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FULL PAPER

One Scaffold, Many Possibilities: CuAAC, SPAAC and Maleimide-Thiol Coupling of Ruthenium(II) Polypyridyl Complexes

Anne Stumper, Martin Lämmle, Alexander K. Mengele, Dieter Sorsche and Sven Rau*

Abstract: The applicability of Ru(II) polypyridyl complexes with appropriate functionalities as substrates for biorthogonal coupling reactions is investigated. In detail, copper(I)-catalyzed azide-alkyne cyclo-additions (CuAAC), strain-promoted azide-alkyne cycloadditions (SPAAC), and maleimide-thiol coupling of ruthenium complexes are examined. The first examples of SPAAC where the organic azide is provided by the metal complex are presented. All chromophores belong to one easy-to-synthesize scaffold, which has proven to be convenient for applications of metal chromophores. The fundamental photophysical properties of the examined compounds do not change with substitution, which is important for the design of chromophore conjugates. Furthermore, limitations of CuAAC reactions will be discussed with regard to copper impurities in the products formed.

Introduction

Providing possibilities to link functional molecular units is essential for the generation of sophisticated supramolecular architectures. Biological applications in particular are limited by the choice of linkage given that bioorthogonal conditions have to be applied. Click chemistry approaches such as the copper(I)-catalyzed azide-alkyne cycloaddition (CuAAC)[1], strain promoted azide-alkyne cycloaddition (SPAAC)[2] or maleimide-thiol coupling[3] represent advantageous strategies for linkage of sensitive (bio)molecules with functional units, e.g. photosensitizers or other imaging agents,[4–6] and control of the coupling is dependent on the corresponding functionalities. For photodynamic therapy (PDT), one feasible approach is to couple selectivity mediating biomolecules to photosensitizers like Ru(II) polypyridyl complexes.[7] Moreover, the choice of azide-alkyne cycloadditions provides the advantage that the reacting groups are not abundant in the body and therefore guarantees high selectivity and control. CuAAC has proven useful for the development of novel drug systems as well as many other applications.[1,8] Even though CuAAC is a chemically very selective coupling method, the employment of cell toxic copper ions for CuAAC reactions requires extensive purification and limits the exploitation of the CuAAC for live cell studies and for reactions where possible copper ion coordination has to be avoided. The use of the strain-activated cyclo-octyne motif has been shown to promote “click” chemistry without the need for a catalyst, and hence, SPAAC depicts a valuable bioorthogonal alternative. Commercially available cyclooctyne derivatives can be employed for the functionalization of biomolecules[9–12]. We hence focused our efforts on the development of ruthenium photosensitizers with stable pending azide groups to undergo SPAAC, which also provide the photophysical properties for a potential PDT application. The reaction of cyclooctynes with azides was observed already in 1967 and originates from the ring strain within cyclooctyne.[13,14] To this end, however, the number of Ru(II) chromophores with stable azide functionalities has been limited.[15] In order to prevent the formation of stereochemically complex structures with respect to the chiral nature of tris(bipyridine) ruthenium(II) complexes, we were further aiming for a substitution pattern that would preserve the C_2 symmetry of the parent complexes. (**Figure 1**). The introduction of dibenzocyclooctyne (DIBO) to Ru(II) and Ir(III) polypyridyl complexes[16,17] as well as SPAAC of an azide ligand on a Ru(II) core has recently been described in literature.[18] To the best of our knowledge, no SPAAC of a Ru(II) polypyridyl complex offering an azide group has been reported to date.

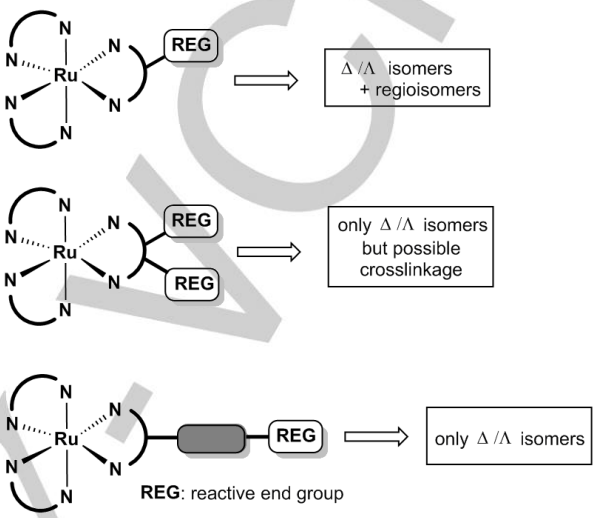

Figure 1. Importance of the design: structural details resulting in sets of possible isomers.

Additionally, our synthetic approach has proven convenient for the facile introduction of a maleimide functionality suitable for maleimide thiole “click” coupling. Among the amino acids, only cysteine provides a thiol (-SH) functionality. Although they are comparably less abundant in the body with respect to the other amino acids, proteins often present distinct cysteines on their accessible surface. Hence, maleimide-thiol coupling reactions[3] can be used to link Ru(II) chromophores to proteins. The cytotoxicity of several Ru(II)–arene complexes has been

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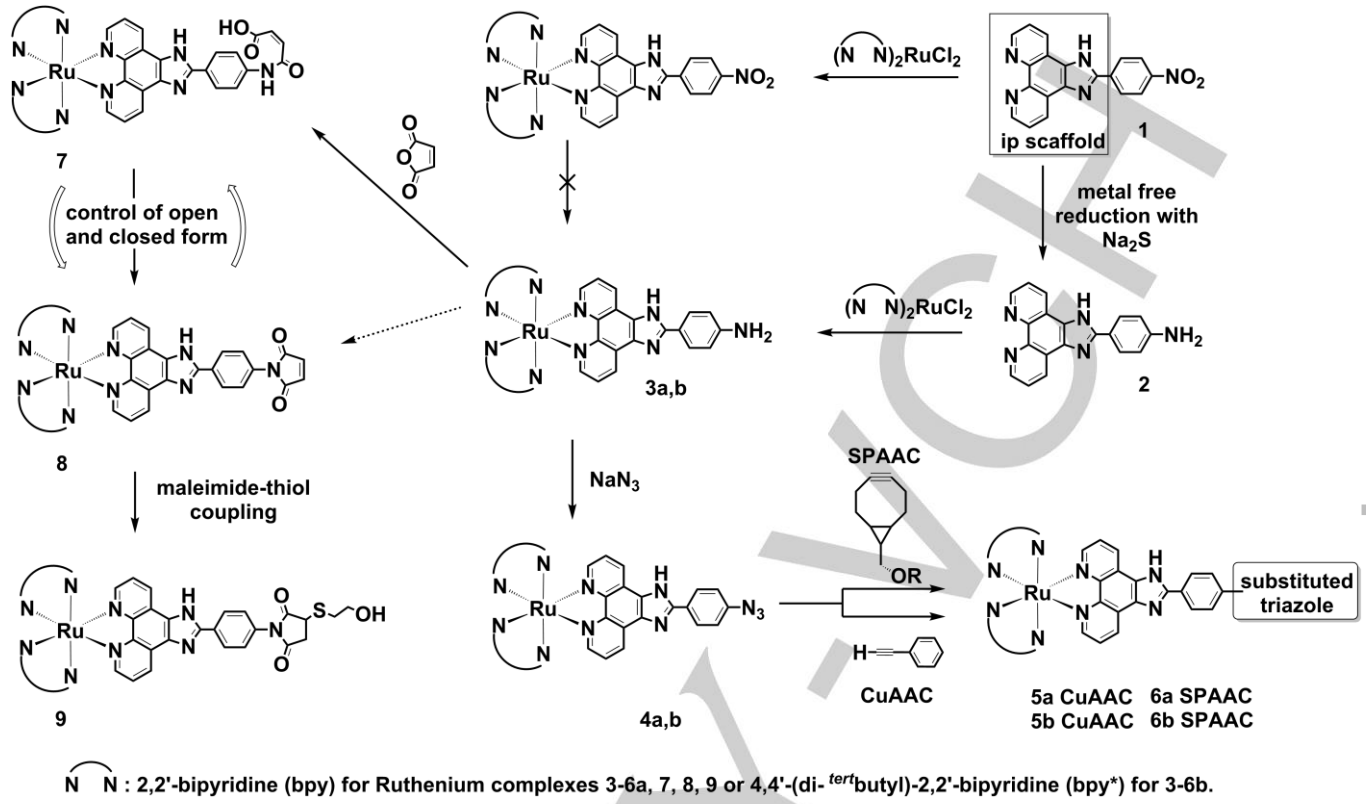

Figure 2. Overview of the examined compounds; conversion of the nitro functionality to the amine on the chromophore core was not possible; reaction of **3a** to yield **8** in a one-pot-synthesis was successful in 50% of the batches; the conversion from **7** to **8** and vice versa can be controlled easily.

previously examined.[19,20] Ru(II) and Ir(III) terpyridine complexes were coupled to cytochrome c[21] and a Ru(II) polypyridine chromophore was linked to different proteins via maleimide-thiol coupling.[22] Hence, providing a scaffold which is easily accessible and undergoes a variety of reactions which can be employed for linking biomolecules to chromophores is desirable. As a result of our ongoing efforts to provide versatile new ligand frameworks for the straightforward linkage of photoredox-active ruthenium chromophores with functional biomolecules, we herein report the development of functionalized imidazophenanthroline (**ip**) scaffolds (Error! Reference source not found.), all resorting to the easily accessible and versatile amine derivatives **3a,b**. We show CuAAC, SPAAC and maleimide-thiol reactions under biological conditions as well as in organic solvents. Desirable photophysical properties of the "click" precursors are shown to be retained upon functionalization which is essential for the establishment of a library of "clickable" chromophores.

Results and Discussion

In order to ensure subsequent applicability, it is necessary to devise strategies where the linking step, i.e. formation of the triazole, does not alter the photochemical properties significantly. The choice of the **ip**-phenyl ligand sphere preserves desired properties as already described in literature covering the creation of highly potent drug candidates and imaging agents.[4,7,23] Intrinsic symmetry of the **ip**-phenyl ligand limits the number of possible isomers to Δ/Λ-forms, whereas ligands showing lower symmetry (e.g. 4-R-bpy type ligands) would yield additional regio-isomers (**Figure 1**).

Synthesis of amine and azide precursor chromophores

Amine functionalized complex **3a**, the corresponding azide **4a** and the performance of the CuAAC to yield **5a** have previously been reported.[24] However, we modified the syntheses and present

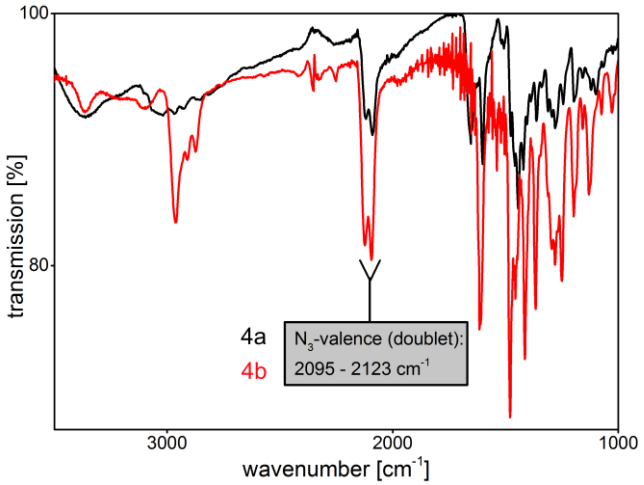

Figure 3. ATR-IR spectra of the azide species **4a** and **4b** with the typical N_3-valence doublet.

here the lipophilic analogues **3b**, **4b** and **5b** which feature additional *tert*-butyl groups on the peripheral bipyridines. First, ligand **2** is needed to generate the corresponding metal complexes. In order to avoid possible metal contamination, we abstained from using Pd/C for the reduction of the nitro group (**1**, crystal data in ESI) to the amine and used Na_2S as a mild, fast and cheap reduction agent with improved yield. We also tested the direct reduction of the nitro functionality on the chromophore implementing the same conditions but no conversion was observable. Presumably, Na_2S is not strong enough in redox potential to reduce the electron withdrawing nitro species on the cationic chromophore core. Complexation reactions to obtain **3a** (92%) and **3b** (89%) were carried out in microwave reactions following previously presented instructions using the corresponding Ru(II) precursor ($[Ru(4,4'\text{-R-bpy})_2Cl_2]$, R=H or ^{t}bu).[7] Azides **4a** (97%) and **4b** (84%) were obtained in excellent yields *via* diazotisation with $NaNO_2$ and following addition of NaN_3. Both azides show the typical doublet N_3-valence vibration band for phenyl azides[25] at 2095-2123 cm^{-1} in the IR spectra (**Figure 3**). All compounds were fully characterized *via* NMR spectroscopic techniques (^{1}H, ^{13}C (UDEFT), HMBC) as well as HRMS. The fundamental photophysical properties were determined via UV/Vis and emission spectroscopy. **3a** is very well soluble in water (153 mg/mL) but under addition of n-octanol to the aqueous solution and shaking at 25°C over night to determine partition coefficients, for some samples there was a decoloring of the aqueous phase and a thin solid film on the glass wall of the vessel was observed. Henceforth, no determination of partition coefficients was possible. The solid-state structure of **3b** shows the two-fold positively charged complex as chloride salt. All distances and angles regarding the coordination chemistry of the tris-(bipyridine) ruthenium(II) fragment are in the expected range. The anions both appear to establish hydrogen bonds with the **ip** ligand (red in **Figure** 4Error! Reference source not found.). The hydrogen bond between the imidazole amine function and one of the chloride ions is rather short with $d_{(N\cdots Cl)}$= 3.127(3) Å, and is supported by additional short contact interactions with the neighboring C-H groups of the phenanthroline and phenyl moieties (black in **Figure 4**, $d_{(phen\text{-}C\text{-}H\cdots Cl)}$ = 3.707 Å, $d_{(ph\text{-}C\text{-}H\cdots Cl)}$ = 3.634 Å). The **ip** ligand hence also appears completely planar, showing no gauche-type repulsion between the ip and phenyl moieties. This observation points to a surprisingly dominant H-bond interaction as related ruthenium complexes show torsion angles between 14° and 38.5°.[26] Additional hydrogen bond interactions are established between the terminal amine function and the second counter ion, with a somewhat longer N···Cl distance of 3.181(4) Å. This observation in the solid state suggests that the multiple H-bond sites available in this particular complex allow the formation of a neutral ion pair with increased lipophilicity. This results in an overall charge of 0 for the supramolecular assembly. Lipophilicity experiments were performed with **3a** and **3b** (respectively Cl-salts). Thereby no partition coefficients could be obtained under commonly applied experimental prodceedings.[27] For **3a** no partition could be obtained, as the compound remained completely within the aqueous phase or did show the before-mentioned formation of a solid. In contrast, **3b** was completely migrating into the organic phase, so that a highly significant effect of the *tert*-butyl substitution can be ascertained. This effect is highly relevant for the prospective design of molecular systems within a biological environment as the solubility properties determine the latter cellular uptake and accumulation characteristics.[28]

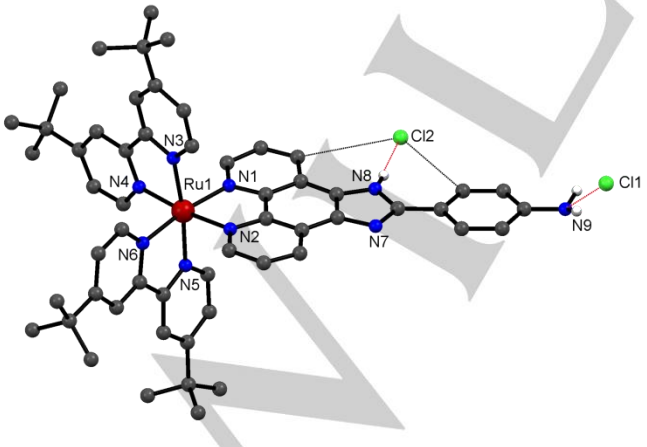

Figure 4. Mercury ball and stick representation of the molecular structure of **3b**, hydrogen bonds between the NH functionalities and the chloride counter ions are depicted in red, supporting short contacts to CH moieties are depicted in black, most hydrogen atoms were omitted for clarity.

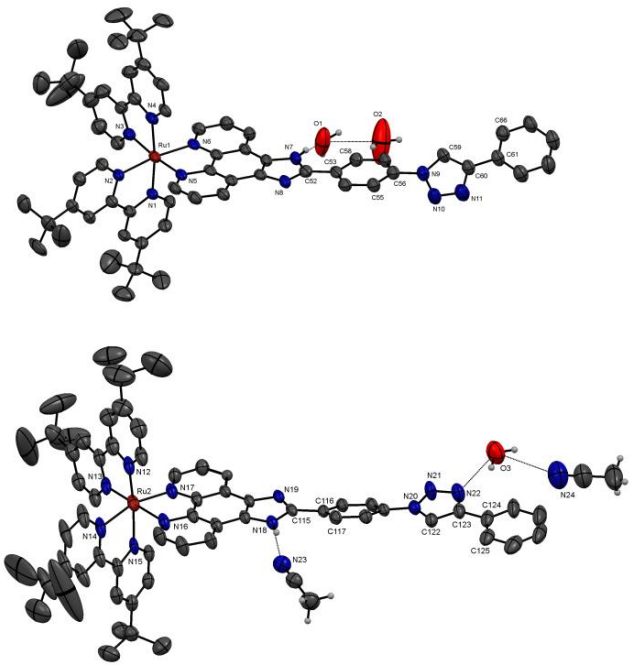

Figure 5. Solid-state structure of the Ru complex **5b** (ellipsoids are drawn at 50% probability level, hydrogen atoms and counter ions were omitted for clarity); **top**: complex **5b** in a hydrogen bonding network with two water molecules, **bottom:** complex **5b** interacting with two acetonitrile and one water molecule.

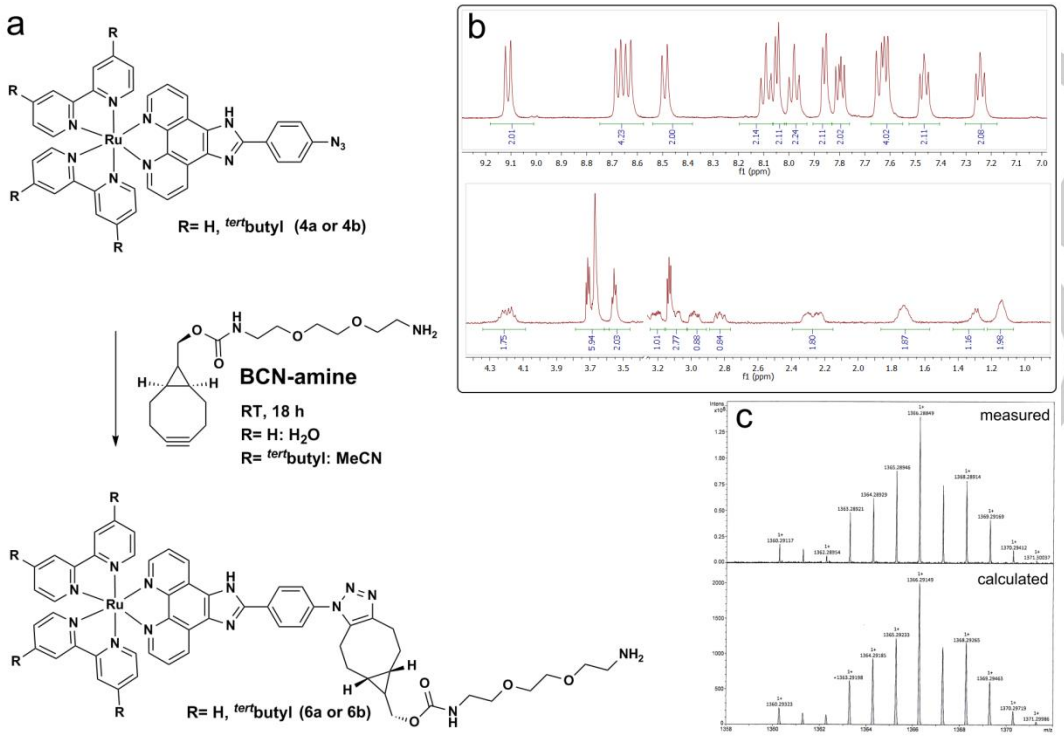

Figure 6. (a) Synthesis scheme for the SPAAC reactions of **4a** and **4b** with BCN-amine; (b) ^{1}H-NMR spectrum of **6a** (Cl-salt) in MeOH-d_4, aromatic region on the top and signals of aliphatic protons on the bottom, solvent signal was cut out for clarity (see **Figure S10** for full spectrum); (c) HRMS (MALDI-FT-ICR) signal for **6a** (PF_6-salt) with calculated isotopic pattern matching for the molecular ion $[M+H^+]^+$.

Copper (I) Catalyzed Azide Alkyne Cycloaddition (CuAAC) and lipophilicity experiments

The CuAAC protocol is a powerful method to link different moieties with each other to generate sophisticated molecular systems. Conversion in CuAAC reactions with model compound phenylacetylene to obtain **5a** and **5b** was performed with $CuSO_4$ 5 H_2O and sodium ascorbate. Formation of the product is readily observed *via* ^{1}H-NMR spectroscopy due to formation of a typical singlet for the triazole proton (see **Figure S3** and **S4** in ESI). All compounds were further characterized *via* HRMS (ESI/MALDI). Crystals of **1** (see supporting information) and **5b** (PF_6 salt) suitable for XRD were obtained by slow evaporation of the solutions in DMSO and MeCN, respectively. The solid-state structures in **Figure 5** show the two-fold positively charged Ru complex **5b** exhibiting the 1,4-regioisomeric triazole group. Within the asymmetric unit two different hydrogen bond motifs are found. As depicted in **Figure 5** (top), a hydrogen bonding pattern between the N-H moiety of the **ip** sphere and two mutually linked water molecules is established. This interaction leads to a planarization of the **ip** framework with the phenyl moieties as reflected by the torsion angle of 1.78° (N7-C52-C53-C58). The triazole ring, as well as the terminal phenyl group, share one common plane as the respective torsion angle (C59-C60-C61-C66) is as small as 1.56°. Interestingly, the above described two planarized spheres of the extended ligand are distorted to each other as the torsion angle C55-C56-N9-N10 exhibits a value of 34.81°. The second hydrogen bonding network found in the asymmetric unit **Figure 5**, bottom) consists of one acetonitrile molecule bound via the N-H group of the **ip** ligand and one water molecule, acting as a bridge between N22 of the triazole ring and a second acetonitrile molecule. Contrary to the structure discussed above, a larger torsion angle of 18.92° (N18-C115-C116-C117) was found between the **ip** sphere and the central phenyl ring, which is clearly caused by the different hydrogen bond framework. This is further corroborated by the fact that the **ip** sphere as well as the triazole and the terminal phenyl ring are virtually planar (torsion angle for N18-C115-N20-C122 is 5.41° and 1.34° for C122-C123-C124-C125) whereas only the central phenyl ring tilts out of this plane. Slow evaporation of a MeOH solution of **3b** yielded suitable crystals for XRD. Only one H-bond interaction with a group of two water molecules at the N-H moiety of the **ip** ligand was observed, which points to strong contribution of hydrogen bonding patterns with regard to the formation of different rotamers within one crystal structure. For both species (**3b**, **5b**) the triazole and terminal phenyl ring are coplanar, which might point to a relatively strong conjugative coupling between these moieties, overcompensating the repulsive gauche-type interactions of the relevant H atoms.

For multiple reproductions of the CuAAC reactions a contamination with copper ions for different batches was observed via HRMS (see **Figure S9** in ESI). With regard to the known cytotoxicity of copper ions this limits a potential application of CuAAC derivatives of **4a,b** for biological applications.[29] In general, the yields for the CuAAC reaction are higher, when utilizing stoichiometric equivalents of copper (2-4 eq), but copper species were also detected when the amount of copper was reduced (0.15 eq) with a loss in yield revealing unreacted educt species. Some batches were still found to be contaminated with copper ions after extensive purification. Pure batches were used for further reactions and characterization. For batches showing copper-containing fragments regarding mass spectrometry an interesting, intensive absorbance band around 360 nm was observed utilizing UV/Vis spectroscopy (see **Figure S8** in ESI).

For these batches the common use of extraction with Na-ETDA solution did not succeed. In future, precipitation with CN^--ions could be exploited. The corresponding ^{1}H-NMR spectra were interpretable which leads to the assumption that copper(I) ions are the contaminant as Cu(II) would lead to a significant broadening for the ^{1}H-NMR signals due to its paramagnetism. Regarding the contamination with metal ions, the choice of CuAAC should be deliberated on. We assume the formation of a Ruthenium(II)-Copper(I)-multinuclear species, where the copper ion might coordinate triazole or imidazo-units. It is known that copper(I) species similar in structure to our assumed species absorb within the relevant wavelength region with significant absorption coefficients.[30,31] These observations have to be examined in more detail and are subject of ongoing studies.

Strain-promoted Azide Alkyne Cycloaddition (SPAAC)

Based on these observations we investigated alternative, metal-free coupling concepts, and we present herein the first examples for SPAAC reactions with Ru(II) polypyridyl complexes where the azide functionality is provided by the chromophore. In order to avoid the necessity of a metal-based catalyst, we utilized the commercially available, strained cyclooctyne BCN-amine. A further advantage of BCN is that its C_s symmetry prevents the formation of complex regioisomers when linked to the chiral chromophore precursors.[32] Additionally, BCN-amine depicts low lipophilicity compared to other cyclooctyne derivatives. As the application of SPAAC reactions is of interest for the generation of molecular systems in general we employed two different Ru(II) polypyridine complexes providing different solubility characteristics. The SPAAC reaction to yield **6a** was carried out stirring **4a** and BCN-amine in water for 18 h. For **6b** a mixture of MeCN and water was employed with respect to its increased lipophilicity (**Figure 6**). Both SPAAC products were purified *via* size exclusion chromatography and characterized *via* NMR spectroscopy and HRMS. High resolution mass spectrometry revealed expected signals for $[M+H^+]^+$ and $[M–2Cl^-–H^+]^+$ species showing the typical isotopic pattern for ruthenium (**Figure S14**, ESI). Screening of the reactions *via* NMR suggested quantitative formation, however only moderate yields of the pure products were obtained (27-40%) after size exclusion chromatography due to the small scale these reactions have been carried out in. However, to the best of our knowledge, no yields have been provided for other SPAAC reactions carried out with Ru(II) chromophores in the literature[16]. A related Ir(III) based system was reported *via* recrystallization[17].

Maleimide-Thiol Coupling

Displaying another metal-free click alternative we envisaged the maleimide sulfide coupling reaction, since the respective chromophore precursor **8** is readily accessible from **3a**. The amine functionality was converted to the open maleimide **7** using maleic anhydride in a room temperature condensation reaction providing quantitative yields. Successful reaction conditions were modified according to literature which dealt with organic maleimides.[33] Surprisingly, when screening different reaction conditions it was observed that reactions in a one-pot microwave

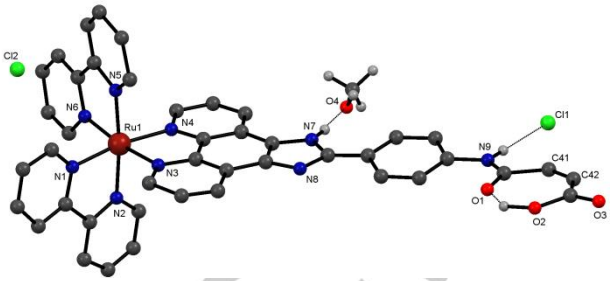

Figure 7. Structural motif of **7** (ball-and-stick representation; most hydrogen atoms were omitted for clarity).

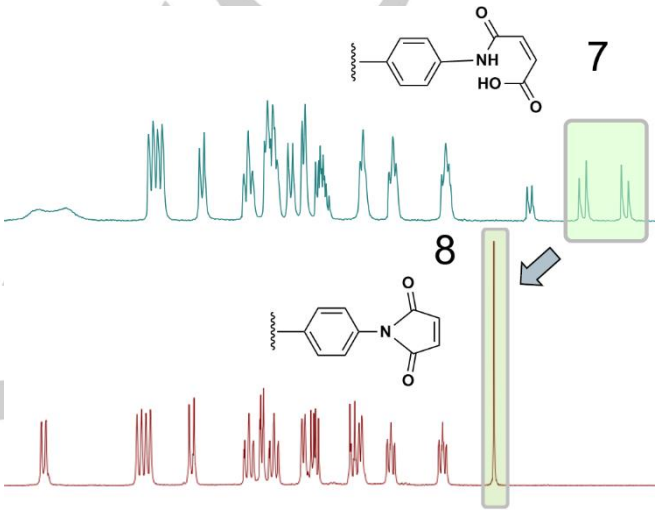

Figure 8. ^{1}H-NMR spectra of open (**a, 7**) and closed (**b, 8**) form of the maleimide in MeCN-d_3; signals at the double bond ease characterization of the ring-closing reaction via NMR spectroscopy.

set up[34] yield the desired closed maleimide either in excellent yields or not at all. Therefore, the two-step way was preferred later-on as it works reliably. The ring-closing reaction to obtain **8** was screened with modified literature conditions[33] (e.g. elongated reaction time for complete conversion) of **7** to **8**. It was determined, that the ring-closing reaction is completed after three hours and the most efficient way of work up is the direct precipitation from the reaction mixture as PF_6 salt. More elongated reaction times will lead to ring-opening due to the released water. Compounds **7** and **8** were characterized in detail *via* NMR and IR spectroscopic techniques, and HRMS. The ^{1}H-NMR spectrum provides direct information about the nature of the substitution, as it reveals either two doublets or a singlet for the open and closed form, respectively. The IR spectrum of **8** depicts a typical strong C=O vibration band at 1716 cm^{-1}. It is known, that maleimide functionalities are reactive in protic environments, leading to ring-opening reactions. We observed decomposition of about 40% within a day in methanol solution, which precluded chromatographic purification for **8** in protic solvents. The ring-closing reaction can be repeated when decomposition is observed and **8** is stable (weeks) in non-protic, dry solvents (e.g. acetonitrile). Therefore, recrystallization protocols were developed to yield **8** in sufficient purity for the next reaction.

Single crystals of **7** suitable for X-ray diffractometry were obtained via slow evaporation out of a MeOH solution of **8** as chloride salt. The structural motif depicted in **Figure 7** shows the complex **7** with a hydrolytic cleavage of the maleimide functionality forming a ring-opened, hydrogen bonding stabilized carboxylic acid moiety. All distances and angles regarding the coordination chemistry of the *pseudo*-octahedral Ru-moiety are within the expected range. The torsion angle between the **ip** motif and the bridging phenyl moiety exhibits a very small value of 6.7° indicating only minor gauche type interactions. Moreover, the torsion angles in the ring-opened maleimide part cover rather small values between 1.2° and 7.5° pointing to a fully conjugated system, which is further supported by the short C-C distance between C41 and C42 (1.276 Å) indicative of a substantial double bond character. It is apparent from **Figure 7** that H-bonding interactions between the solvent and the N-H functionality of the ip-ligand occur, whereas one of the two chloride counter ions interacts with the hydrolytically generated amide N-H group of the former maleimide moiety.

To verify the potential of this functionality to serve as agent for the

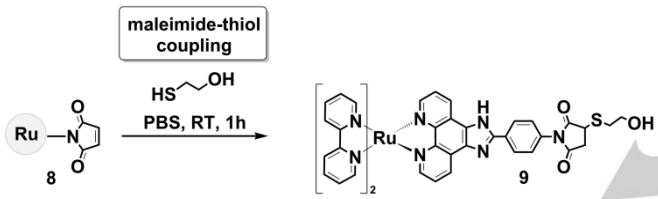

Figure 10. Reaction of **8** with 2-mercapto-ethanol to yield the coupling product **9**; the reaction can be carried out in pure PBS or in PBS acetonitrile mixtures.

functionalization of proteins, we converted **8** with 2-mercapto-ethanol as model thiol (**Figure** 9). Educt **8** was reprecipitated as chloride salt for the reaction. The reaction was carried out at room temperature in aqueous PBS buffer (pH=7.4). Upon addition of the thiol to the PBS solution of **8** a darkening of the red solution was observable and the reaction was completed after one hour, indicated by the naked eye with a slight brightening of the red color and as confirmed *via* NMR spectroscopy. The product was purified via size exclusion chromatography and the conversion was confirmed via NMR spectroscopy and HRMS. The reaction can also be carried out in a PBS/acetonitrile mixture (1:1) utilizing the PF_6 salt of **8**.

Photophysical Characterization

For the straightforward introduction of chromophores into clicked systems it is generally of paramount importance that such click-motifs retain the photophysical properties of the former. Steady state absorption and emission properties in aqueous solution were determined for the ruthenium complexes and are summarized in **Table 1**. As can be concluded with respect to the virtually identical absorption and emission profiles, the **ip** scaffold is indeed capable of preserving the photophysical properties of its precursor chromophores upon click-substitution using either CuAAC, SPAAC, or maleimide sulfide coupling, as no significant shifts of LC and ^{1}MLCT transitions can be observed. The preliminary data show that the UV/Vis and emission

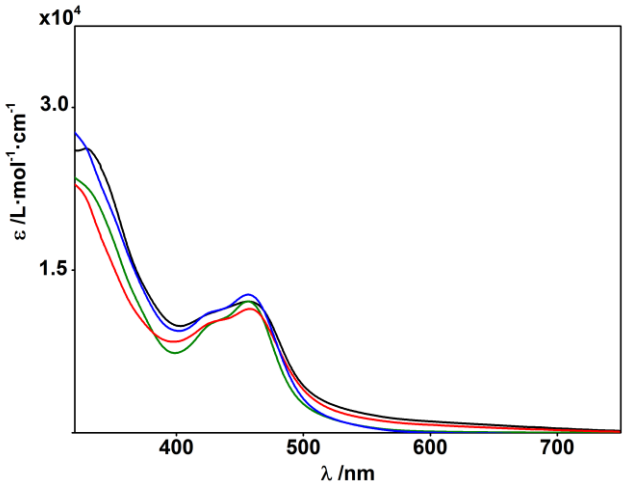

Figure 9. Exemplary absorbance of the series of Ru complexes **3a**, **4a**, **5a** and **6a** in MilliQ water.

spectroscopic properties of the [(bpy)$_2$Ru(**ip**-phenyl)]-type complexes are not influenced by the substitution of the peripheral phenyl moiety.

Table 1. Absorption and emission maxima in aqueous solution at concentrations of about c=10^{-5} mol/L.

compounds	LC $\lambda_{max,abs}$ [nm]	^{1}MLCT $\lambda_{max,abs}$ [nm]	$\lambda_{max,em}$ [nm]
3a	287	460	603.5
4a	285.5	458.5	603
5a	286	459.5	603.5
6a	285	458	604
3b	287.5	465.5	621
4b	285.5	461	622
5b	287	467.5	616.5
6b	285	463	624
7	286	460	605
8	286	462	604
9	285	460	604

Conclusions

We present a series of Ru(II)polypyridyl complexes which can undergo a variety of click reactions such as CuAAC, SPACC and maleimide-thiol coupling. The utilized precursor compounds all resort to the easy-to-synthesize compounds **3a** and **3b**. We here present the first examples for SPAAC reactions where the organic azide is provided by the metal chromophore. All compounds have been characterized in detail and their fundamental photophysical properties were determined. The introduced stable azide functionalities can be converted in cycloaddition reactions under biorthogonal (CuAAC, SPAAC) or highly selective conditions (maleimide-thiol coupling). Therefore, these complexes may be employed for the functionalization of biomolecules enabling various applications. Additionally, the reactions can also be carried out in organic solvents which is important for the establishment of molecular systems for other applications beyond a biological background. We want to point out, that this scaffold offers a variety of possible functionalizations without change in

essential photophysical properties which can be exploited for the prediction of characteristics of established conjugates. The carry-off of copper ions is obviously a problem for the use of CuAAC reactions with regard to biological applications, especially as the contamination was varying from batch to batch. Therefore it is highly interesting to provide efficient linking strategies for metal complex conjugates avoiding contaminants like copper ions.

Experimental Section

2-(4-nitrophenyl)-1H-imidazo[4,5-f][1,10]-phenanthroline (**1**) was prepared according to literature.[35] Other compounds were synthesized utilizing optimized or alternate protocols presented below.

Ligand Synthesis

2-(4-nitrophenyl)-1H-imidazo[4,5-f][1,10]-phenanthroline (1): 4-Nitrobenzaldehyde (1.25 g, 8.27 mmol) and phenanthroline-5,6-dione (1.68 g, 8.00 mmol) were suspended in acetic acid (100%, 50 mL). Ammonium acetate (12 g) was added and the suspension was heated to 85°C to yield a yellow solution. Upon heating to 120°C a thick yellow suspension was observed which was refluxed for 2 h. The precipitate was filtered and washed with water to yield **1** (95%, 2.59 g, 7.6 mmol) as a bright yellow solid. ^{1}H NMR (400.13 MHz, DMSO-d_6): δ= 9.53 (dd, *J* = 4.3, 1.8 Hz, 2H), 9.43 (dd, *J* = 8.1, 1.7 Hz, 2H), 9.05 (d, *J* = 9.1 Hz, 2H), 8.97–8.94 (d, 2H), 8.31 (dd, *J* = 8.1, 4.3 Hz, 2H) ppm. Crystal data for **1**: C_{21} H_{17} Cl N_5 O_3 S, M_r = 419.47 g mol^{-1}, white block, monoclinic, space group $P2_1/n$, a = 7.9969(2) Å, b = 16.9498(4) Å, c = 14.3417(4) Å, α = 90°, β= 90.503(2)°, γ = 90°, V = 1917.28(9) Å^3, T = 150 K, Z = 4, $\rho_{calcd.}$ = 1.453 Mg/m^3, μ (Cu-K$_\alpha$) = 1.804 mm^{-1}, F(000) = 872.0, altogether 9268 reflexes up to h(-9/9), k(-11/21), l(-17/16) measured in the range of 15.346° ≤ Θ ≤ 148.978°, completeness Θ_{max} = 99.7 %, 3884 independent reflections, R_{int} = 0.0220, 273 parameters, 0 restraints, $R1_{obs}$ = 0.0429, $wR2_{obs}$ = 0.1183, $R1_{all}$ = 0.0458, $wR2_{all}$ = 0.1218, GOOF = 1.059, largest difference peak and hole: 0.79/-0.46 e·Å^{-3}. CCDC 1573110 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

4-(1H-imidazo[4,5-f][1,10]phenanthroline-2-yl)-phenyl-1-aniline (2): 2-(4-nitrophenyl)-1H-imidazo-[4,5-f]-[1,10]-phenanthroline (3.2 g, 9.38 mmol) was suspended in 1,4-dioxane (76 mL) and heated to 80°C. Na_2S was dissolved in water and heated to 80 °C. The warm Na_2S solution was combined with the warm yellow suspension and heated to 80 °C for 4 h. The color changed instantaneously from orange to red and the solid was dissolved completely. The dioxane was evaporated under precipitation of the product. The solid was filtered off and washed respectively with water and Et_2O. After drying the product was received as ochre colored solid in a yield of 78% (2.3 g, 7.29 mmol). ^{1}H NMR (400.13 MHz, RT, DMSO-d_6): δ = 8.90 (dd, 2 H), 8.87 (d, 2H), 8.01 (d, 2 H), 7.73 (dd, 2 H), 6.68 (d, 2H), 5.42 (s, 2 H) ppm.

Ruthenium Complexes without tert-butyl groups

[Bis(2,2'-bipyridine)-(4-(1*H*-imidazo[4,5-f][1,10]phenanthroline-2-yl)-phenyl-1-aniline)-Ruthenium-(II)]Cl_2 (3a): 300 mg (620 μmol) [$(bpy)_2RuCl_2$] and 232 mg (746 mmol) of **2** were dissolved in EtOH (15 mL), H_2O (5 mL) and 3 drops of an aqueous KOH solution (1 plate dissolved in 15 mL H_2O) were added. The solution was reacted in the microwave for 2h at 180 W and changed the color from dark violet to red. The solvents were rotary evaporated, the residue was dissolved in EtOH, Et_2O was added and red crystals precipitated. The red crystals were filtered off. Via size exclusion chromatography (Sephadex©) in MeOH the desired complex was isolated as a bright orange band. After the solvent was removed via rotary evaporation the product was isolated as red crystals (92%, 414.5 mg, 571 μmol). ^{1}H NMR (400.13 MHz, RT, MeOD-d_4): δ = 9.12 (d, *J* = 7.6 Hz, 2H), 8.71 (dd, *J* = 16.5, 7.9 Hz, 4H), 8.25 – 8.12 (m, 2H), 8.11 – 8.00 (m, 6H), 7.94 (ddd, *J* = 5.6, 1.4, 0.7 Hz, 2H), 7.85 (dd, *J* = 8.4, 5.3 Hz, 2H), 7.69–7.63 (m, 2H), 7.54 (ddd, *J* = 7.6, 5 .6, 1.3 Hz, 2H), 7.31 (ddd, *J* = 7.6, 5.7, 1.3 Hz, 2H), 6.90 – 6.81 (m, 2H) ppm. ^{13}C-NMR (101 MHz, RT, MeOD-d_4): δ = 157.55, 157.38, 152.01, 150.66, 146.14, 138.15, 138.02, 134.87, 130.68, 127.85, 127.73, 127.42, 127.21, 126.37, 124.55, 124.46 ppm. HRMS (MALDI-FT-ICR): 724.15194 [M-H$^+$-2Cl$^-$]$^+$ (calculated: 724.15057).

[Bis(2,2-bipyridine)-(4-(1*H*-imidazo[4,5-f][1,10]phenanthroline-2-yl)-phenyl-1-azido)-Ruthenium(II)] Cl_2 (4a): 3a (200 mg, 275μmol) was dissolved in 10 mL H_2O, 10mL MeCN and 0.45 mL 3 M HCl. $NaNO_2$ (23 mg, 330μmol) dissolved in 0.45 mL H_2O was added and stirred for 10 min at RT, then the reaction mixture was cooled to 0°C with an ice bath and NaN_3 (36 mg, 550μmol) in 0.45 mL H_2O was added and stirred for further 10 min at 0°C. Then it was allowed to come to RT and stirred for 3h. The reaction mixture was neutralized with NH_3 (25% in water) and the solvents were removed under reduced pressure. The residue was dissolved in EtOH and the precipitated white salts were filtered off over a PTFE filter. Then the product was precipitated with Et_2O and n-hexane. The precipitate was filtered via a glass frit (POR5). Via size exclusion chromatography (Sephadex©) in MeOH the desired complex was isolated as a bright red band. After the solvent was removed via rotary evaporation the product was received as dark red-brown crystals in a yield of 97% (202 mg, 269 μmol). ^{1}H NMR (400.13 MHz, RT, MeOD-d_4): δ=9.10 (d, *J* = 8.4 Hz, 2H), 8.77 (dd, *J* = 14.2, 8.2 Hz, 4H), 8.28 – 8.15 (m, 4H), 8.15 – 8.05 (m, 4H), 7.97 (d, *J* = 5.7 Hz, 2H), 7.86 (dd, *J* = 8.3, 5.3 Hz, 2H), 7.76 (d, *J* = 5.5 Hz, 2H), 7.64 – 7.50 (m, 2H), 7.45 – 7.28 (m, 2H), 7.06 (d, *J* = 8.6 Hz, 2H) ppm. ^{13}C-NMR (101 MHz, RT, MeOD-d_4): δ = 158.80, 158.60, 152.85, 152.80, 151.24, 143.98, 139.33, 139.19, 132.05, 129.71, 128.96, 128.88, 127.32, 127.21, 125.69, 125.62, 120.86 ppm. HRMS (MALDI-FT-ICR): 750.14197 [M-H$^+$-2Cl$^-$]$^+$ (calculated: 750.14208). IR: ν =2095-2123 (s, doublet N_3-stretching) cm^{-1}.

[Bis(2,2-bipyridine)-(4-(1*H*-imidazo-4,5-f-1,10-phenanthroline-2-yl)-phenyl-1-(1-*H*-1,2,3-triazolyl-4-phenyl)-Ruthenium(II)]Cl_2 (5a): 57 mg (0.07mmol) of **4a** and 10.5 mg (11 μl, 0.1mmol) of phenylacetylene were dissolved in water. NaAsc (4 eq, 55 mg) and $CuSO_4$·5 H_2O (2 eq, 34 mg) were dissolved in 2 mL water respectively, added to the reaction mixture and stirred overnight at rt. The product was precipitated as PF_6 salt out of the aqueous solution and filtered off over a glass frit (POR4). Reprecipitation to the Cl salt with TBACl in acetone and purification via size exclusion chromatography (Sephadex©) in MeOH yielded the product as a bright red-orange band. After removal of the solvent, the product was isolated as red crystals in 95% yield (61 mg, 0.067 mmol). ^{1}H-NMR (MeCN-d_3, 400.13 MHz): δ=9.05 (d, *J*=8.5 Hz, 2H), 8.70 (s, 1H), 8.63 (d, *J*=8.4 Hz, 2H), 8.54 – 8.43 (m, 4H), 8.07 (s, 2H), 7.96 (d, *J*=9.9 Hz, 6H), 7.87 (s, 2H), 7.76 (s, 2H), 7.63 (s, 4H), 7.51 (s, 2H), 7.42 (d, *J*=6.6 Hz, 4H),

7.20 (s, 2H) ppm. ^{13}C-NMR (101 MHz, MeCN-d_3)=158.29, 158.00, 152.67, 148.82, 148.08, 145.44, 138.40, 138.23, 131.16, 129.92, 129.27, 128.66, 128.28, 127.62, 126.48, 125.64, 124.99, 124.89, 121.33, 119.96 ppm. HRMS (MALDI-FT-ICR): 852.18794 $[M\text{-}H^+\text{-}2Cl^-]^+$ (calculated: 852.18802).

[Bis(2,2-bipyridine)-(4-(1*H*-imidazo-4,5-f-1,10-phenanthroline-2-yl)-phenyl-1-(1*H*-1,2,3-triazolyl-4,5- ((1R,8S,Z)-bicyclo[6.1.0]non-4-en-9-yl)methyl (2-(2-(2-aminoethoxy)-ethoxy)-ethyl)-carbamate)-Ruthenium(II)]Cl_2 (6a): 20.58mg (25.05µmol) of **4a** were dissolved in a small amount of H_2O. 8.1 mg (24.968µmol) *N*- [(1*R*, 8*S*, 9*S*) - Bicyclo[6.1.0]-non-4-yn-9-yl-methyl-oxy-carbonyl]-1,8-diamino-3,6-dioxa-octane (BCN-amine) was dissolved in an ethanol/water mixture and added subsequently. A slight warming of the resulting solution was observed and it was stirred for 18 h at rt. 60.2 mg (369.33µmol) NH_4PF_6 dissolved in water were added to precipitate the product. The solid was filtered via a syringe filter (nylon), washed with water and dried (crude product yield: 85%). The product was dissolved from the filter with MeCN and the solvent was evaporated. Reprecipitation with TBACl yielded the Cl-salt of **6a** which was purified via size exclusion chromatography to remove the educt surplus. **6a** was isolated as red solid in a moderate yield (7.6 mg, 6.631 µmol, 26.6%).^{1}H-NMR (MeOD-d_4, 500.13 MHz): δ=9.21 (d, *J*= 8.3 Hz, 2H), 8.76 (dd, *J*=15.9, 8.3 Hz, 4H), 8.60 (d, *J*= 8.5 Hz, 2H), 8.14 (ddd, *J*= 32.6, 15.9, 8.0 Hz, 6H), 7.96 (d, *J*= 5.5 Hz, 2H), 7.90 (dd, *J*= 8.3, 5.3 Hz, 2H), 7.73 (dd, *J*= 11.4, 7.2 Hz, 4H), 7.61–7.52 (m, 2H), 7.40–7.29 (m, 2H), 4.27–4.08 (m, 2H), 3.70 (dd, *J*= 13.3, 8.2 Hz, 6H), 3.55 (t, *J*= 5.4 Hz, 2H), 3.26–3.16 (m, 1H), 3.16–3.05 (m, 3H), 3.03–2.92 (m, 1H), 2.89–2.77 (m, 1H), 2.36–2.18 (m, 2H), 1.81–1.63 (m, 2H), 1.39–1.23 (m, 1H), 1.14 (s, 2H) ppm.^{13}C-NMR (101 MHz, MeOD-d_4): δ=158.80, 158.61, 153.71, 153.28, 152.86, 152.79, 151.90, 151.42, 147.23, 146.62, 146.45, 139.34, 139.21, 136.74, 132.15, 129.25, 128.97, 128.88, 127.89, 127.41, 125.69, 125.62, 71.38, 71.31, 71.10, 67.88, 63.62, 63.39, 41.54, 40.71, 26.46, 24.54, 23.77, 23.38, 21.16, 20.72, 19.16 ppm. HRMS (MALDI-FT-ICR): 1366.28849 $[M+H^+]^+$ (calcd. 1366.28998), 1220.31673 $[M\text{–}PF_6^-]^+$(calcd. 1220.31798).

[Bis(2,2-bipyridine)-(4-(1*H*-imidazo[4,5-f][1,10]phenanthroline-2-yl)-phenyl-1-((*Z*)-4-(λ^2-azanyl)-4-oxobut-2-enoic acid))-Ruthenium(II)]$(PF_6)_2$ (open maleimide 7): 84 mg (0.08mmol) of **3a** (PF_6 salt) were dissolved in MeCN (5 mL) and maleic anhydride. 8 mg (10 eq, 0.80mmol) were added. The resulting solution was stirred at RT for 12 h. Product **7** was precipitated with diethylether from the reaction mixture, filtered off via a POR3 glass frit and washed intensively with diethylether. The reaction yielded **7** as a red solid (98%, 88 mg, 0.08 mmol). ^{1}H NMR (400.13 MHz, MeCN-d_3): δ=12.36 (s, 1H, NH_{ip}), 9.62 (s, 1H, NH_{amide}), 8.98 (d, *J* = 48.4 Hz, 2H), 8.52 (dd, *J* = 16.2, 8.2 Hz, 4H), 8.31 (d, *J* = 8.6 Hz, 2H), 8.10 (t, *J* = 7.9 Hz, 2H), 8.04 – 7.94 (m, 4H), 7.91 (d, *J* = 8.6 Hz, 2H), 7.85 (d, *J* = 5.4 Hz, 2H), 7.76 (ddd, *J* = 10.7, 8.3, 5.3 Hz, 4H), 7.59 (d, *J* = 5.1 Hz, 2H), 7.45 (t, *J* = 6.1 Hz, 2H), 7.21 (t, *J* = 6.5 Hz, 2H), 6.59 (d, *J* = 12.7 Hz, $1H_{openMI}$), 6.40 (d, *J* = 12.7 Hz, $1H_{openMI}$) ppm. HRMS (MALDI-FT-ICR): 3193.29621 $[3M\text{-}PF_6]^+$ (calcd. 3193.29054), 2081.21209 $[2M\text{–}PF_6]^+$(calcd. 2081.21066), 968.12270 $[M\text{–}PF_6]^+$(calcd. 968.12297), 822.15048 $[M\text{–}2PF_6\text{-}H^+]^+$(calcd. 822.15096). Crystal data for **7**: C_{44} H_{35} $Cl_{1.78}$ N_9 O_4 Ru, M_r = 918.16 g mol^{-1}, red block, orthorhombic, space group Pbcn, a = 28.3662(10) Å, b = 15.9805(4) Å, c = 20.9099(6) Å, α = 90°, □= 90.° γ = 90°, V = 9478.6(5) Å^3, T = 149.95(10) K, Z = 8, $\rho_{calcd.}$ = 1.287 Mg/m^3, µ (Cu-K_α) = 4.000 mm^{-1}, F(000) = 3747.0, altogether 37069 reflexes up to h(-31/34), k(-12/19), l(-23/25) measured in the range of 15.096° ≤ Θ ≤ 148.782°, completeness Θ_{max} = 97 %, 9375 independent reflections, R_{int} = 0.0398, 561 parameters, 3 restraints, $R1_{obs}$ = 0.1358, $wR2_{obs}$ = 0.3927, $R1_{all}$ = 0.1690, $wR2_{all}$ = 0.4224, GOOF = 1.540, largest difference peak and hole: 1.66/-0.74 e·Å^{-3}.

[Bis(2,2-bipyridine)-(4-(1*H*-imidazo[4,5-f][1,10]phenanthroline-2-yl)-phenyl-1-(1-λ^2-pyrrole-2,5-dione)-Ruthenium(II)]Cl_2 (closed maleimide 8):

a) One pot synthesis from 3a (working 50% of batches): 70 mg (0.07mmol) of **3a** (PF_6 salt) were dissolved in MeCN (1.5 mL) and 2 mL acetic anhydride. 8 mg (0.09mmol) of NaAc were added and the obtained solution was refluxed (MW, 450 W) for 30 min. **8** was isolated via precipitation from the reaction mixture with ether.

b) Ring-closing reaction from 7 (works reliably): 60 mg (0.05mmol) of **7** (PF_6 salt) were dissolved in MeCN (5 mL) and 6 mg (0.07mmol) of NaAc were added. The resulting solution was refluxed for 3 h and the product was precipitated with diethylether from the reaction mixture. To avoid decomposition the product was reprecipitated as Cl-salt and again reprecipitated to yield the PF_6 salt, purification via chromatography would have to be performed in non protic solvents to avoid a massive loss of yield due to ring-opening. As the product is sensitive towards protic solvents it should be dried intensively and should not be kept in protic solution for long time. The product was obtained as red solid (45 mg, 76%, 0.04 mmol). ^{1}H NMR (400.13 MHz, MeOD-d_4): δ=9.00 (d, *J* = 8.3 Hz, 2H), 8.64 (dd, *J* = 15.2, 8.2 Hz, 4H), 8.33 (d, *J* = 8.3 Hz, 2H), 8.17 (t, *J* = 7.9 Hz, 2H), 8.11 (d, *J* = 5.1 Hz, 2H), 8.06 (t, *J* = 7.9 Hz, 2H), 7.93 (d, *J* = 5.4 Hz, 2H), 7.83 (dd, *J* = 8.2, 5.1 Hz, 2H), 7.67 (dd, *J* = 15.4, 6.9 Hz, 4H), 7.56 – 7.50 (m, 2H), 7.36 – 7.25 (m, 2H), 7.05 (s, $2H_{MI}$) ppm. ^{13}C NMR (101 MHz, MeOD-d_4): δ= 170.10, 157.55, 157.38, 152.53, 152.01, 150.66, 146.14, 138.15, 138.02, 134.87, 134.26, 130.68, 127.85, 127.73, 127.42, 127.21, 126.37, 124.55, 124.46, 123.94 ppm. IR(KBr): 1716 (C=O-valence) cm^{-1}. HRMS (MALDI-FT-ICR): 2045.2001 $[2M\text{-}PF_6]^+$ (calculated: 2045.18953), 950.11248 $[M\text{-}PF_6]^+$ (calculated: 950.11355), 804.14309 $[M\text{-}2PF_6\text{-}H]^+$ (calculated: 804.14040).

Bis(2,2-bipyridine)-(4-(1*H*-imidazo[4,5-f][1,10]phenanthroline-2-yl)-phenyl-1-(3-((2-hydroxyethyl)thio)-1-λ^2-pyrrolidine-2,5-dione))-Ruthenium (II)] Cl_2 (9): 8 (30 mg, 0.03mmol) was dissolved in PBS (1.5 mL) and 2-mercapto-ethanol (3 µL, 0.04mmol) was added whereupon dark red streaks appeared in the solution. The mixture was stirred at RT for 1 h during which the red streaks vanished. The product was precipitated with NH4PF6, filtered via a glass frit (POR5) and washed with diethylether. It was reprecipitated to the Cl-salt with TBACl and the product was purified twice by size exclusion chromatography (Sephadex©, MeOH) to yield **9** as red solid (74%, 0.02mmol, 21 mg). ^{1}H NMR (400.13 MHz, MeCN-d_3): δ=12.48 (s, 1H), 8.97 (s, 2H), 8.52 (dd, J = 16.2, 8.2 Hz, 4H), 8.37 (dd, J = 8.3, 5.6 Hz, 1H), 8.21 (dd, J = 8.9, 2.1 Hz, 1H), 8.10 (t, J = 7.9 Hz, 2H), 8.05 – 7.96 (m, 4H), 7.92 (s, 1H), 7.89 – 7.73 (m, 5H), 7.59 (t, J = 5.0 Hz, 2H), 7.52 (d, J = 8.5 Hz, 1H), 7.45 (dd, J = 7.4, 6.1 Hz, 2H), 7.25 – 7.16 (m, 2H), 2.89 (m, 3H), 2.77 (m, 2H), 1.32 (m, 2H). HRMS (MALDI-FT-ICR): 1028.12559 $[M\text{-}PF_6]^+$ (calculated: 1028.12747). This reaction can also be carried out with the PF_6 salt of **8** in a MeCN/PBS (1:1) mixture.

Ruthenium Complexes bearing *tert*-butyl groups

[Bis(4,4'-di-*tert*-butyl-2,2'-bipyridine)-4-(1*H*-imidazo[4,5-f]-[1,10]-phenanthroline-2-yl)aniline-Ruthenium(II)]Cl_2 (3b): 454 mg (640 µmol) $[(bpy^*)_2RuCl_2]$ and 283 mg (833 µmol) of **2** were dissolved in EtOH/H_2O (20 mL, 3:1) and 3 drops of an aqueous KOH solution (1 plate dissolved in 15 mL H_2O) were added. The solution was reacted in the microwave for

2h at 180 W and changed the color from dark violet to red. The solvents were rotary evaporated, the residue was dissolved in EtOH, Et_2O was added and red crystals precipitated. The red crystals were filtered off. Via size exclusion chromatography (Sephadex©) in MeOH the desired complex was isolated as a bright orange band. After the solvent was removed via rotary evaporation the product was isolated as red crystals (89%, 590.7 mg, 570 µmol). ^{1}H NMR (400.13 MHz, RT, MeCN-d_3): δ = 9.03 (d, *J* = 8.2 Hz, 2H), 8.51 (m,6), 8.30 (d, *J* = 8.8 Hz, 2H), 7.92-7.82 (m, 6H), 7.71 (d, *J* = 6.0 Hz, 2H), 7.64 (dd, *J* = 8.2, 5.2 Hz, 2H), 7.56 (d, *J* = 6.1 Hz, 2H), 7.45 (dd, *J* = 6.0, 2.0 Hz, 2H), 7.26 (dd, *J* = 6.1, 1.9 Hz, 2H), 1.42 (s, 18H), 1.31 (s, 18H). ^{13}C-NMR (101 MHz, RT, MeOD-d_4): δ [ppm] = 164.48, 164.31, 158.54, 158.39, 154.02, 152.28, 152.15, 151.80, 147.44, 131.93, 130.30, 127.52, 126.27, 126.12, 124.10, 122.83, 122.75, 119.65, 36.69, 36.58, 30.65, 30.55. HRMS: (MALDI-FT-ICR): 948.40126 [M–2PF_6^-–H^+]$^+$ (calcd. 948.4010). MS (ESI): 474 [M-2Cl^-]$^{2+}$. Crystal data for **3b**: $C_{61}H_{37}Cl_2N_{12}Ru$, M_r = 1140.23 g mol^{-1}, red prism, monoclinic, space group *P* 2$_1$/n, a = 14.43810(10) Å, b = 25.6586(2) Å, c = 15.6446(2) Å, α= 91.2930(10)°, V = 5794.25(10) Å^3, T = 150(2) K, Z = 4, $\rho_{calcd.}$ = 1.307 Mg/m^3, µ (Cu-K$_\alpha$) = 3.423 mm^{-1}, F(000) = 2380, altogether 41016 reflexes up to h(-18/15), k(-32/30), l(-17/19) measured in the range of 7.463° ≤ Θ ≤ 74.479°, completeness Θ_{max} = 99.7 %, 11819 independent reflections, R_{int} = 0.0342, 10349 reflections with $F_0 > 2\sigma(F_0)$, 648 parameters, 13 restraints, $R1_{obs}$ = 0.0480, $wR2_{obs}$ = 0.01316, $R1_{all}$ = 0.0556, $wR2_{all}$ = 0.1373, GOOF = 1.097, largest difference peak and hole: 1.404/-1.221 e·Å^{-3}. The position of the amine function in the imidazole ring has been assigned with respect to the hydrogen bond towards the chloride anion. Due to the considerable disorder of solvent molecules, restraints were applied in order to fix their respective geometry, i.e. in particular the DFIX for C-C distances (1.400 Å) and C≡N distances (1.050 Å), and DANG C-N (2.450 Å) in acetonitrile. The solvent molecules were left isotropic. Protons at the amine function were added using standard N-H distances (0.91 Å). CCDC 1520547 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

[Bis(4,4'-di-*tert*-butyl-2,2-bipyridine)-(4-(1*H*-imidazo-4,5-f-1,10-phenanthroline-2-yl)azido)-Ruthenium(II)]Cl_2 (4b): 3b (0.455 g, 0.45 mmol) was dissolved in a solution of H_2O (7 ml), MeCN (7 ml) and 3 M HCl (0.3 ml) and stirred for 20 min. $NaNO_2$ (15.8 mg, 0.229 mmol, 1.2 eq) dissolved in H_2O (0.3 ml) was added. The red solution was stirred for 10 min. The solution was cooled to 0° C and NaN_3 (25.0 mg, 0.385 mmol, 2.0 eq.) dissolved in H_2O (0.3 ml) was added. The red solution was stirred at 0° C for 10 min and 2.5 h at room temperature (RT). The solution was neutralized with ammonia solution (25 % in water. To the remaining red solution aqueous NH_4PF_6 was added. The precipitate was filtered off, reprecipitated as Cl salt with TBACl and purified via size exclusion chromatography (Sephadex©, MeOH) to receive the product as a red solid (84%, 0.398 g, 0.38 mmol). ^{1}H NMR (400.13 MHz, RT, MeCN-d_3): δ = 12.13 (s, 1H), 9.06 (d, *J* = 8.2 Hz, 1H), 8.85 (d, *J* = 7.9 Hz, 1H), 8.50 (dd, *J* = 18.9, 1.6 Hz, 4H), 8.29 (d, *J* = 8.8 Hz, 2H), 7.99 (d, *J* = 5.2 Hz, 2H), 7.79 (dd, *J* = 8.3, 5.3 Hz, 2H), 7.69 (d, *J* = 6.0 Hz, 2H), 7.46 (dd, *J* = 6.0, 2.0 Hz, 4H), 7.33 (d, *J* = 8.8 Hz, 2H), 7.19 (d, *J* = 4.8 Hz, 2H), 1.45 (s, 18H), 1.35 (s, 18H). ^{13}C NMR (101 MHz, MeCN-d_3): δ= 163.47, 163.34, 157.90, 157.76, 152.70, 152.09, 151.93, 150.85, 146.71, 142.84, 130.85, 128.79, 126.79, 126.45, 125.57, 125.43, 122.41, 122.31, 120.42, 118.26, 36.26, 36.16, 30.42, 30.32 ppm. HRMS (MALDI-FT-ICR): 1120.36379, [M-PF_6^-]$^+$ (calcd. 1120.36347), 2385.69779 [2 M-PF_6^-]$^+$ (calcd. 2385.69167), MS (ESI): 488 [M-2Cl^-]$^{2+}$, 506 [M-Cl^-+H^+]$^{2+}$, 1011 [M-Cl^-]$^+$. IR: v=2095-2123 (s, doublet, N_3-stretching) cm^{-1}.

[Bis(4,4'-di-*tert*-butyl-2,2-bipyridine)-(4-(1*H*-imidazo-4,5-f-1,10-phenanthroline-2-yl)-1-*H*-1,2,3-triazolyl-4-phenyl)-Ruthenium(II)]Cl_2 (5b): 52 mg (0.04 mmol) of **4b** (PF_6 salt) and 6 mg (6.5 µl, 0.06 mmol, 1.5 eq) of phenylacetylene were dissolved in MeCN. NaAsc (8 eq, 62 mg) and $CuSO_4$·5 H_2O (4 eq, 39 mg) were dissolved in 2 mL water respectively, added to the reaction mixture and the emulsion was stirred rapidly overnight at RT. The product was extracted with dichloromethane (DCM), dried over Na_2SO_4. Reprecipitation as Cl salt with TBACl and purification via size exclusion chromatography (Sephadex©) in MeOH yielded the product as a bright orange band. After removal of the solvent, the product was isolated as red crystals in good yield (77%, 42 mg, 0.03 mmol). ^{1}H NMR (400.13 MHz, MeCN-d_3): δ= 8.97 (d, *J* = 8.3 Hz, 2H), 8.74 (s, 1H), 8.51 (d, *J* = 18.5 Hz, 4H), 8.44 (d, *J* = 8.3 Hz, 2H), 8.08 (d, *J* = 7.8 Hz, 2H), 7.99 (d, *J* = 4.9 Hz, 2H), 7.94 (d, *J* = 7.4 Hz, 2H), 7.76 (dd, *J* = 18.4, 13.0 Hz, 2H), 7.71 (d, *J* = 5.9 Hz, 2H), 7.55 – 7.35 (m, 7H), 7.22 (d, *J* = 4.6 Hz, 2H), 1.46 (s, 18H), 1.35 (s, 18H). ^{13}C-NMR (101 MHz, MeCN-d_3): δ [ppm] = 163.11, 162.88, 158.19, 157.87, 151.99, 151.91, 148.70, 147.17, 145.30, 139.03, 136.66, 133.37, 131.74, 130.73, 130.64, 129.98, 129.72, 129.23, 128.35, 128.02, 126.52, 125.39, 125.28, 122.25, 122.15, 121.23, 119.84, 36.28, 36.17, 30.51, 30.42. HRMS (MALDI -FT-ICR):1076.43889 [M–2 PF_6–H]$^+$ (calcd. 1076.43842), 2297.84968 [2 M–3 PF_6—2 H^+]$^+$ (calcd. 2297.84156). Crystal data for **5b**: $C_{134}H_{148}F_{24}N_{26}O_3P_4Ru_2$, M_r = 2952.79 g mol^{-1}, red block, triclinic, space group P-1, a = 16.1146(5) Å, b = 19.6835(6) Å, c = 23.6612(8) Å, α = 80.3508(15)°, β= 87.6796(16)°, γ = 69.9649(15)°, V = 6950.0(4) Å^3, T = 150.0 K, Z = 2, $\rho_{calcd.}$ = 1.411 Mg/m^3, µ (Mo-K$_\alpha$) = 0.357 mm^{-1}, F(000) = 3044.0, altogether 120459 reflexes up to h(-18/18), k(-23/23), l(-27/27) measured in the range of 4.234° ≤ 2Θ ≤ 49.436°, completeness Θ_{max} = 100 %, 23619 independent reflections, R_{int} = 0.0628, 1850 parameters, 525 restraints, $R1_{obs}$ = 0.0773, $wR2_{obs}$ = 0.2014, $R1_{all}$ = 0.1119, $wR2_{all}$ = 0.2397, GOOF = 1.054, largest difference peak and hole: 1.65/-0.81 e·Å^{-3}. DFIX and ISOR restraints were used to fix the geometry of disordered solvent molecules. CCDC 1573111 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from The Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.

[Bis(4,4'-di-*tert*-butyl-2,2-bipyridine)-(4-(1*H*-imidazo-4,5-f-1,10-phenanthroline-2-yl)-phenyl-1-(1*H*-1,2,3-triazolyl-4,5- ((1R,8S,Z)-bicyclo[6.1.0]non-4-en-9-yl) methyl (2-(2-(2-aminoethoxy)ethoxy)-ethyl)carbamate)-Ruthenium(II)]Cl_2 (6b): 41.0 mg (32.41µmol) of **4b** (PF_6 salt) were dissolved in a small amount of MeCN. 8.2 mg (25.276µmol) *N*- [(1*R*, 8*S*, 9*S*)-bicyclo[6.1.0]-non-4-yn-9-yl-methyl-oxy-carbonyl]-1,8-diamino-3,6-dioxa-octane (BCN-amine) was dissolved in an ethanol and added subsequently. A slight warming of the resulting solution was observed and it was stirred for 16 h at rt. The solvent was removed and the product was reprecipitated as Cl-salt (crude yield: 80%). The solid was filtered via a syringe filter (PTFE), washed with diethylether and dried. The product was dissolved from the filter with MeOH and purified via size exclusion chromatography (Sephadex©). **6b** was isolated as red solid in a moderate yield (13.80 mg (10.069µmol), 40%).^{1}H-NMR (MeOD-d_4, 500.13 MHz): δ = 9.18 (d, *J*= 8.3 Hz, 2H), 8.77 (dd, *J*= 21.0, 1.8 Hz, 4H), 8.53 (dd, *J*= 8.7, 2.3 Hz, 2H), 8.11 (dd, *J*= 5.3, 1.1 Hz, 2H), 7.93 –7.79 (m, 4H), 7.66 –7.56 (m, 6H), 7.39 (dd, *J*= 6.1, 1.6 Hz, 2H), 4.18 (d, *J*= 8.1 Hz, 2H), 3.78 –3.63 (m, 6H), 3.55 (t, *J*= 5.6 Hz, 2H), 3.35 (s, 2H), 3.26 –3.16 (m, 1H),

3.16 –3.03 (m, 3H), 3.03 –2.91 (m, 1H), 2.89 –2.74 (m, 1H), 2.37 –2.18 (m, 2H), 1.82 –1.63 (m, 2H), 1.50 (s, 18H), 1.39 (s, 18H), 1.32 (d, *J*= 17.2 Hz, 2H), 1.15 (s, 2H) ppm. ^{13}C NMR (101 MHz, MeOD-d_4): δ = 164.42, 164.25, 158.59, 158.46, 153.48, 152.19, 152.13, 151.24, 147.37, 146.42, 139.16, 136.68, 131.94, 131.80, 129.16, 127.73, 127.68, 127.34, 126.24, 126.11, 122.82, 122.74, 71.37, 71.31, 71.09, 67.87, 63.61, 61.74, 49.00, 41.54, 40.71, 36.70, 36.58, 30.65, 30.55, 26.52, 26.43, 24.56, 24.50, 23.81, 23.43, 23.35, 21.16, 20.79, 20.63, 19.13 ppm. HRMS (MALDI-FT-ICR): 1298.59747 $[M–2Cl^{-}–H^{+}]^{+}$ (calcd. 1298.59637).

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Keywords: Ruthenium • click chemistry • CuAAC • SPAAC • maleimides

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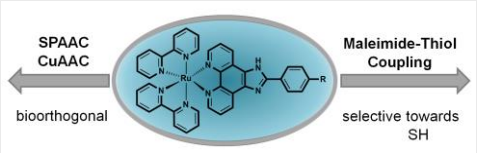

A library of Ru(II) poylpyridine complexes is presented which allows the application of different coupling reactions with potentially biologically active substrates. Therefore precursor substrates for CuAAC, SPAAC and maleimide-thiol coupling are generated and the feasibility of the click reactions is evaluated. All presented precursor compounds resort to one easy-to-synthesize scaffold to retain essential photophysical properties.

Ruthenium; Click Chemistry

Anne Stumper, Martin Lämmle, Alexander K. Mengele, Dieter Sorsche and Sven Rau

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One Scaffold, Many Possibilities: CuAAC, SPAAC and Maleimide-Thiol Coupling of Ruthenium(II) Polypyridyl Complexes