br, 1H), 7.56-7.53 (m, 5H, H<sub>Ar</sub>), 7.44-7.42 (m, 2H, H<sub>Ar</sub>), 7.38-7.31 (m, 14H, H<sub>Ar</sub>), 7.28-7.26 (m, 12H, H<sub>Ar</sub> with CHCl<sub>3</sub>), 7.11-7.06 (m, 4H, H<sub>Ar</sub>), 7.00-6.96 (m, 12H, H<sub>Ar</sub>), 6.55 (d,  $J$  = 9.2 Hz, 2H, H<sub>xanthene</sub>), 6.44 (d,  $J$  = 2.0 Hz, 2H, H<sub>xanthene</sub>), 6.33 (d,  $J$  = 8.0 Hz, 2H, H<sub>xanthene</sub>), 5.55 (s, 4H, H<sub>a-b</sub>), 4.75 (s, 2H, H<sub>e</sub>), 3.89 (s, 6H, H<sub>e</sub>), 3.35 (q,  $J$  = 6.9 Hz, 8H, NEt (CH<sub>2</sub>)), 2.49 (t,  $J$  = 7.2 Hz, 6H, H<sub>f</sub>), 1.66-1.58 (m, 14H, alkyl chains including H<sub>2</sub>O), 1.39-1.25 (m, 54H, CH<sub>2</sub>Alkyl), 1.16 (t,  $J$  = 7.2 Hz, 12H, NEt (CH<sub>3</sub>)), 0.87 (t,  $J$  = 6.8 Hz, 9H, CH<sub>3</sub>).

**<sup>13</sup>C NMR** (150 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 166.2, 153.8, 148.0, 143.6, 140.7, 140.3, 136.8, 134.2, 133.9, 133.9, 133.8, 133.6, 129.2, 128.9, 128.7, 128.5, 127.8, 127.2, 127.1, 124.4, 122.1, 121.9, 120.0, 119.5, 111.3, 108.6, 97.4, 88.2, 88.0, 87.2, 53.8, 44.6, 37.3, 31.9, 29.7, 29.7, 29.6, 29.4, 29.2, 29.0, 22.7, 14.1, 12.5.

**HR-MS** (ESI, positive):  $m/z$  calculated for C<sub>143</sub>H<sub>165</sub>BN<sub>15</sub>O<sub>4</sub>RuS<sub>3</sub> [M+H]<sup>+</sup> 2365.1468; found: 2365.1184.

**UV-Vis** (conc. 10  $\mu\text{M}$  in chloroform)  $\lambda_{\text{max}}$  ( $\epsilon$ ): 550 nm (16270 mol<sup>-1</sup>dm<sup>3</sup>cm<sup>-1</sup>).

**Emission** ( $\lambda_{\text{exc}}$  = 520 nm, conc. 10  $\mu\text{M}$  in chloroform)  $\lambda_{\text{max}}$ : 569 nm

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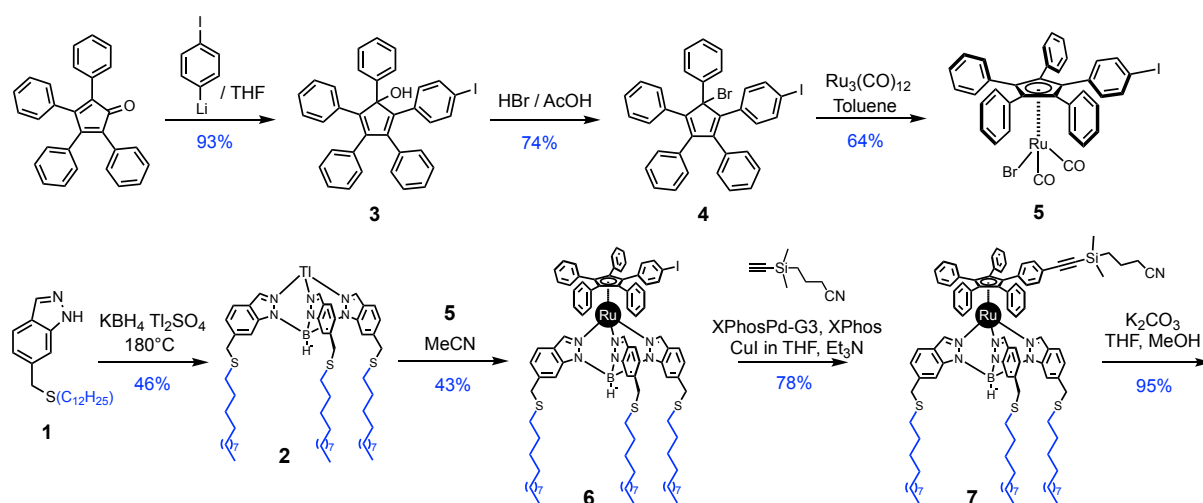
### 1. General Information: Materials and methods

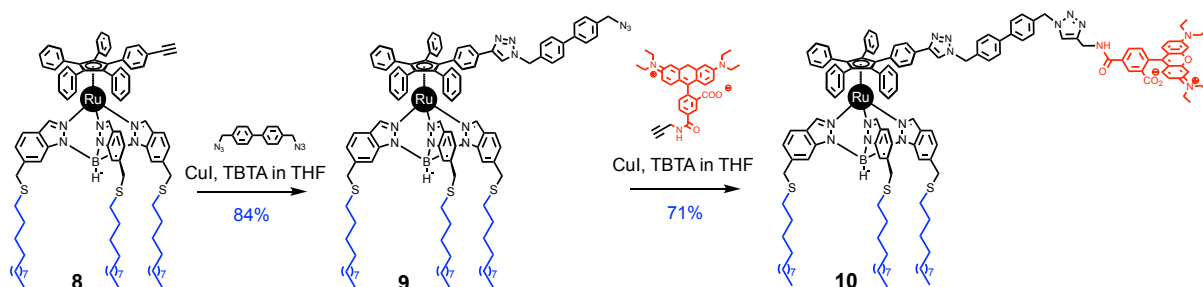
Synthetic procedures were carried out under a dry nitrogen atmosphere, unless otherwise specified. All reagents and solvents were purchased at the highest commercial quality available and used without further purification, unless otherwise stated. Thionyl chloride, acetic



anhydride, potassium acetate, hydrogen chloride, 1,8-diazabicyclo[5.4.0]-7-undecene (DBU), thallium(I) sulfate, copper(I) iodide (CuI), potassium carbonate (K<sub>2</sub>CO<sub>3</sub>), ammonium chloride (NH<sub>4</sub>Cl), sodium sulfate (Na<sub>2</sub>SO<sub>4</sub>), acetic acid (AcOH), triethylamine (NEt<sub>3</sub>), sodium azide, 4-methylmorpholine, tetrahydrofuran (THF), ethylacetate (AcOEt), methanol (MeOH), dichloromethane (CH<sub>2</sub>Cl<sub>2</sub>), chloroform (CHCl<sub>3</sub>), ethanol (EtOH), anhydrous THF, anhydrous N,N-dimethylformamide (DMF), anhydrous toluene, and anhydrous acetonitrile (MeCN) were purchased from FUJIFILM Wako Pure Chemical Corporation. 3-Amino-4-methylbenzoic acid, isoamyl nitrite, lithium aluminum hydride (LiAlH<sub>4</sub>), hydrogen bromide (30% in acetic acid), potassium borohydride, 4,4'-bis(bromomethyl)biphenyl, tris[(1-benzyl-1H-1,2,3-triazol-4-yl)methyl]-amine, propargylamine, HATU, HOAt, 1,4-iodobenzene, tetraphenylcyclopentadienone, *n*-butyllithium (nBuLi, 1.6 M in hexane), triruthenium dodecacarbonyl, trimellitic anhydride, and *N,N*-diethyl-3-aminopheanol were purchased from TCI. 1-Dodecanethiol, XPhosPd-G3, and XPhos were purchased from Sigma-Aldrich. [(3-Cyanopropyl)dimethylsilyl]acetylene,<sup>[1]</sup> 4,4'-bis(azidomethyl)biphenyl,<sup>[2]</sup> 5-carboxytetraethyl-rhodamine,<sup>[3]</sup> 2-[6-(diethylamino)-3-(diethyliminio)-3*H*-xanthen-9-yl]-5-(prop-2-yn-1-ylcarbonyl)benzoate,<sup>[4]</sup> and 6-(bromomethyl)-1*H*-indazole<sup>[5]</sup> were prepared according to literature procedure. The complexation reaction under microwave irradiation was carried out utilizing a CEM Discover SP in a glass pressure tube equipped with PTFE-coated magnetic stirring bar. <sup>13</sup>C spectra were recorded on a JEOL JNM-ECA600 spectrometer and <sup>1</sup>H spectra on a JEOL JNM-ECX400P spectrometer at a constant temperature of 298 K. Tetramethylsilane (TMS) was used as an internal reference for <sup>1</sup>H and <sup>13</sup>C NMR measurements in CDCl<sub>3</sub>. Chemical shifts ( $\delta$ ) are reported in ppm. Coupling constants (*J*) are given in Hz and the following abbreviations have been used to describe the signals: singlet (s); broad singlet (br. s); doublet (d); triplet (t); quadruplet (q); quintuplet (quint); multiplet (m). Full assignments of <sup>1</sup>H and <sup>13</sup>C NMR spectra were made with the assistance of COSY, HMBC, and HSQC spectra when necessary. The mass spectrometry was performed using JEOL JMS-700 MStation for EI and JEOL JMS-T100LP AccuTOF for ESI. The absorption and emission spectra were obtained using Hitachi U-3310 spectrometer and Hitachi F-7000 spectrometer in CHCl<sub>3</sub> solution at 20 ± 0.1 °C in 1.0 cm quartz cell. Silica gel column chromatography and thin-layer (TLC) chromatography were performed using PSQ60B (Fuji Silysia Chemical LTD.) and Merck silica gel 60 (F254) TLC plates, respectively.

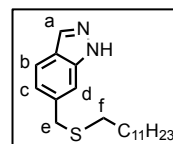
## 2. Synthetic procedures





### a. Synthesis of 1

1-Dodecanethiol (17.9 mL, 75 mmol) and DBU (11.2 mL, 75 mmol) were dissolved in anhydrous THF (100 mL). 6-(Bromomethyl)-1H-indazole<sup>[5]</sup> (10.6 g, 50 mmol) was added to the solution and the resulting mixture was refluxed for 1 h. After cooling the reaction mixture to room temperature, H<sub>2</sub>O (200 mL) was added to the mixture. The aqueous phase was extracted with CH<sub>2</sub>Cl<sub>2</sub> (200 mL x 4). The combined organic extracts were washed with brine (200 mL), dried over anhydrous sodium sulfate, filtered and evaporated under vacuum to obtain the crude compound which was purified by silica gel column chromatography (hexane : AcOEt = 4 : 1) to afford **1** as a colorless solid (14.5 g, 87%).



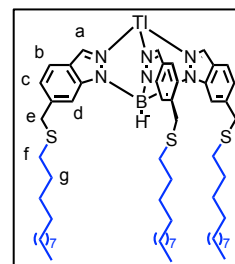
**<sup>1</sup>H NMR** (400 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 10.05 (s, 1H, NH), 8.05 (d,  $J$  = 0.8 Hz, 1H, H<sub>a</sub>), 7.70 (dd,  $J$  = 8.0 Hz, 1.1 Hz, 1H, H<sub>b</sub>), 7.42 (s, 1H, H<sub>d</sub>), 7.17 (dd,  $J$  = 8.2, 1.4 Hz, 1H, H<sub>c</sub>), 3.84 (s, 2H, H<sub>e</sub>), 2.42 (t,  $J$  = 7.4 Hz, 2H, H<sub>f</sub>), 1.60-1.52 (m, 3H, alkyl chain including H<sub>2</sub>O), 1.35-1.24 (m, 18H, alkyl chain), 0.88 (t,  $J$  = 7.0 Hz, 3H, CH<sub>3</sub>)

**<sup>13</sup>C NMR** (150 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 140.2, 138.7, 134.6, 123.1, 122.1, 121.2, 109.4, 36.7, 31.9, 31.5, 29.7, 29.6, 29.6, 29.5, 29.4, 29.2, 29.2, 28.9, 22.7, 14.1.

**HR-MS** (EI, positive):  $m/z$  calc. for C<sub>20</sub>H<sub>32</sub>N<sub>2</sub>S [M]<sup>+</sup>: 332.2286; found: 332.2281.

### b. Synthesis of 2

**1** (3.0 g, 9.0 mmol), KBH<sub>4</sub> (195 mg, 3.6 mmol) and Ti<sub>2</sub>SO<sub>4</sub> (910 mg, 1.8 mmol) were added to a Schlenk tube equipped with PTFE stopcock. The mixture was heated at 140 °C for 1 h under a N<sub>2</sub> flow, then the stopcock was closed and the reaction temperature was raised to 180 °C. After 5 h heating, the reaction mixture was cooled to room temperature. The reaction mixture was extracted with CH<sub>2</sub>Cl<sub>2</sub> (100 mL) and insolubilities were removed by centrifugation. The obtained supernatant was evaporated under vacuum to afford the crude compound which was redissolved in a minimum quantity of CH<sub>2</sub>Cl<sub>2</sub> and purified by reprecipitation by adding the same volume of MeOH to afford **2** as a colorless solid (1.7 g, 46%).



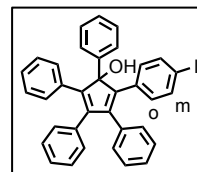
**<sup>1</sup>H NMR** (400 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 7.99 (s, 3H, H<sub>a</sub>), 7.97 (s, 3H, H<sub>d</sub>), 7.58 (d,  $J$  = 8.4 Hz, 3H, H<sub>b</sub>), 7.09 (dd,  $J$  = 8.2, 1.4 Hz, 3H, H<sub>c</sub>), 3.90 (s, 6H, H<sub>e</sub>), 2.43 (t,  $J$  = 7.2 Hz, 6H, H<sub>f</sub>), 1.58-1.51 (m, 7H, H<sub>g</sub> with H<sub>2</sub>O), 1.28-1.18 (m, 54H, CH<sub>2</sub> Alkyl), 0.88 (t,  $J$  = 6.8 Hz, 9H, CH<sub>3</sub>).

**<sup>13</sup>C NMR** (150 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 144.5, 137.0, 132.9, 122.3, 122.1, 120.6, 112.2, 37.3, 31.9, 31.6, 31.5, 29.7, 29.6, 29.5, 29.4, 29.3, 29.3, 28.9, 22.7, 14.2.

**HR-MS** (ESI, negative):  $m/z$  calc. for C<sub>60</sub>H<sub>94</sub>BN<sub>6</sub>S<sub>3</sub> [M-Ti]<sup>-</sup>: 1005.6795; found: 1005.6823.

### c. Synthesis of 3

1,4-diodobenzene (13.0 g, 39 mmol) was dissolved in anhydrous THF (100 mL). *n*-BuLi in hexane (1.6 M, 20 mL, 32 mmol) was added dropwise to the solution at -80 °C and the mixture was stirred for 30 min. A solution of tetraphenylcyclopentadienone (10.1 g, 26 mmol) in anhydrous THF (100 mL) was added to the reaction mixture *via* canula and the resulting mixture was stirred at -80 °C for 1 h, then gradually warmed to room temperature. The reaction was quenched by adding saturated aqueous NH<sub>4</sub>Cl (100 mL) and the mixture was extracted with AcOEt (100 mL). The extract was washed with H<sub>2</sub>O (100 mL x 2) and brine (100 mL x 2), dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>, filtered and evaporated under vacuum. The crude compound was washed with hexane and the precipitate was filtered to afford **3** as a pale yellow solid (14.3 g, 93%).



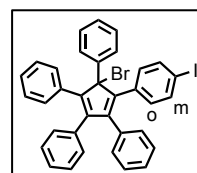
**<sup>1</sup>H NMR** (400 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 7.57 (d, *J* = 8.4 Hz, 2H, H<sub>m</sub>), 7.32 (d, *J* = 8.0 Hz, 2H, H<sub>o</sub>), 7.17-7.04 (m, 12H, H<sub>Ar</sub>), 7.00-6.94 (m, 8H, H<sub>Ar</sub>), 2.46 (s, 1H, OH).

**<sup>13</sup>C NMR** (150 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 147.5, 142.8, 140.4, 137.5, 134.8, 133.5, 129.8, 129.4, 127.9, 127.9, 127.2, 127.2, 92.4, 89.8.

**HR-MS** (EI, positive): *m/z* calc. for C<sub>35</sub>H<sub>25</sub>IO [**M**]<sup>+</sup>: 588.0950; found: 588.0955.

#### d. Synthesis of **4**

A suspension of **3** (14.0 g, 24 mmol) in AcOH (80 mL) was refluxed until its full dissolution. HBr in AcOH (30%, 50 mL) was then added to the mixture and the resulting mixture refluxed for 3 h. After cooling the reaction mixture to room temperature, H<sub>2</sub>O (100 mL) was added and a solid precipitated. The precipitate was filtered and washed with H<sub>2</sub>O to obtain the crude compound which was pre-adsorbed on silica gel (50 mL) and purified by silica gel column chromatography (hexane : CH<sub>2</sub>Cl<sub>2</sub> = 4 : 1) to afford **4** as a mixture of regioisomers (11.5 g, 74%). This compound was used in the next step without further purification.



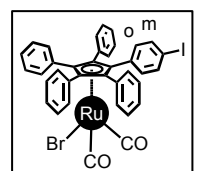
**<sup>1</sup>H NMR** (400 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 7.55 (d, *J* = 8.8 Hz, 2H), 7.46-7.40 (m, 19H), 7.35 (d, *J* = 8.8 Hz, 7H), 7.25-7.22 (m, 20H), 7.20-7.01 (m, 76H), 6.98-6.91 (m, 49H), 6.73-6.67 (m, 13H).

**<sup>13</sup>C NMR** (150 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 149.1, 148.7, 146.9, 142.5, 141.7, 141.3, 140.5, 137.4, 136.8, 136.6, 135.5, 135.4, 134.4, 134.1, 133.9, 133.8, 133.6, 132.1, 131.9, 130.4, 130.4, 130.3, 130.0, 129.9, 128.5, 128.4, 128.0, 127.9, 127.9, 127.8, 127.7, 127.6, 127.4, 127.4, 127.2, 127.2, 127.2, 93.3.

**HR-MS** (EI, positive): *m/z* calc. for C<sub>35</sub>H<sub>24</sub>BrI [**4**]<sup>+</sup>: 650.0106; found: 650.0109.

#### e. Synthesis of **5**

A mixture of **4** (3.7 g, 5.6 mmol) and Ru<sub>3</sub>(CO)<sub>12</sub> (1.3 g, 2.0 mmol) in anhydrous toluene (80 mL) was refluxed for 2 h. The reaction mixture was pre-adsorbed on silica gel (20 mL) and purified by silica gel column chromatography (hexane : CH<sub>2</sub>Cl<sub>2</sub> = 7 : 3 to 5 : 5) to give **5** as a yellow solid (2.9 g, 64%).



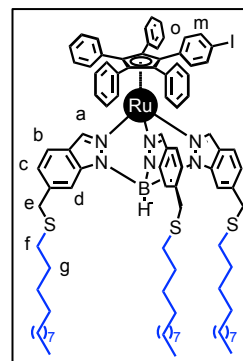
**<sup>1</sup>H NMR** (400 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 7.42 (d, *J* = 8.4 Hz, 2H, H<sub>m</sub>), 7.23-7.18 (m, 4H, H<sub>Ar(yl)</sub>), 7.16-7.08 (m, 8H, H<sub>Ar(yl)</sub>), 7.05-6.99 (m, 8H, H<sub>Ar(yl)</sub>), 6.75 (d, *J* = 8.8 Hz, 2H, H<sub>o</sub>).

**<sup>13</sup>C NMR** (150 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 196.3, 136.9, 134.0, 132.3, 132.2, 129.3, 129.3, 129.2, 128.6, 128.5, 128.0, 127.9, 127.8, 106.3, 106.2, 106.0, 94.9.

**HR-MS** (ESI, positive): *m/z* calc. for C<sub>37</sub>H<sub>24</sub>BrIO<sub>2</sub>RuNa [**M**+Na]<sup>+</sup>: 830.8946; found: 830.8945.

#### f. Synthesis of **6**

**2** (605 mg, 500  $\mu\text{mol}$ ) and **5** (324 mg, 400  $\mu\text{mol}$ ) were introduced to a MW pressure tube. After substitution of inner atmosphere of the tube into  $\text{N}_2$ , anhydrous MeCN (20 mL) was added. The resulting mixture was heated under microwave irradiation (120  $^\circ\text{C}$ , 30 min, 2 cycles). The reaction mixture was transferred into a conical tube and centrifuged. The supernatant was removed by decantation and  $\text{CH}_2\text{Cl}_2$  was added. The insoluble parts in  $\text{CH}_2\text{Cl}_2$  were removed by centrifugation and the supernatant was evaporated to afford the crude compound. The crude compound was purified by silica gel column chromatography (hexane :  $\text{CH}_2\text{Cl}_2$  = 7 : 3) to give **6** as a yellow solid (580 mg, 43 %).



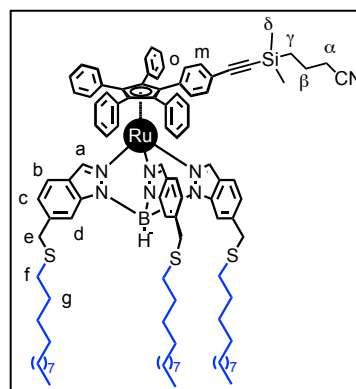
**$^1\text{H}$  NMR** (400 MHz,  $\text{CDCl}_3/\text{TMS}$ ):  $\delta$  = 7.86 (s, 6H,  $\text{H}_{d,a}$ ), 7.34-7.28 (m, 12H,  $\text{H}_{Ar}$ ), 7.12-7.07 (m, 6H,  $\text{H}_{Ar}$ ), 7.03-6.96 (m, 12H,  $\text{H}_{Ar}$ ), 3.87 (s, 6H,  $\text{H}_e$ ), 2.48 (t,  $J$  = 7.4 Hz, 6H,  $\text{H}_f$ ), 1.61 (quint,  $J$  = 7.5 Hz, 6H,  $\text{H}_g$ ), 1.39-1.25 (m, 54H,  $\text{CH}_2_{\text{Alkyl}}$ ), 0.88 (t,  $J$  = 6.8 Hz, 9H,  $\text{CH}_3$ )

**$^{13}\text{C}$  NMR** (150 MHz,  $\text{CDCl}_3/\text{TMS}$ ):  $\delta$  = 143.6, 140.4, 140.3, 137.1, 137.0, 136.3, 135.3, 133.9, 133.7, 133.6, 133.6, 133.5, 127.3, 127.2, 127.2, 127.1, 127.0, 122.1, 122.0, 121.9, 120.0, 111.2, 93.2, 88.1, 87.9, 87.7, 86.8, 37.1, 31.9, 31.7, 29.7, 29.7, 29.6, 29.4, 29.3, 29.0, 22.7, 14.1.

**HR-MS** (ESI, positive):  $m/z$  calc. for  $\text{C}_{95}\text{H}_{118}\text{BIN}_6\text{RuS}_3$  [ $\text{M}$ ] $^+$ : 1678.6761; found: 1678.6756.

#### g. Synthesis of **7**

**6** (500 mg, 298  $\mu\text{mol}$ ), (3-cyanopropyl)-dimethylsilylacetylene<sup>[1]</sup> (451 mg, 3.0 mmol), XPhos Pd G3 (26 mg, 30  $\mu\text{mol}$ ), XPhos (19 mg, 41  $\mu\text{mol}$ ) and CuI (13 mg, 68  $\mu\text{mol}$ ) were suspended in anhydrous toluene (10 mL) and anhydrous  $\text{Et}_3\text{N}$  (10 mL). The resulting mixture was deoxygenated by 3 cycles of a freeze-pump-thaw method and then refluxed for 20 h. The reaction mixture was filtered through Celite and the filtrate evaporated under vacuum. The residue was extracted with hexane (20 mL). The insolubilities were filtered off and the filtrate was evaporated under vacuum. The crude compound was purified by silica gel column chromatography (hexane :  $\text{CH}_2\text{Cl}_2$  = 3 : 2 – 1 : 1), followed by recycling GPC (JAIGEL 1H-2H, eluent :  $\text{CHCl}_3$ ) to afford **7** as an orange solid (396 mg, 78%).



**$^1\text{H}$  NMR** (400 MHz,  $\text{CDCl}_3/\text{TMS}$ ):  $\delta$  = 7.86 (s, 6H,  $\text{H}_{d,a}$ ), 7.34-7.28 (m, 12H,  $\text{H}_{Aryl}$ ), 7.12-7.08 (m, 6H,  $\text{H}_{Aryl}$ ), 7.02-6.97 (m, 12H,  $\text{H}_{Aryl}$ ), 3.87 (s, 6H,  $\text{H}_e$ ), 2.48 (t,  $J$  = 7.4 Hz, 6H,  $\text{H}_f$ ), 2.39 (t,  $J$  = 7.0 Hz, 2H,  $\text{H}_\alpha$ ), 1.77 (m, 2H,  $\text{H}_\beta$ ), 1.62 (quint,  $J$  = 7.4 Hz, 6H,  $\text{H}_g$ ), 1.39-1.25 (m, 54H,  $\text{CH}_2_{\text{Alkyl}}$ ), 0.88 (t,  $J$  = 6.8 Hz, 9H,  $\text{CH}_3$ ), 0.79 (m, 2H,  $\text{H}_\gamma$ ), 0.19 (s, 6H,  $\text{H}_\delta$ ).

**$^{13}\text{C}$  NMR** (150 MHz,  $\text{CDCl}_3/\text{TMS}$ ):  $\delta$  = 143.6, 140.3, 137.1, 135.3, 133.7, 133.7, 133.5, 133.4, 130.8, 127.3, 127.2, 127.2, 122.1, 121.9, 121.0, 120.0, 119.7, 111.2, 106.4, 92.6, 88.2, 87.6, 87.5, 37.1, 31.9, 31.7, 29.7, 29.7, 29.6, 29.4, 29.3, 29.0, 22.7, 20.6, 20.5, 15.7, 14.1, -1.9.

**HR-MS** (ESI, positive):  $m/z$  calc. for  $\text{C}_{103}\text{H}_{130}\text{BN}_7\text{RuS}_3\text{Si}$  [ $\text{M}$ ] $^+$ : 1701.8456; found: 1701.8476.

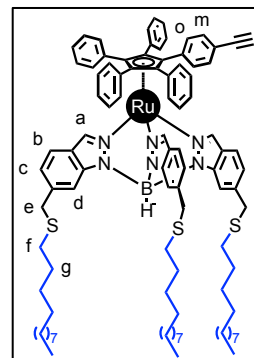
#### h. Synthesis of **8**

**7** (791 mg, 465  $\mu\text{mol}$ ) and  $\text{K}_2\text{CO}_3$  (642 mg, 4.7 mmol) was stirred in a mixture of MeOH (13 mL) and THF (13 mL) at room temperature for 1 h. The reaction mixture was diluted with  $\text{CH}_2\text{Cl}_2$  (200 mL). The organic phase was washed with  $\text{H}_2\text{O}$  (100 mL) and brine (100 mL x 2), dried over anhydrous  $\text{Na}_2\text{SO}_4$ , filtered and evaporated. The crude compound was purified by silica gel column chromatography (hexane :  $\text{CH}_2\text{Cl}_2$  = 2 : 1) to afford **8** as an orange solid (675 mg, 95%).

$^1\text{H NMR}$  (400 MHz,  $\text{CDCl}_3/\text{TMS}$ ):  $\delta$  = 7.87-7.86 (d,  $J$  = 1.8 Hz, 6H,  $\text{H}_{\text{d,a}}$ ), 7.35-7.28 (m, 12H,  $\text{H}_{\text{Aryl}}$ ), 7.13-7.08 (m, 6H,  $\text{H}_{\text{Aryl}}$ ), 7.02-6.97 (m, 12H,  $\text{H}_{\text{Aryl}}$ ), 3.87 (s, 6H,  $\text{H}_{\text{e}}$ ), 3.02 (s, 1H, alkyne proton), 2.47 (t,  $J$  = 7.4 Hz, 6H,  $\text{H}_{\text{f}}$ ), 1.60 (quint,  $J$  = 7.4 Hz, 6H,  $\text{H}_{\text{g}}$ ), 1.39-1.25 (m, 54H,  $\text{CH}_2$  Alkyl), 0.88 (t,  $J$  = 6.8 Hz, 9H,  $\text{CH}_3$ ).

$^{13}\text{C NMR}$  (150 MHz,  $\text{CDCl}_3/\text{TMS}$ ):  $\delta$  = 143.6, 140.3, 137.1, 135.3, 133.7, 133.7, 133.5, 133.4, 130.9, 127.3, 127.2, 127.2, 127.1, 122.1, 121.9, 120.5, 120.0, 111.2, 88.1, 87.7, 87.3, 83.6, 37.1, 31.9, 31.7, 29.7, 29.7, 29.6, 29.4, 29.3, 29.0, 22.7, 14.1.

**HR-MS** (ESI, positive):  $m/z$  calc. for  $\text{C}_{97}\text{H}_{119}\text{BN}_6\text{RuS}_3$  [ $\text{M}$ ] $^+$ : 1576.7795; found: 1576.7824.



#### i. Synthesis of **9**

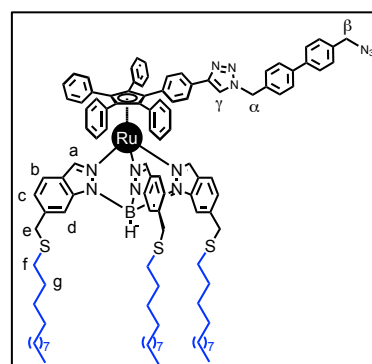
**8** (507 mg, 321  $\mu\text{mol}$ ), 4,4'-bis(azidomethyl)-1,1'-biphenyl<sup>[2]</sup> (841 mg, 3.2 mmol), CuI (35 mg, 181  $\mu\text{mol}$ ) and TBTA (86 mg, 161  $\mu\text{mol}$ ) were suspended in anhydrous THF (32 mL). The resulting mixture was refluxed for 20 h. After removal of volatiles under vacuum, the crude compound was purified by silica gel column chromatography ( $\text{CHCl}_3$ ) to afford **9** as an orange solid (489 mg, 84%).

$^1\text{H NMR}$  (400 MHz,  $\text{CDCl}_3/\text{TMS}$ ):  $\delta$  = 7.90 (s, 6H,  $\text{H}_{\text{d}}$ ), 7.87 (s, 6H,  $\text{H}_{\text{a}}$ ), 7.58-7.55 (m, 5H,  $\text{H}_{\text{Aryl}}$ ), 7.45-7.32 (m, 17H,  $\text{H}_{\text{Aryl}}$ ), 7.28 (s, 1H,  $\text{H}_{\text{y}}$ ), 7.11-7.07 (m, 4H,  $\text{H}_{\text{Aryl}}$ ), 7.00-6.96 (m, 12H,  $\text{H}_{\text{Aryl}}$ ), 5.56 (s, 2H,  $\text{H}_{\alpha}$ ), 4.39 (s, 2H,  $\text{H}_{\beta}$ ), 3.87 (s, 6H,  $\text{H}_{\text{e}}$ ), 2.47 (t,  $J$  = 7.4 Hz, 6H,  $\text{H}_{\text{f}}$ ), 1.61 (quint,  $J$  = 7.5 Hz, 6H,  $\text{H}_{\text{g}}$ ), 1.39-1.25 (m, 54H,  $\text{CH}_2$  Alkyl), 0.88 (t,  $J$  = 6.8 Hz, 9H,  $\text{CH}_3$ ).

$^{13}\text{C NMR}$  (150 MHz,  $\text{CDCl}_3/\text{TMS}$ ):  $\delta$  = 148.0, 143.6, 141.1, 140.3, 140.2, 136.9, 134.8, 134.2, 133.9, 133.9, 133.8, 133.6, 129.0, 128.7, 128.6, 127.8, 127.5, 127.2, 127.1, 127.1, 124.4, 122.1, 121.9, 120.0, 119.5, 111.2, 88.2, 87.9, 87.3, 54.4, 53.9, 37.2, 31.9, 31.8, 29.7, 29.7, 29.6, 29.4, 29.2, 29.0, 22.7, 14.1.

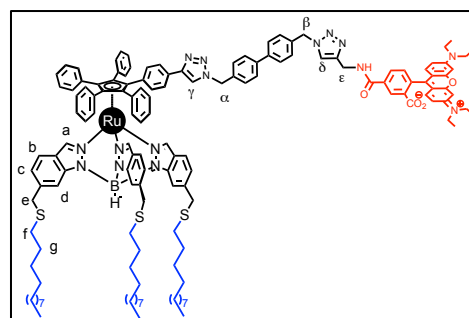
**HR-MS** (ESI, positive):  $m/z$  calc. for  $\text{C}_{111}\text{H}_{132}\text{BN}_{12}\text{RuS}_3$  [ $\text{M}+\text{H}$ ] $^+$ : 1841.8997; found: 1841.9000.

**UV-Vis** (conc. 10  $\mu\text{M}$  in chloroform)  $\lambda_{\text{max}}$  ( $\epsilon$ ) : 333 nm (27250  $\text{mol}^{-1}\text{dm}^3\text{cm}^{-1}$ ).



#### j. Synthesis of **10**

**9** (295 mg, 160  $\mu\text{mol}$ ), alkyne-substituted rhodamine derivative<sup>[4]</sup> (102 mg, 195  $\mu\text{mol}$ ), CuI (19 mg, 100  $\mu\text{mol}$ ) and TBTA (44 mg, 82  $\mu\text{mol}$ ) were suspended in anhydrous THF (17 mL). The resulting mixture was refluxed for 18 h. After removal of volatiles under vacuum, the crude compound was purified by silica gel column chromatography ( $\text{CHCl}_3$  : MeOH = 10 : 1) to give **10** as a brown solid (270 mg, 71%).



**<sup>1</sup>H NMR** (400 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 8.38 (br, 1H, NH), 8.11 (d,  $J$  = 0.8 Hz, 1H, H<sub>δ</sub>), 7.93 (s, 3H, H<sub>d</sub>), 7.89 (s, 3H, H<sub>a</sub>), 7.63 (br, 1H), 7.56-7.53 (m, 5H, H<sub>Ar</sub>), 7.44-7.42 (m, 2H, H<sub>Ar</sub>), 7.38-7.31 (m, 14H, H<sub>Ar</sub>), 7.28-7.26 (m, 12H, H<sub>Ar</sub> with CHCl<sub>3</sub>), 7.11-7.06 (m, 4H, H<sub>Ar</sub>), 7.00-6.96 (m, 12H, H<sub>Ar</sub>), 6.55 (d,  $J$  = 9.2 Hz, 2H, H<sub>xanthene</sub>), 6.44 (d,  $J$  = 2.0 Hz, 2H, H<sub>xanthene</sub>), 6.33 (d,  $J$  = 8.0 Hz, 2H, H<sub>xanthene</sub>), 5.55 (s, 4H, H<sub>α-β</sub>), 4.75 (s, 2H, H<sub>ε</sub>), 3.89 (s, 6H, H<sub>e</sub>), 3.35 (q,  $J$  = 6.9 Hz, 8H, NEt (CH<sub>2</sub>)), 2.49 (t,  $J$  = 7.2 Hz, 6H, H<sub>f</sub>), 1.66-1.58 (m, 14H, alkyl chains including H<sub>2</sub>O), 1.39-1.25 (m, 54H, CH<sub>2</sub> Alkyl), 1.16 (t,  $J$  = 7.2 Hz, 12H, NEt (CH<sub>3</sub>)), 0.87 (t,  $J$  = 6.8 Hz, 9H, CH<sub>3</sub>).

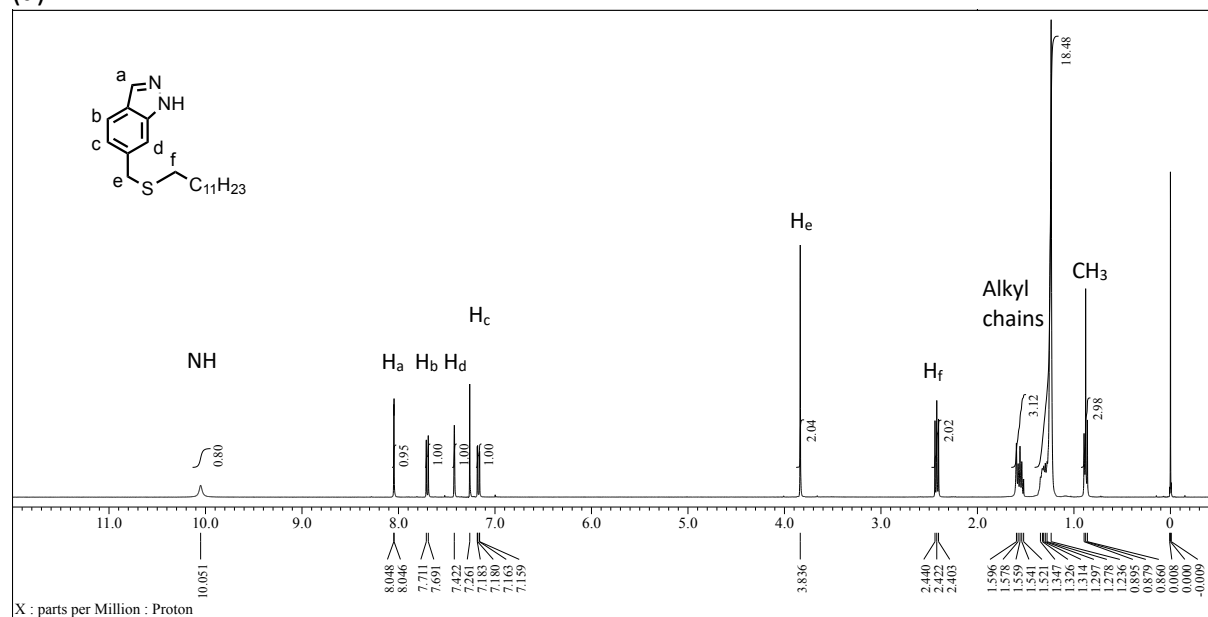
**<sup>13</sup>C NMR** (150 MHz, CDCl<sub>3</sub>/TMS):  $\delta$  = 166.2, 153.8, 148.0, 143.6, 140.7, 140.3, 136.8, 134.2, 133.9, 133.9, 133.8, 133.6, 129.2, 128.9, 128.7, 128.5, 127.8, 127.2, 127.1, 124.4, 122.1, 121.9, 120.0, 119.5, 111.3, 108.6, 97.4, 88.2, 88.0, 87.2, 53.8, 44.6, 37.3, 31.9, 29.7, 29.7, 29.6, 29.4, 29.2, 29.0, 22.7, 14.1, 12.5.

**HR-MS** (ESI, positive):  $m/z$  calc. for C<sub>143</sub>H<sub>165</sub>BN<sub>15</sub>O<sub>4</sub>RuS<sub>3</sub> [M+H]<sup>+</sup> 2365.1468; found: 2365.1184.

**UV-Vis** (conc. 10 μM in chloroform)  $I_{\max}$  (e) : : 550 nm (16270 mol<sup>-1</sup>dm<sup>3</sup>cm<sup>-1</sup>).

**Emission** ( $\lambda_{\text{exc}}$  = 520 nm, conc. 10 μM in chloroform)  $\lambda_{\max}$  : 569 nm.

(a)



(b)

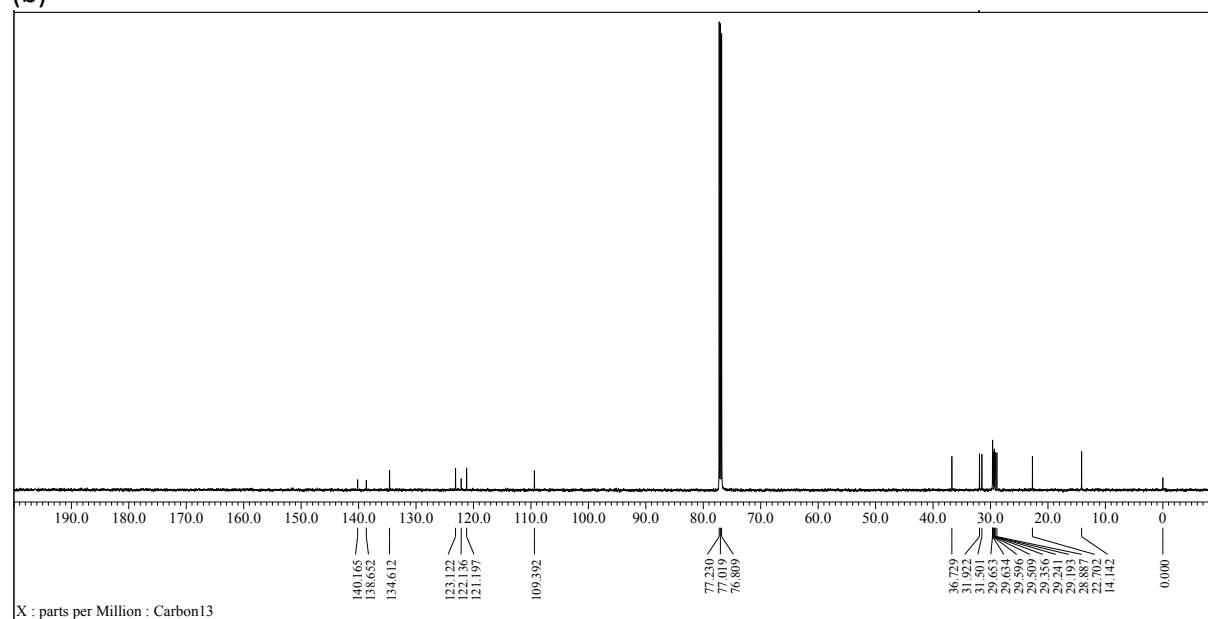


Figure S1 (a) 400 MHz  $^1\text{H}$  and (b) 150 MHz  $^{13}\text{C}$  NMR spectrum of **1** in  $\text{CDCl}_3$

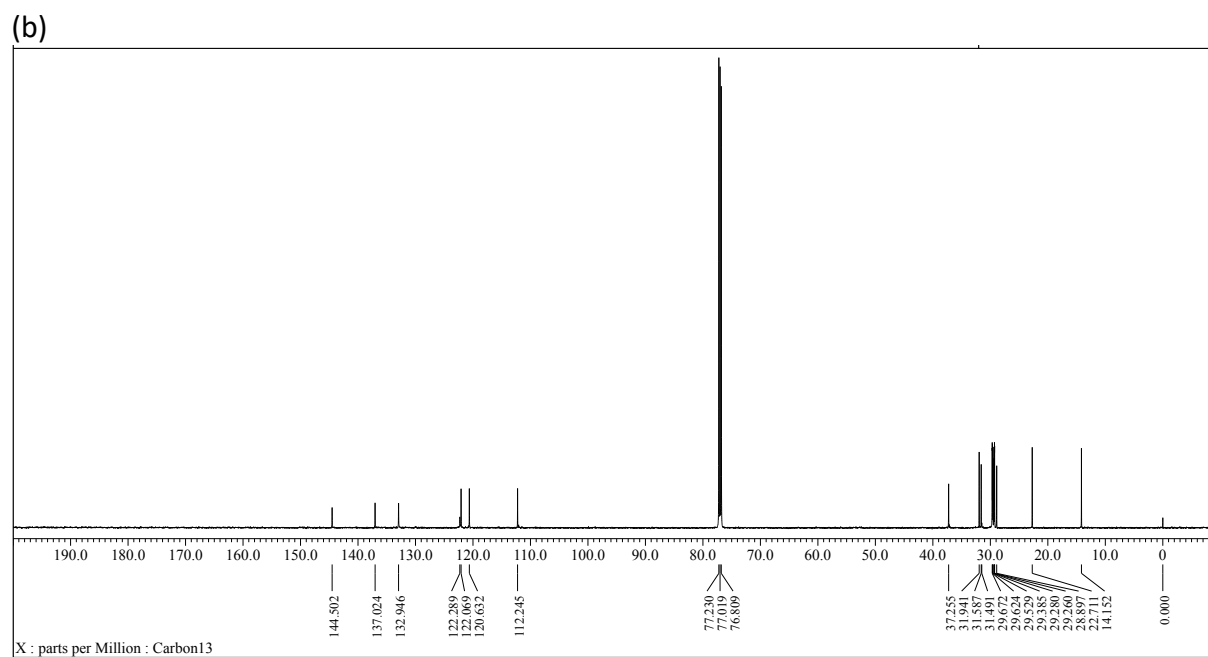
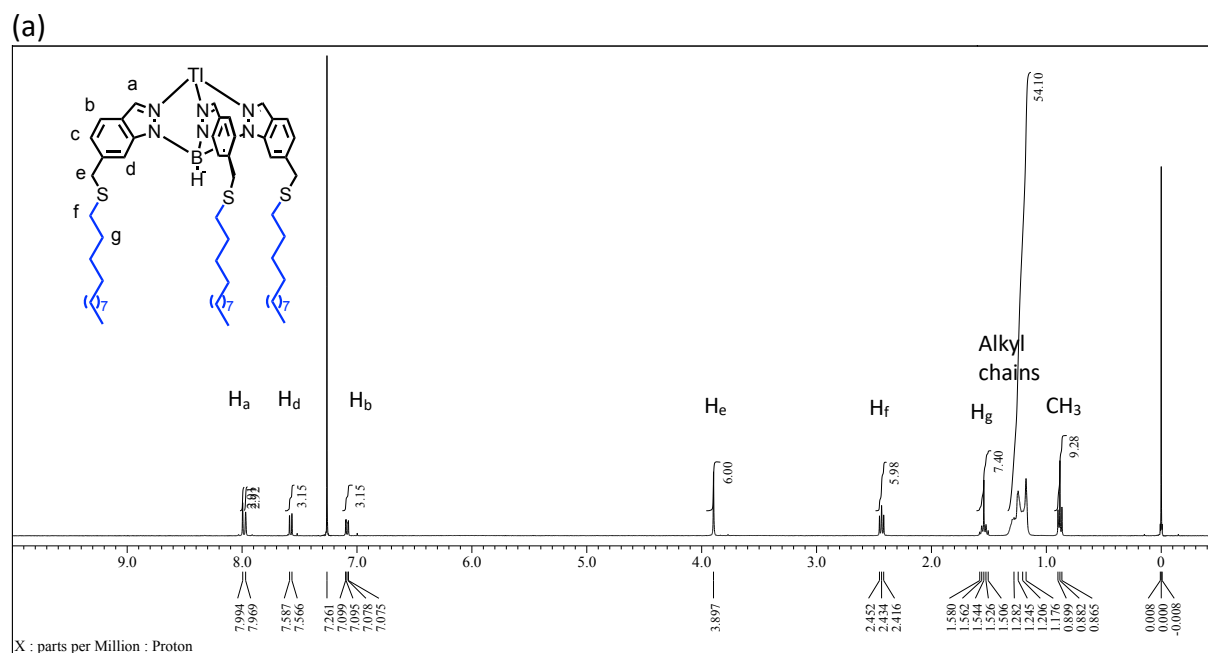
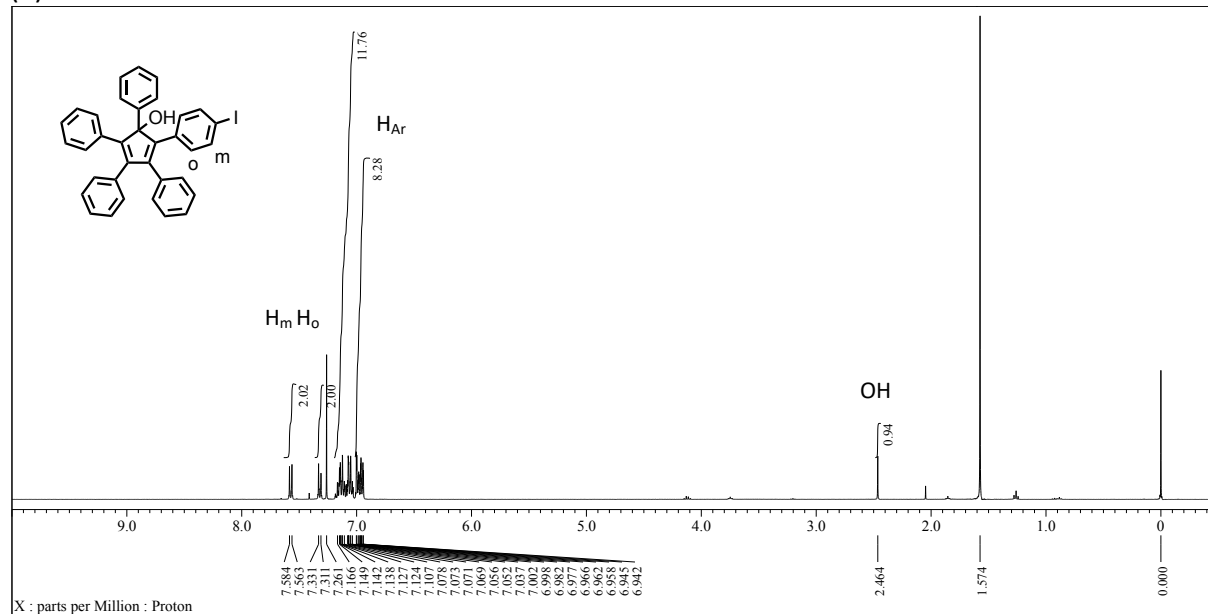


Figure S2 (a) 400 MHz  $^1\text{H}$  and (b) 150 MHz  $^{13}\text{C}$  NMR spectrum of **2** in  $\text{CDCl}_3$



(a)



(b)

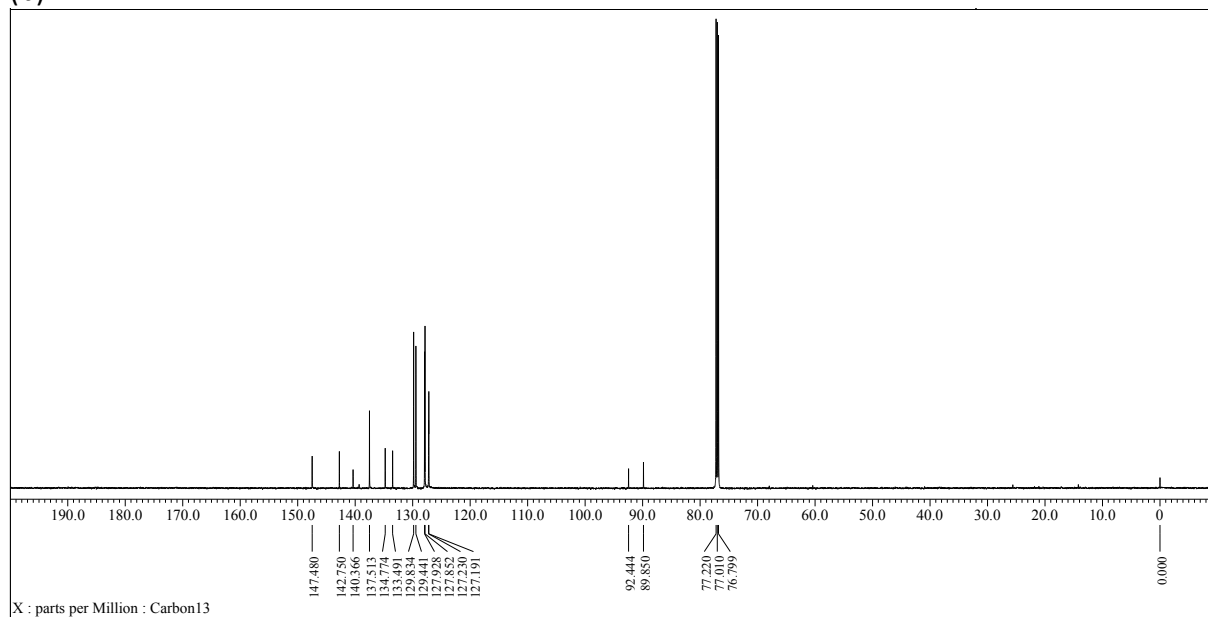
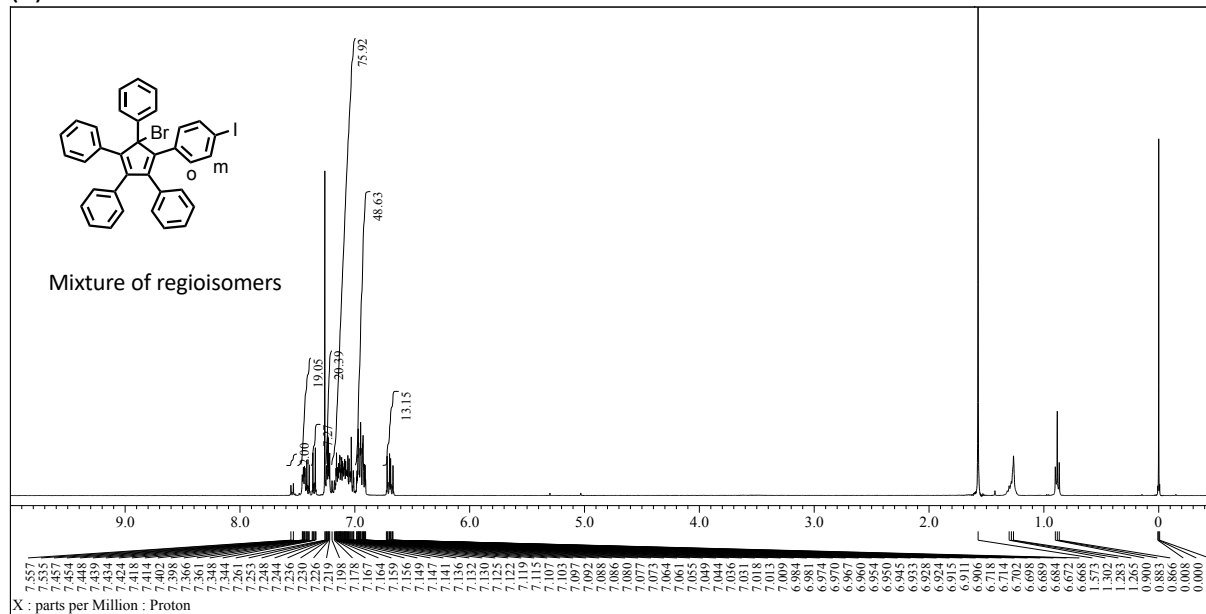


Figure S3 (a) 400 MHz  $^1\text{H}$  and (b) 150 MHz  $^{13}\text{C}$  NMR spectrum of **3** in  $\text{CDCl}_3$

(a)



(b)

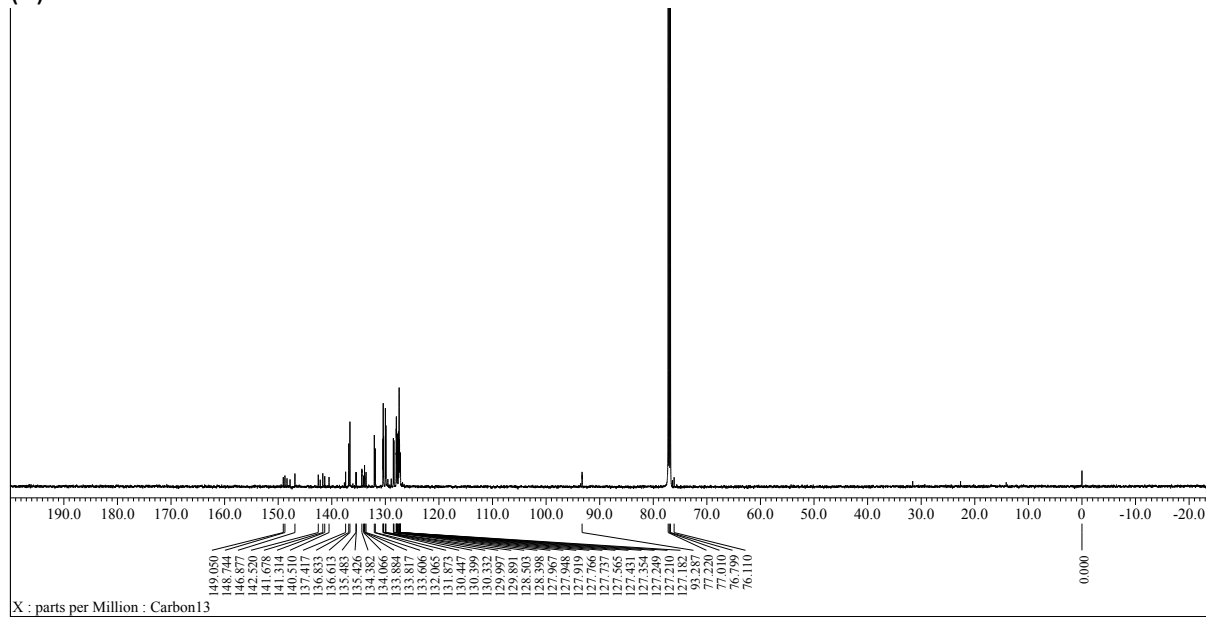


Figure S4 (a) 400 MHz  $^1\text{H}$  and (b) 150 MHz  $^{13}\text{C}$  NMR spectrum of **4** in  $\text{CDCl}_3$  (mixture of regioisomers)

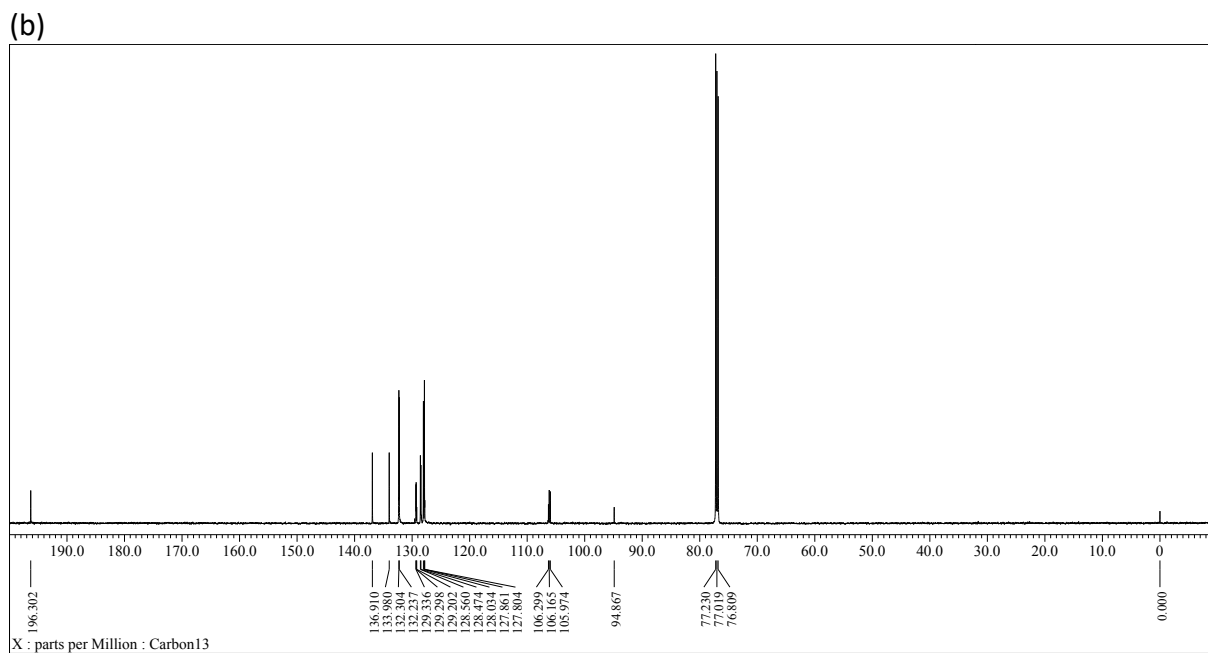
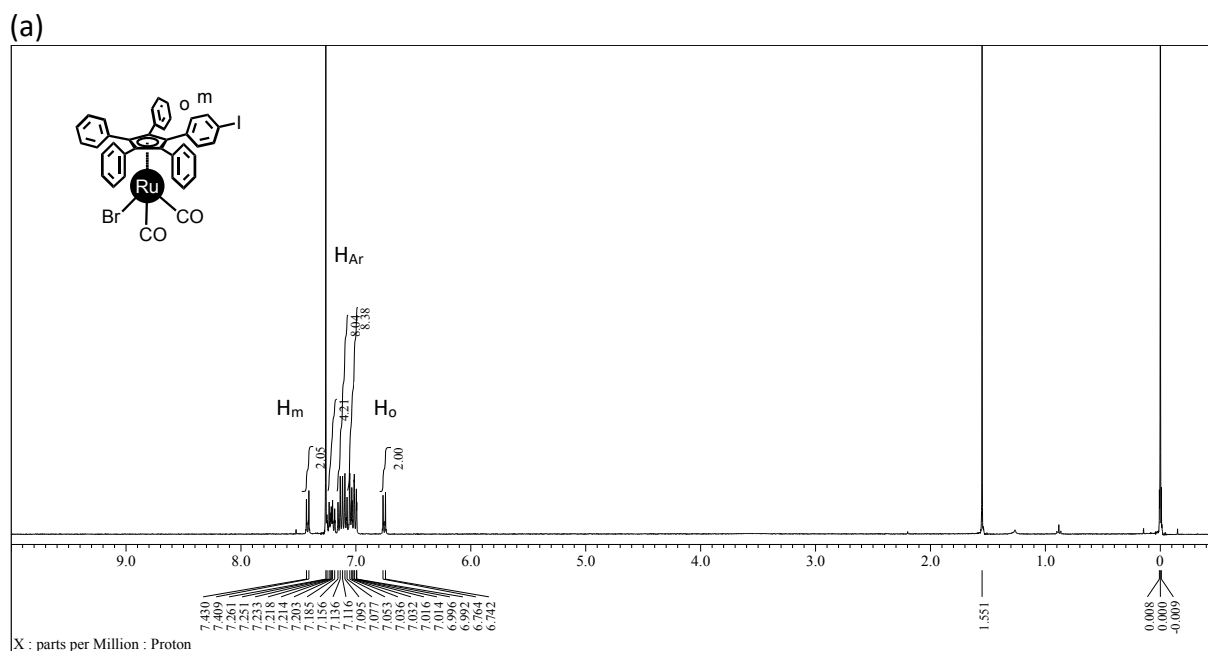


Figure S5 (a) 400 MHz  $^1\text{H}$  and (b) 150 MHz  $^{13}\text{C}$  NMR spectrum of **5** in  $\text{CDCl}_3$

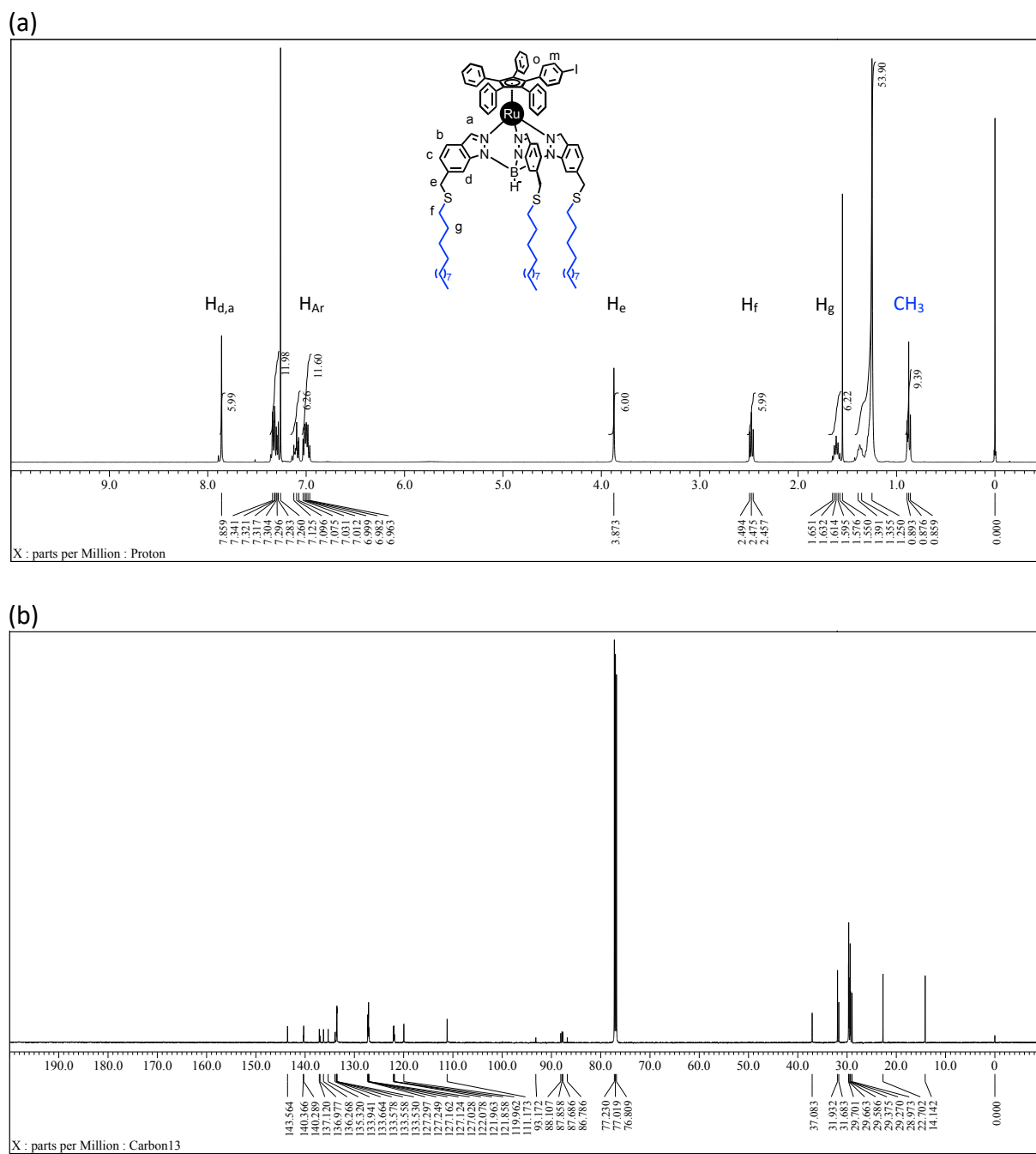


Figure S6 (a) 400 MHz <sup>1</sup>H and (b) 150 MHz <sup>13</sup>C NMR spectrum of **6** in CDCl<sub>3</sub>



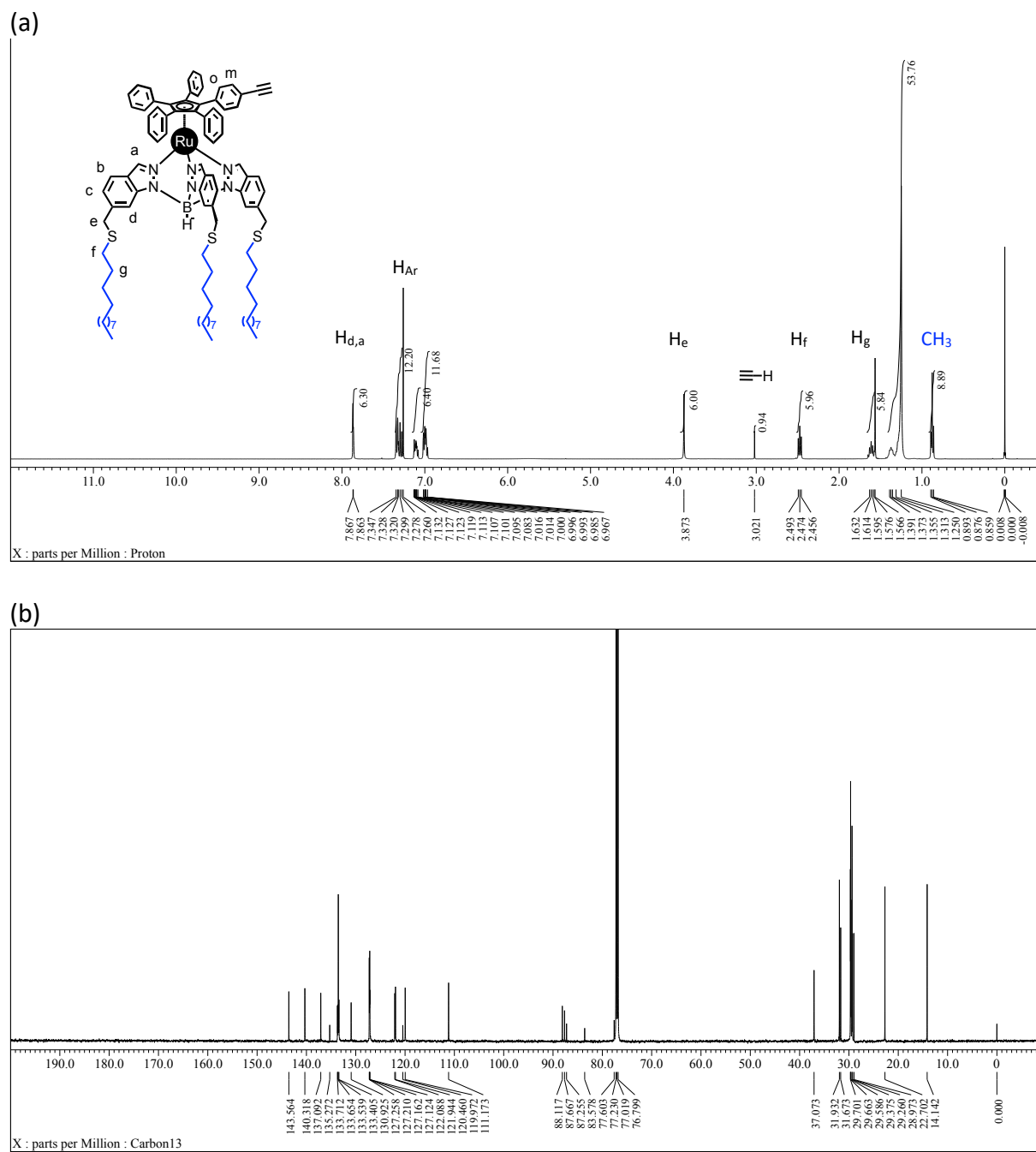


Figure S8 (a) 400 MHz  $^1\text{H}$  and (b) 150 MHz  $^{13}\text{C}$  NMR spectrum of **8** in  $\text{CDCl}_3$

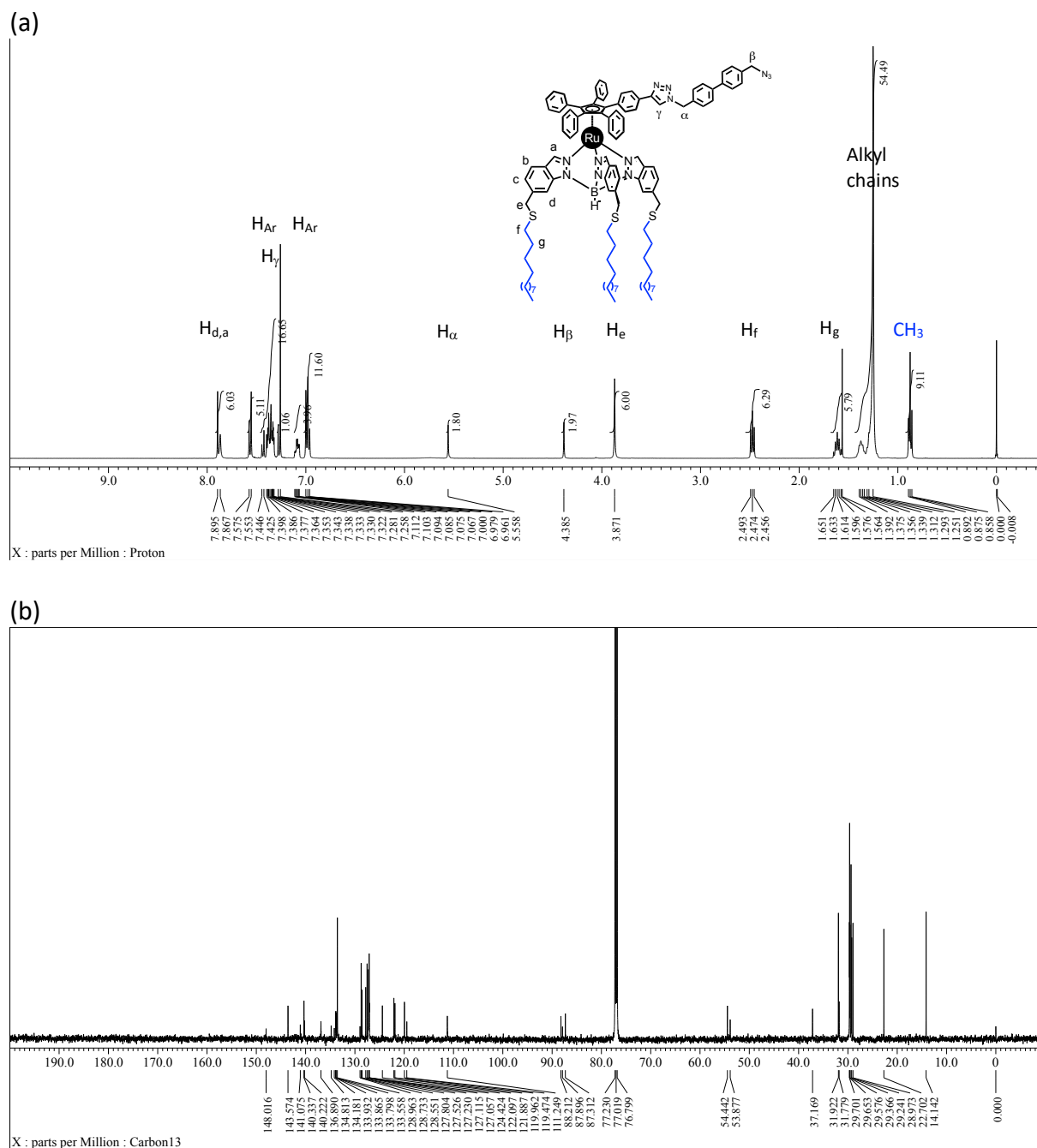


Figure S9 (a) 400 MHz  $^1\text{H}$  and (b) 150 MHz  $^{13}\text{C}$  NMR spectrum of **9** in  $\text{CDCl}_3$

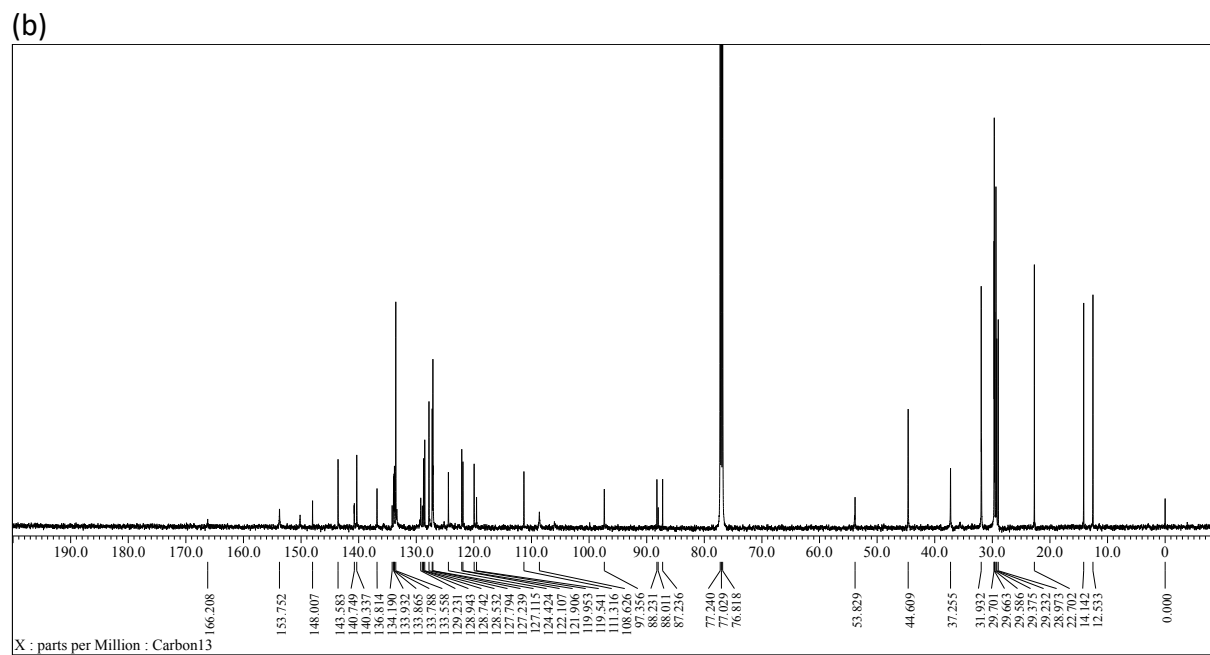
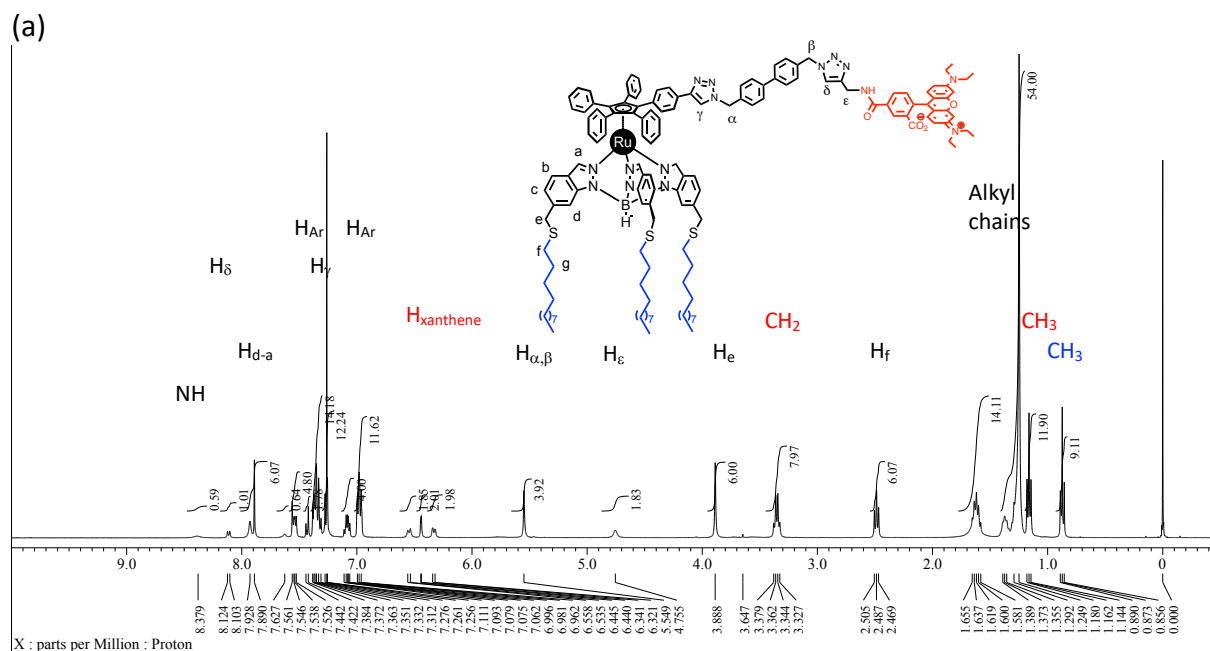


Figure S10 (a) 400 MHz  $^1\text{H}$  and (b) 150 MHz  $^{13}\text{C}$  NMR spectrum of **10** in  $\text{CDCl}_3$



#### 4. HR-MS spectra

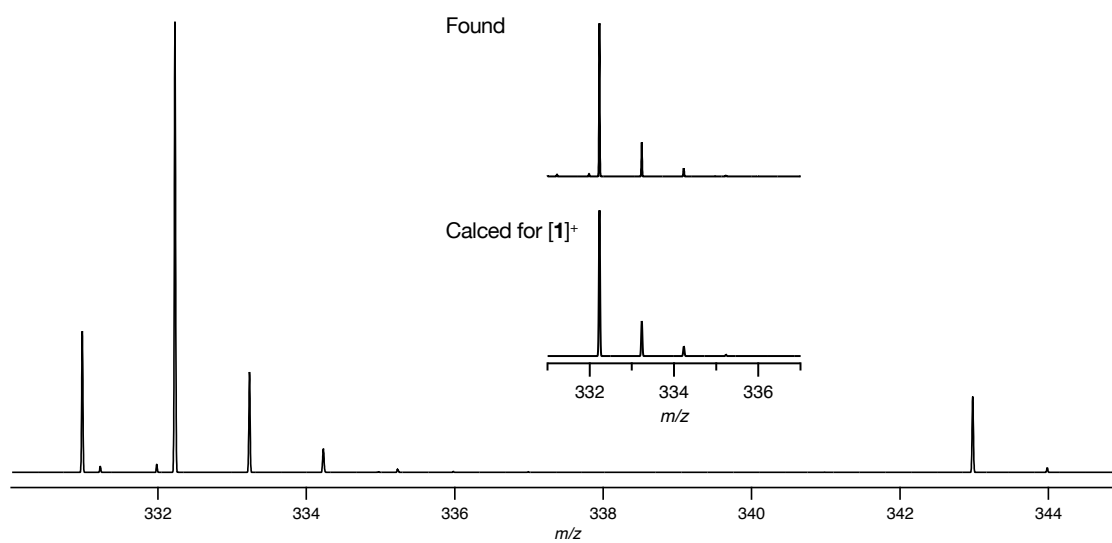


Figure S11 EI MS spectrum of **1**

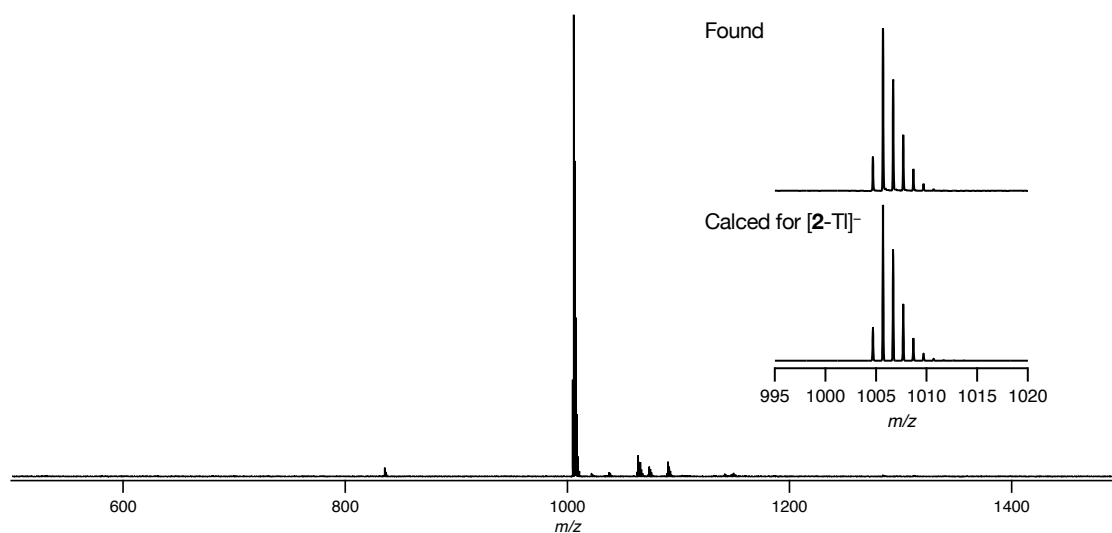


Figure S12 ESI-TOF MS spectrum of **2**

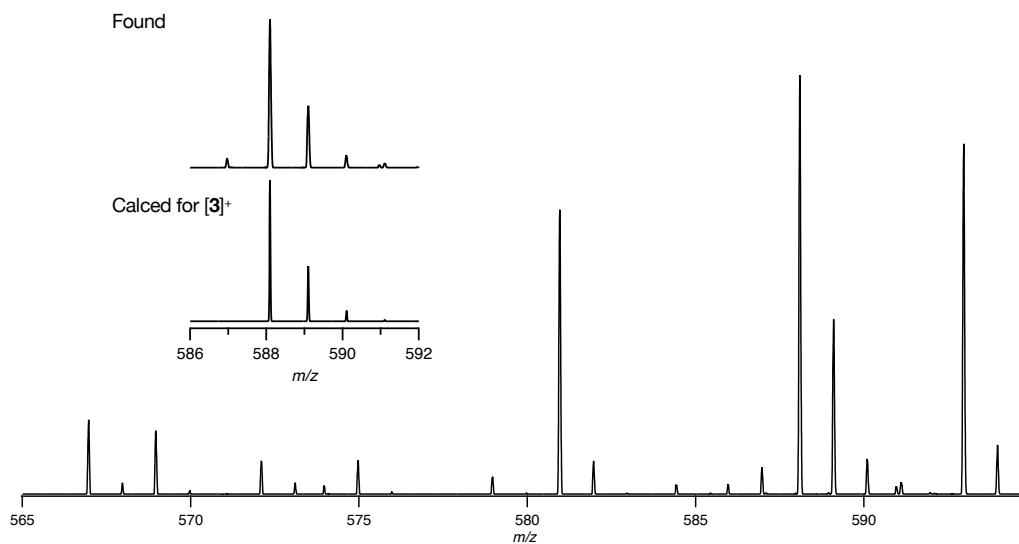


Figure S13 EI MS spectrum of **3**

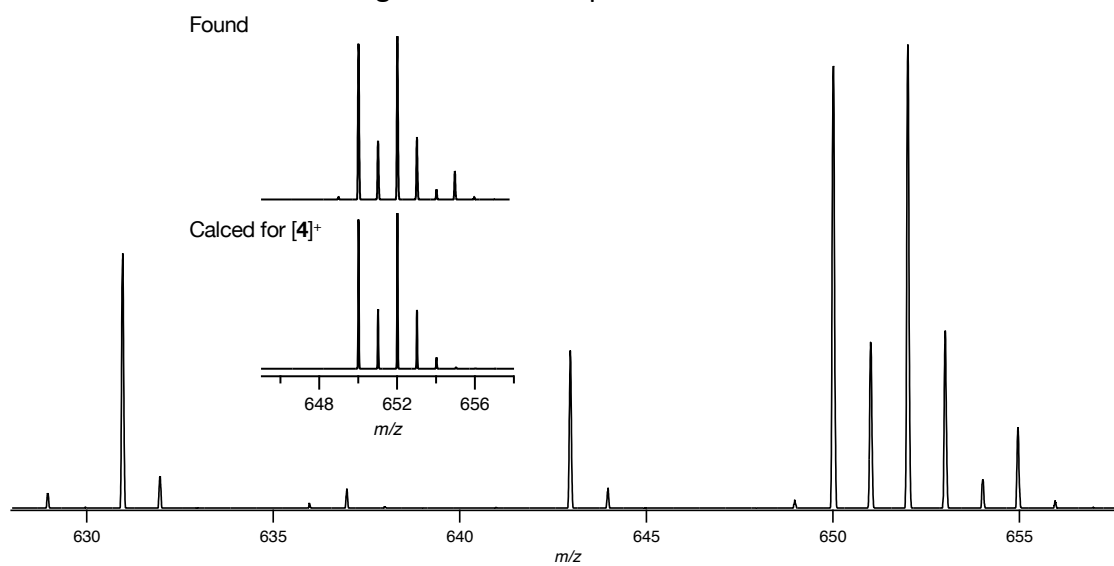


Figure S14 EI MS spectrum of **4**

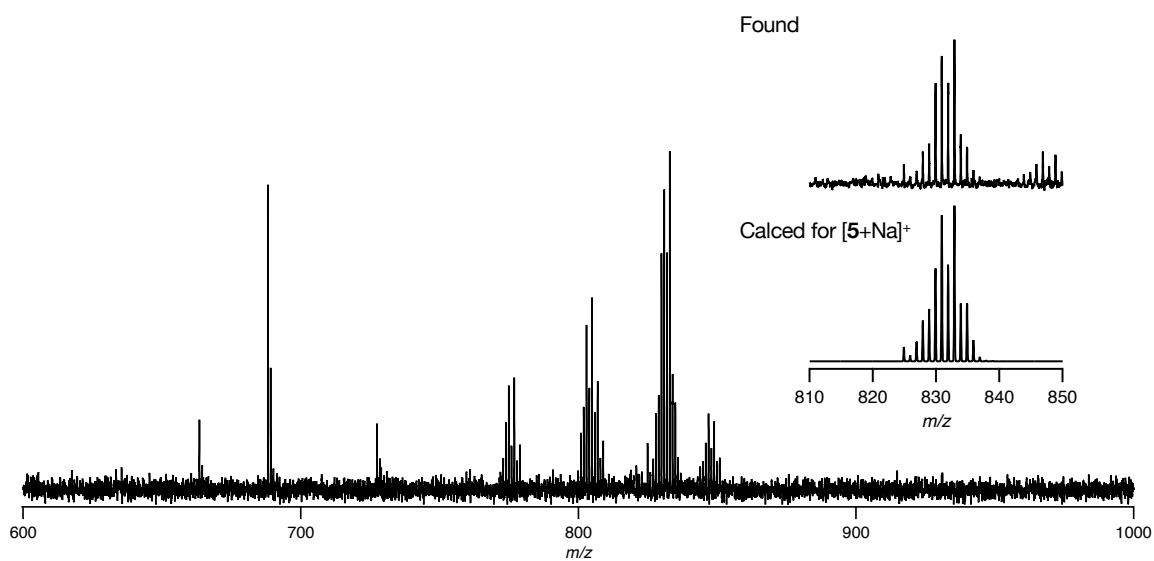


Figure S15 ESI-TOF MS spectrum of **5**

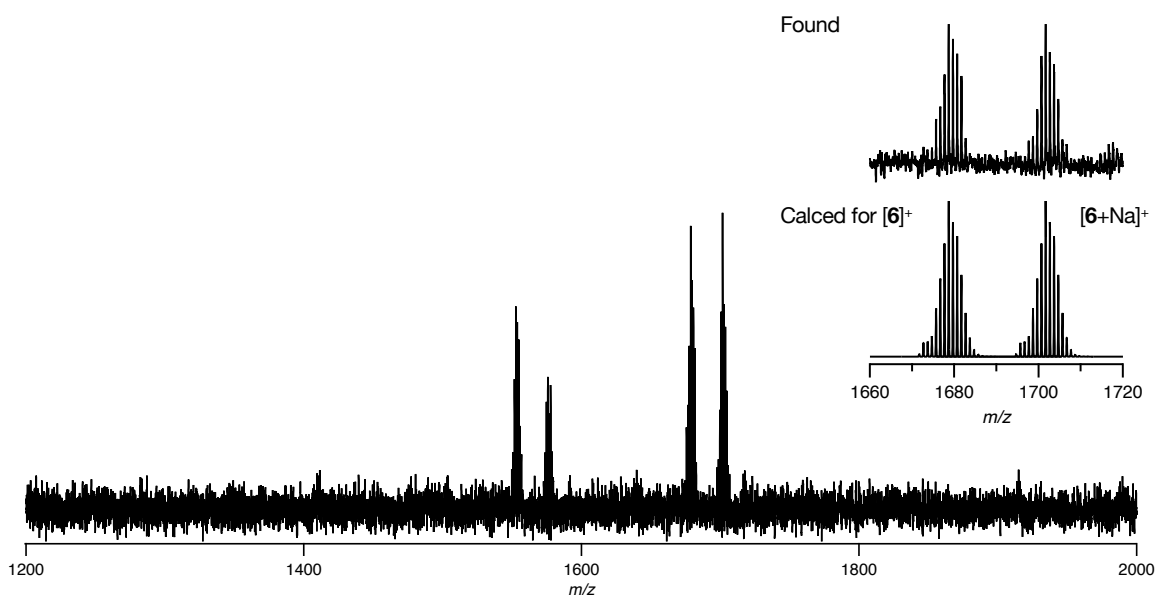


Figure S16 ESI-TOF MS spectrum of **6**

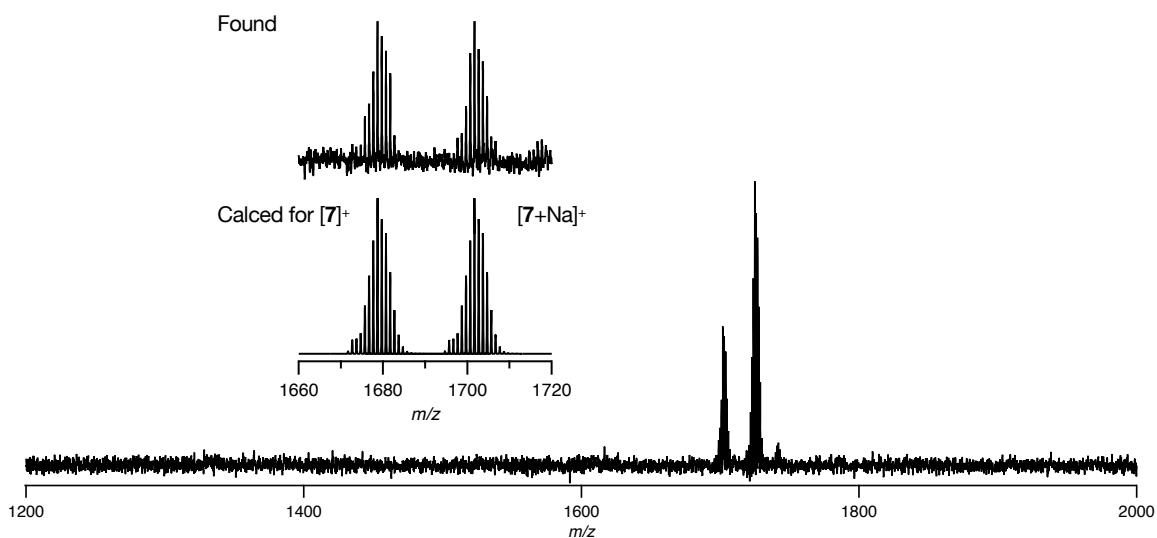


Figure S17 ESI-TOF MS spectrum of **7**

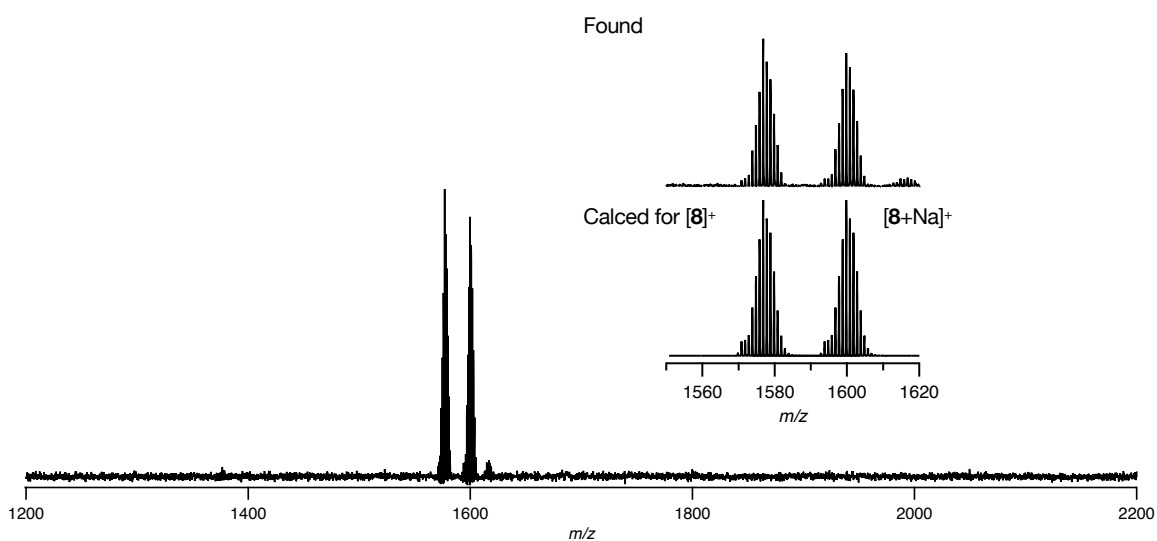


Figure S18 ESI-TOF MS spectrum of **8**

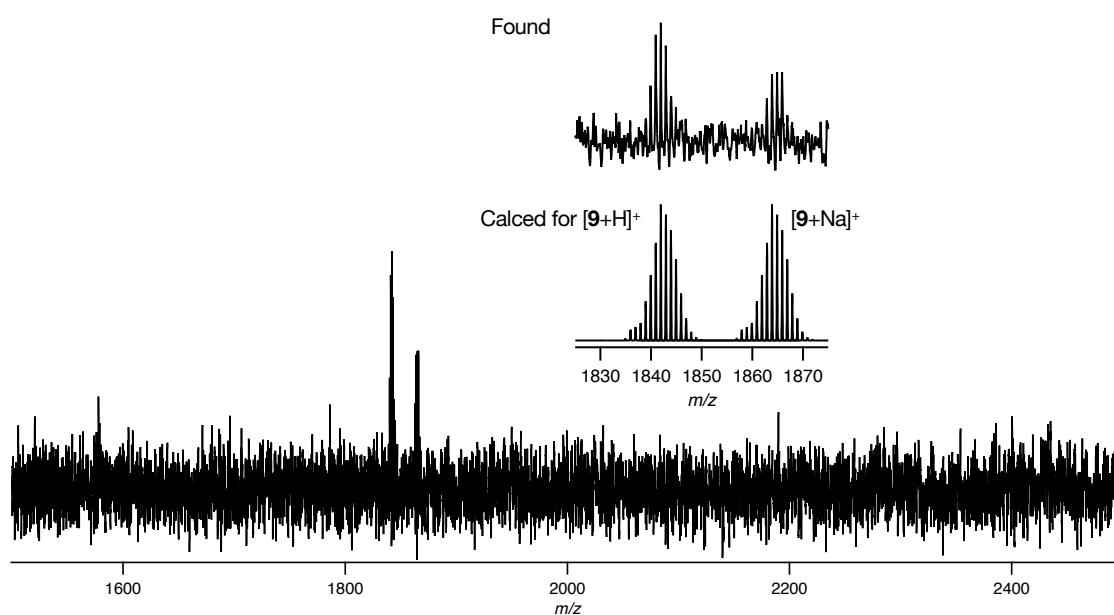


Figure S19 ESI-TOF MS spectrum of **9**

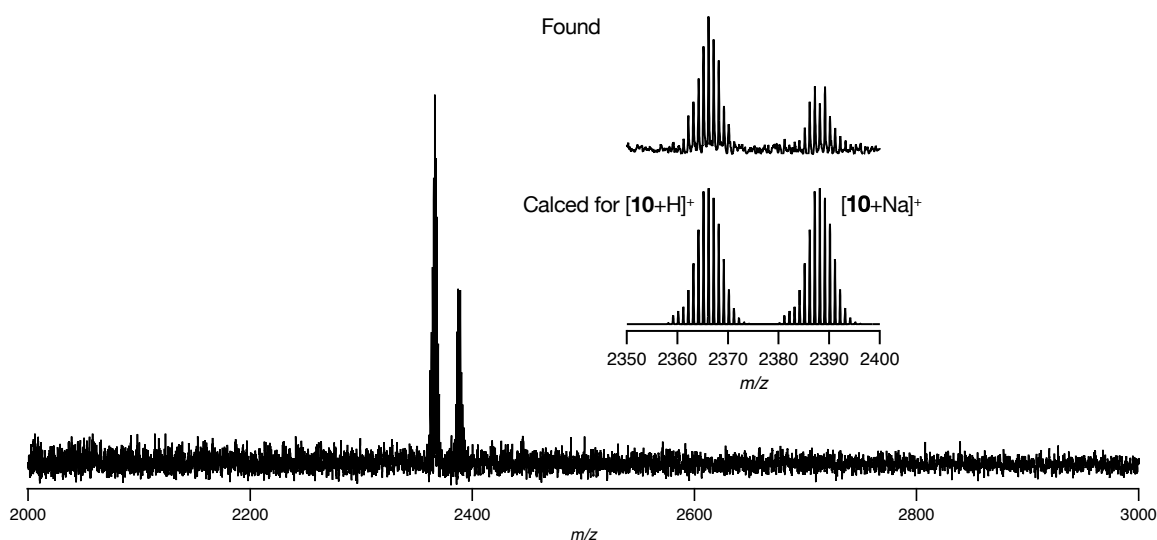


Figure S20 ESI-TOF MS spectrum of **10**

## 5. Fluorescence Microscopy

TIRF microscopic observation of the molecular motor **10** was performed using an inverted fluorescence microscope (Olympus IX 73, Tokyo, Japan) equipped with a 100x objective lens (NA = 1.49), and a diode laser for the excitation at 561 nm. Fluorescence images were recorded with an ORCA-Flash 4.0 CMOS camera (Hamamatsu Photonics, Shizuoka, Japan). Image acquisition and analysis was performed using a HImage software (Hamamatsu Photonics, Shizuoka, Japan).

To prepare the specimen for observation, 20  $\mu\text{L}$  of a highly diluted chloroform solution ( $7.42 \times 10^{-12}$  M) of molecular motor **10** was deposited onto a glass coverslip and then air-dried.

#### References Supporting Informations

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- [2] Thomas, J. R.; Liu, X.; Hergenrother, P. J. *J. Am. Chem. Soc.* **2005**, *127*, 12434.
- [3] Ma, C.; Bian, T.; Yang, S.; Liu, C.; Zhang, T.; Yang, J.; Li, Y.; Li, J.; Yang, R.; Tan, W. *Anal. Chem.* **2014**, *86*, 6508.
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- [5] Sirven, A. M.; Stefak, R.; Rapenne, G. *Heterocycl. Commun.* **2015**, *21*, 5.