SUPPORTING INFORMATION

An anthracene-pendant ruthenium(II) complex conjugated to a biotin anchor, an essential handle for photo-induced anti-cancer activity

Geângela de Fátima Sousa Oliveira,[a] Aryane de Azevedo Pinheiro[b], Luiz Gonzaga do Nascimento Neto[b,c], Mayron Alves de Vasconcelos[b,d], Edson Holanda Teixeira[b], Ana Claudia Silva Gondim,[a] Florencio Sousa Gouveia Jr[a], Luiz Gonzaga de França Lopes,[a] Idalina Maria Moreira de Carvalho,[a*] Eduardo Henrique Silva Sousa[a*]

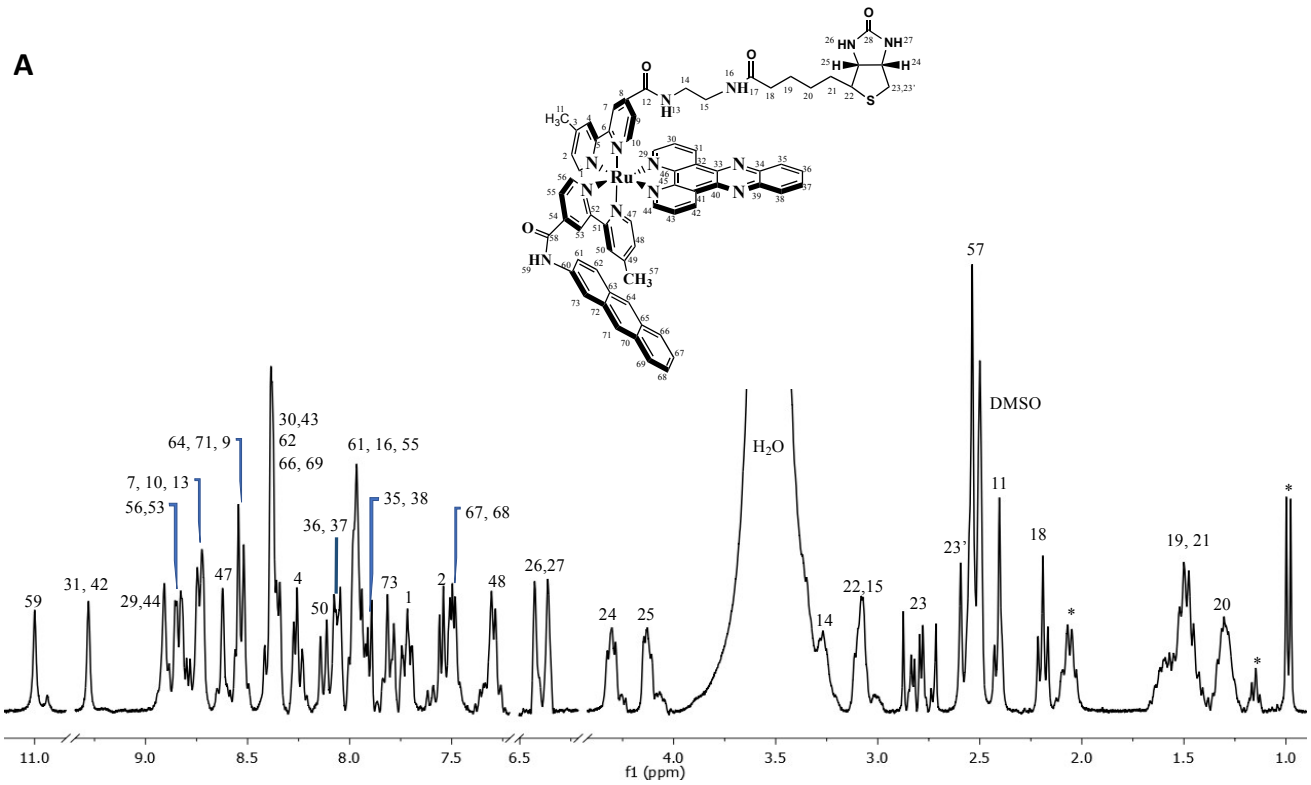
a- Group of Bioinorganic, Department of Organic and Inorganic Chemistry, Federal University of Ceará, Fortaleza, Brazil

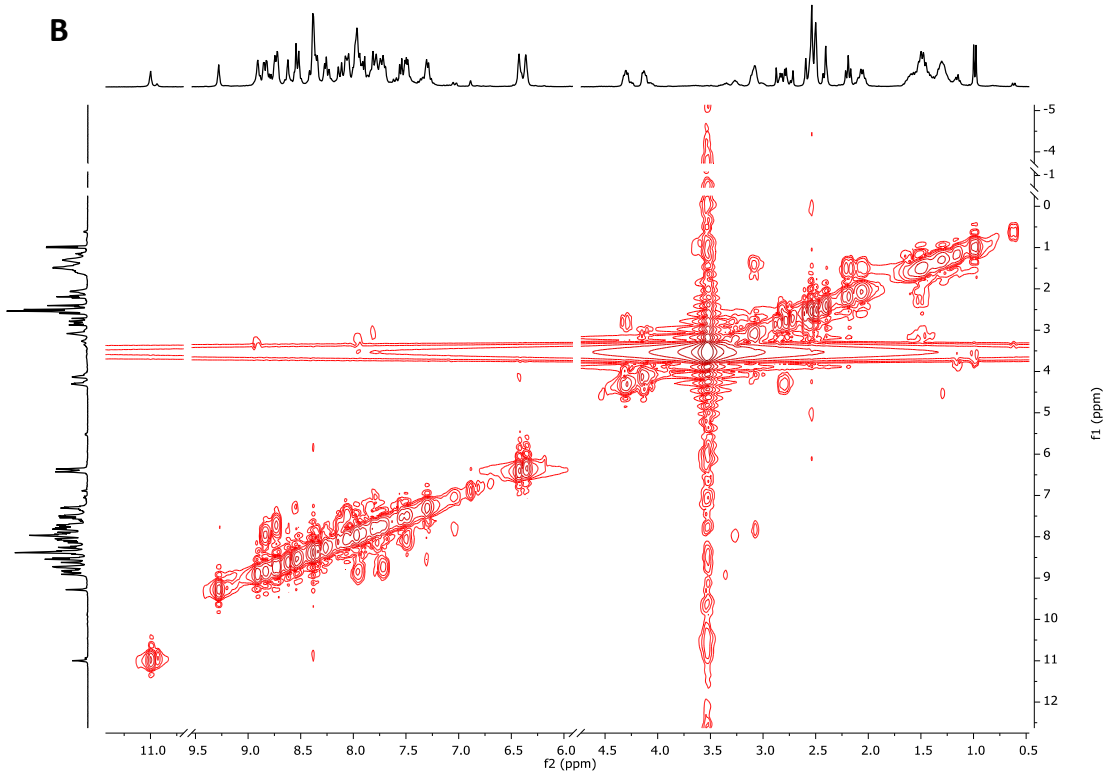
b- Laboratório Integrado de Biomoléculas, Departamento de Patologia e Medicina Legal, Universidade Federal do Ceará, 60441−750, Fortaleza, Ceará, Brazil

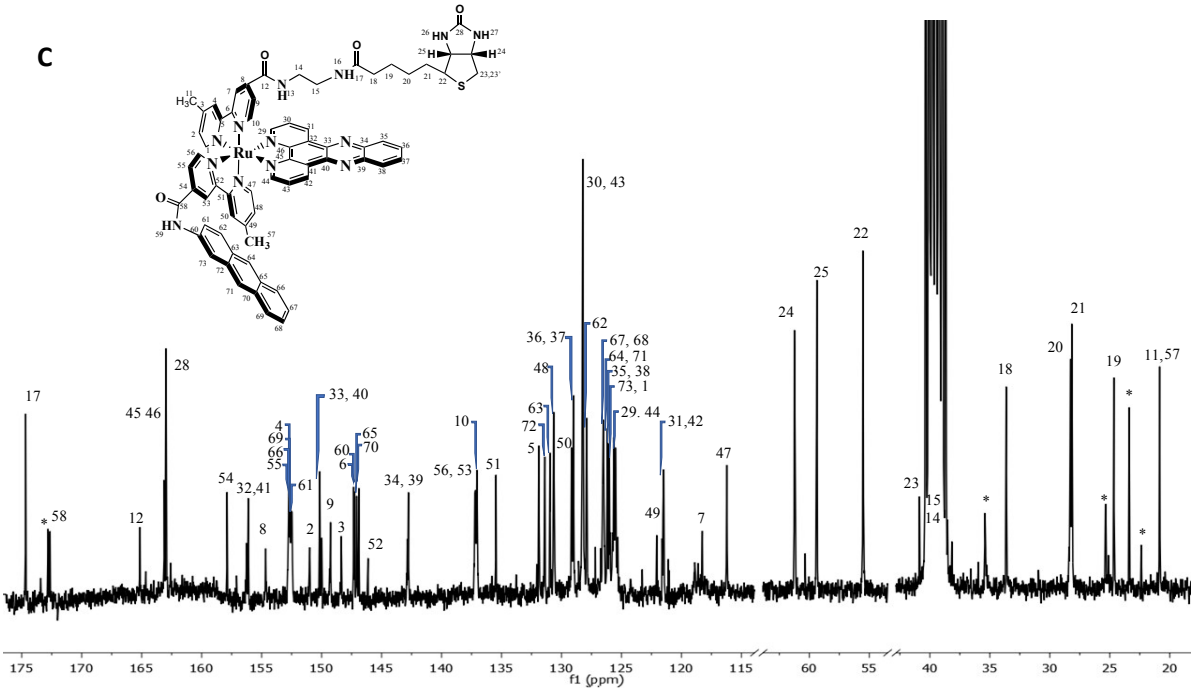
c- Departamento do Núcleo Comum, Instituto Federal de Educação, Ciência e Tecnologia do Ceará, Campus Limoeiro do Norte, 62930-000, Limoeiro do Norte, Ceará, Brazil

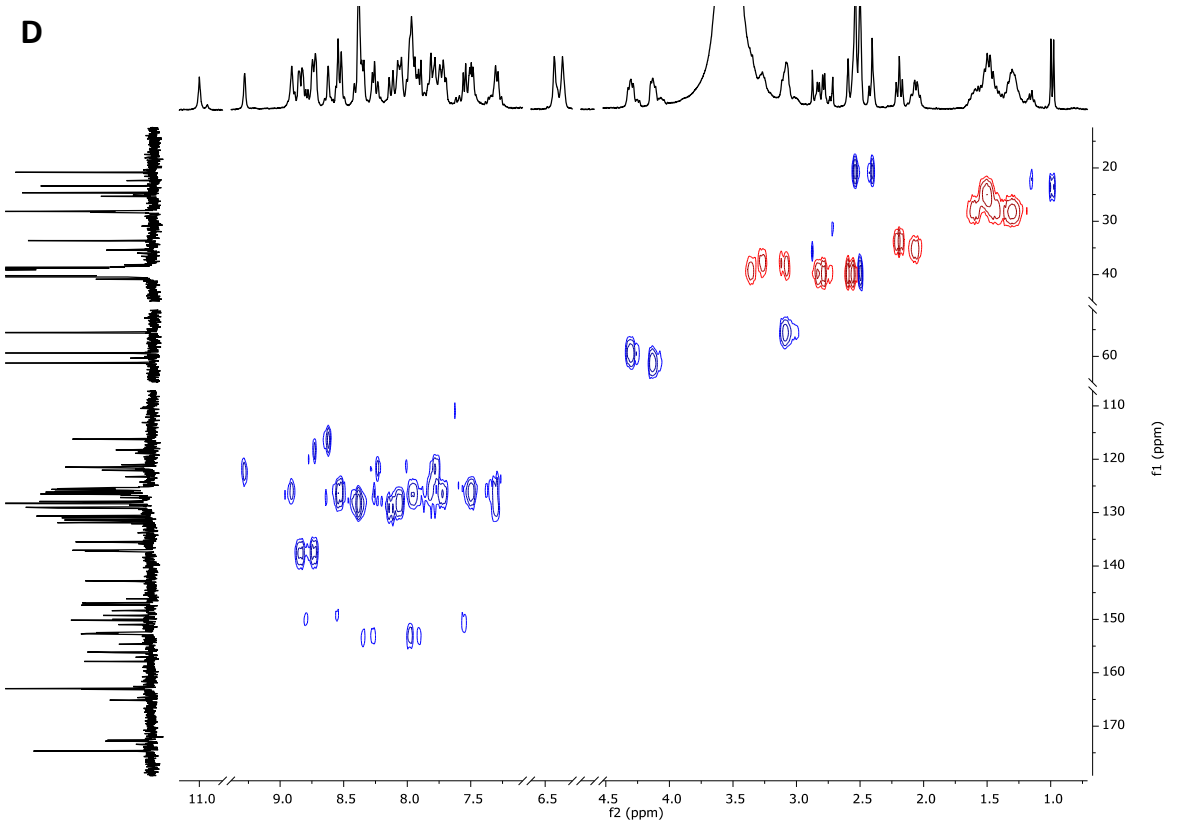
d - Faculdade de Ciências Exatas e Naturais, Universidade do Estado do Rio Grande do Norte, 59625-620, Mossoró, Rio Grande do Norte, Brazil

*corresponding authors: idalina@dqoi.ufc.br, eduardohss@dqoi.ufc.br


A
59
31, 42
29,44
56,53
7, 10, 13
64, 71, 9
47
30,43
62
66, 69
4
50
36, 37
61, 16, 55
35, 38
73
1
2
67, 68
48
26,27
24
25
H2O
14
22,15
23
23'
57
DMSO
11
18
19, 21
20
*
f1 (ppm)


B
f2 (ppm)
f1 (ppm)


C
17
28
45 46
58
12
54
32,41
8
4
69
66
55
61
2
33, 40
9
3
52
60
6
65
70
34, 39
56, 53
10
51
36, 37
48
63
72
5
50
62
67, 68
64, 71
35, 38
73, 1
29. 44
49
31,42
7
47
24
25
22
30, 43
23
15
14
18
20
21
19
11,57
f1 (ppm)


D
f2 (ppm)
f1 (ppm)

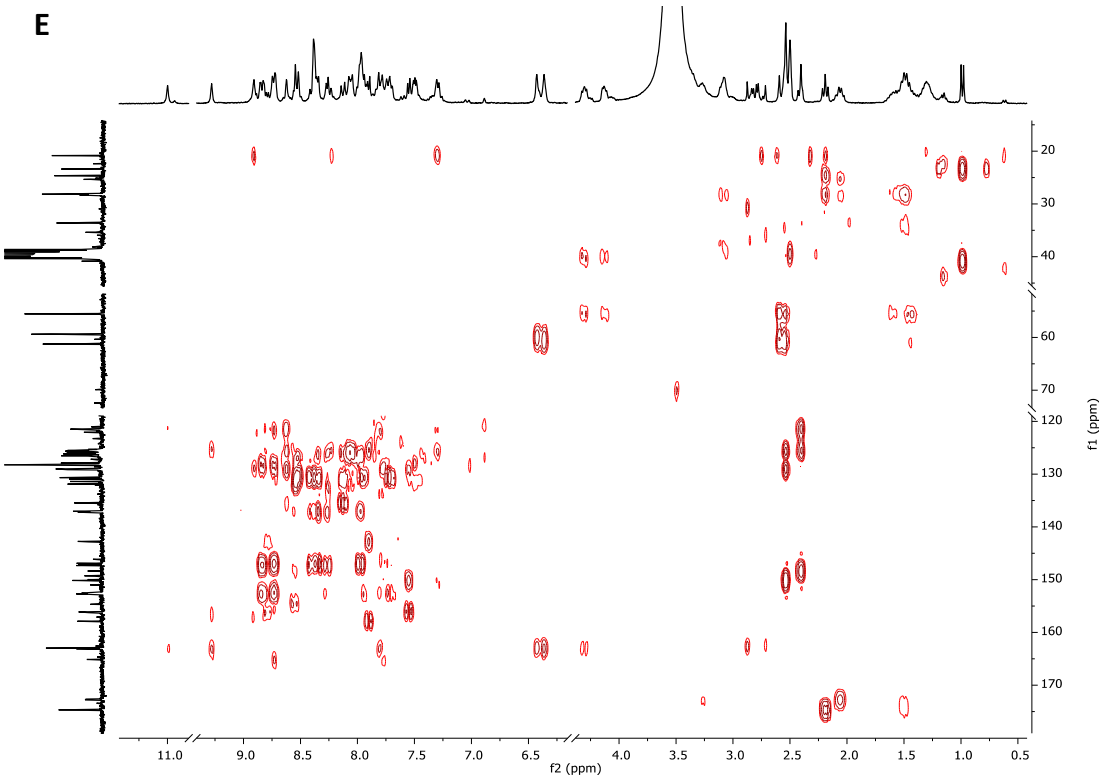


Fig. S1. NMR for **Ru-biot** in $(CD_3)_2SO$ (300 MHz, 303 K). ^1H NMR **(A),** ^1H-^1H COSY **(B),** ^{13}C NMR **(C)**, ^1H-^{13}C HSQC **(D,** red color for CH_2, and blue for CH and CH_3**)**, ^1H-^{13}C HMBC **(E)**.

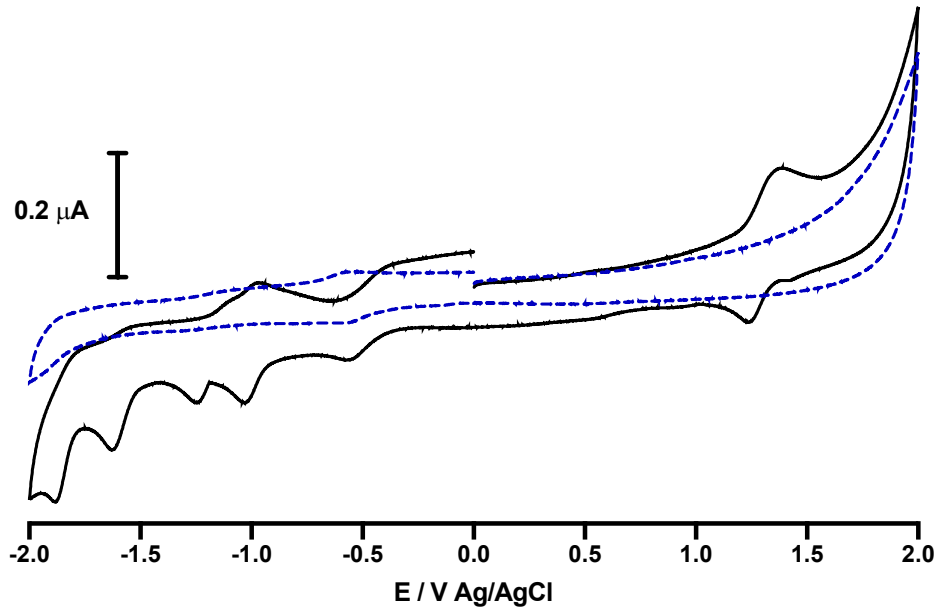


Fig. S2. Cyclic voltammogram of **Ru-biot** in acetonitrile (0.1 M PTBA, 100 mV s^{-1}), initial anodic scan (blank in dashed blue line).

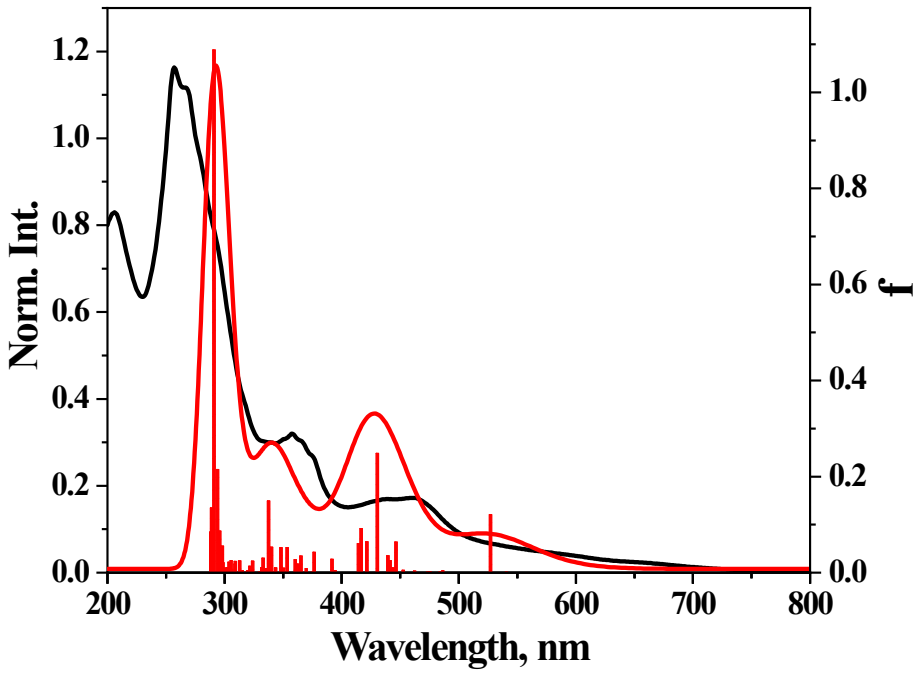


Fig. S3. Calculated (**—**) and experimental (—) electronic absorption spectra of **Ru-biot** (right side Y-axis is normalized intensity, left side is the oscillator strength)

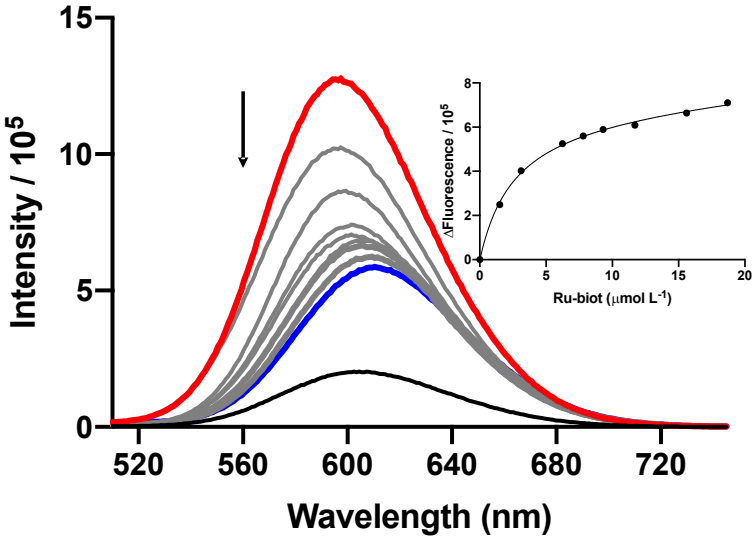


Fig. S4. Competition assay of ethidium bromide (1.5 μmol L^{-1}) with DNA upon titration with Ru-biot (data was fit to a single binding equation, where K_d^{app} was of 2.58 x 10^6, R^2 = 0.998).

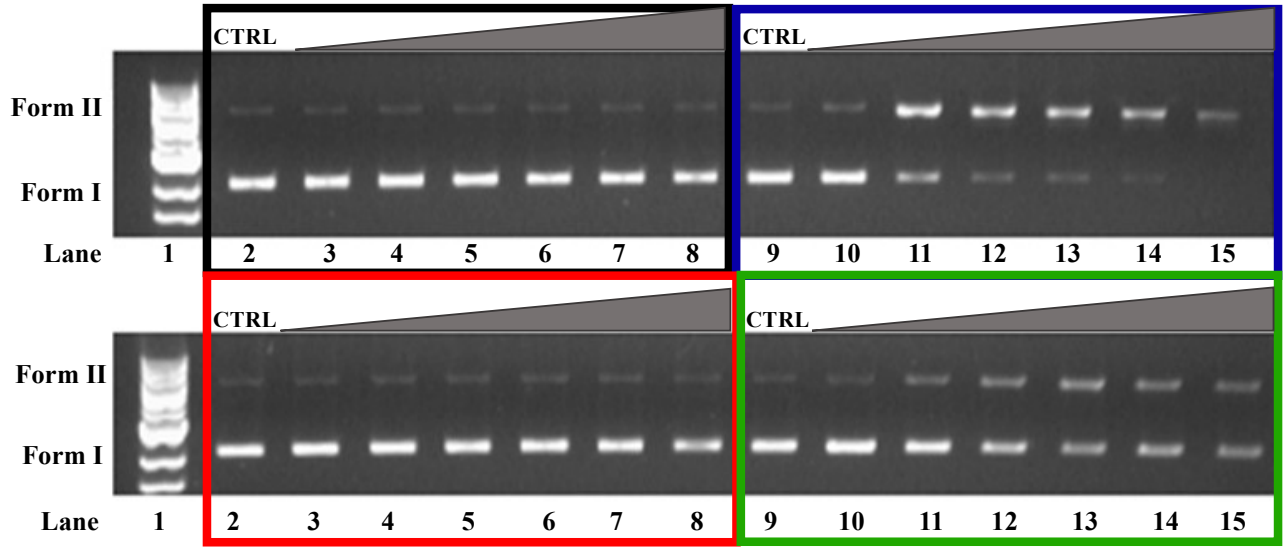


Fig. S5. Photocleavage of 20 μM (in base pair) pBR322 DNA in the presence of **Ru-anth** in the dark and after 1 h of irradiation with blue, green and red LEDs. In all experiments, lane 1 contains only linear DNA ladder and lanes 2 and 9 only pBR322 DNA, while lanes 3−8 and 10−15 contained **Ru-anth** in the following concentrations of 0.3, 3.0, 7.0, 15 and 30 μM. Dark, blue, green and red boxes indicate either the experiment was carried out in the dark or with blue, green or red-light irradiation.

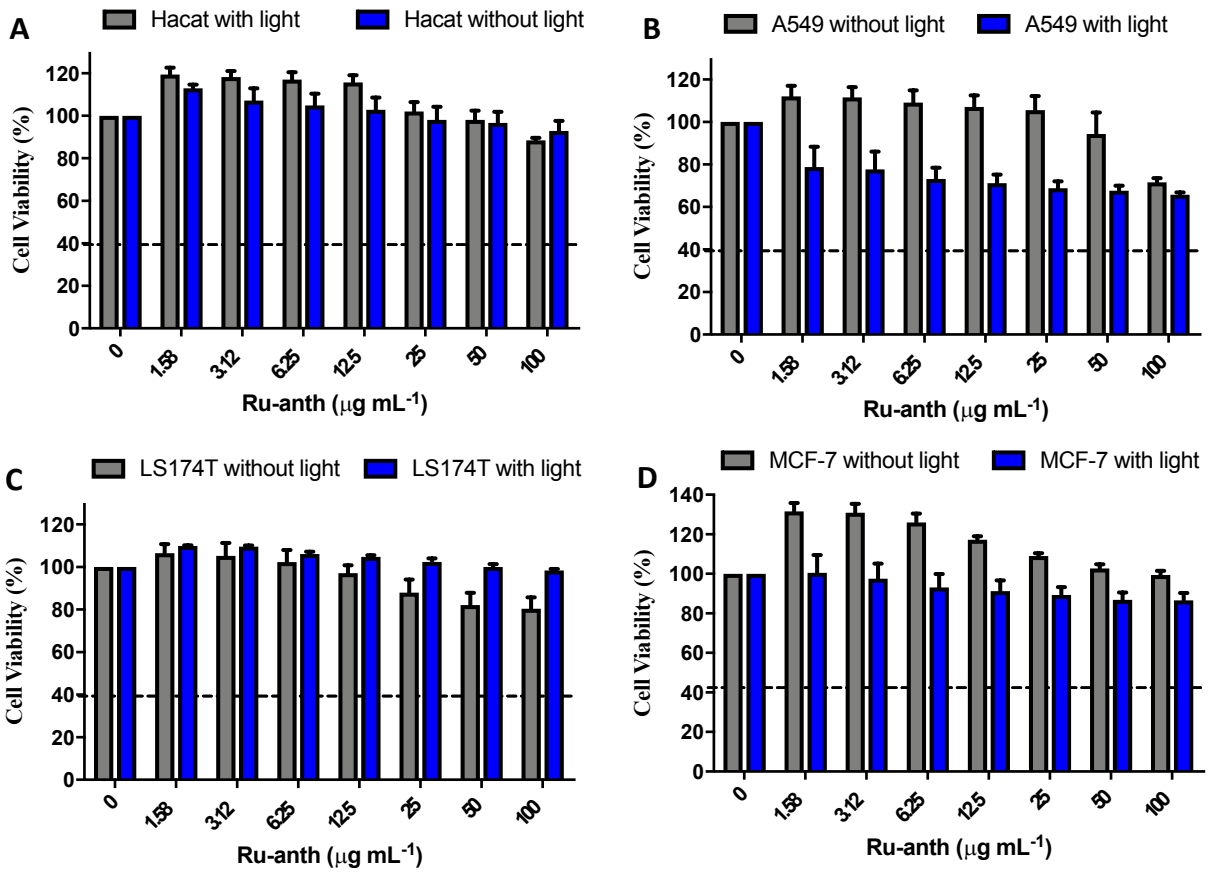


Fig. S6. HaCaT, A549, LS174T and MCF-7 cell viability determined by MTS assay after 48 h of treatment with **Ru-anth** in different doses, without or with blue light exposition for 1h (463 nm, 425 mW cm^{-2}).

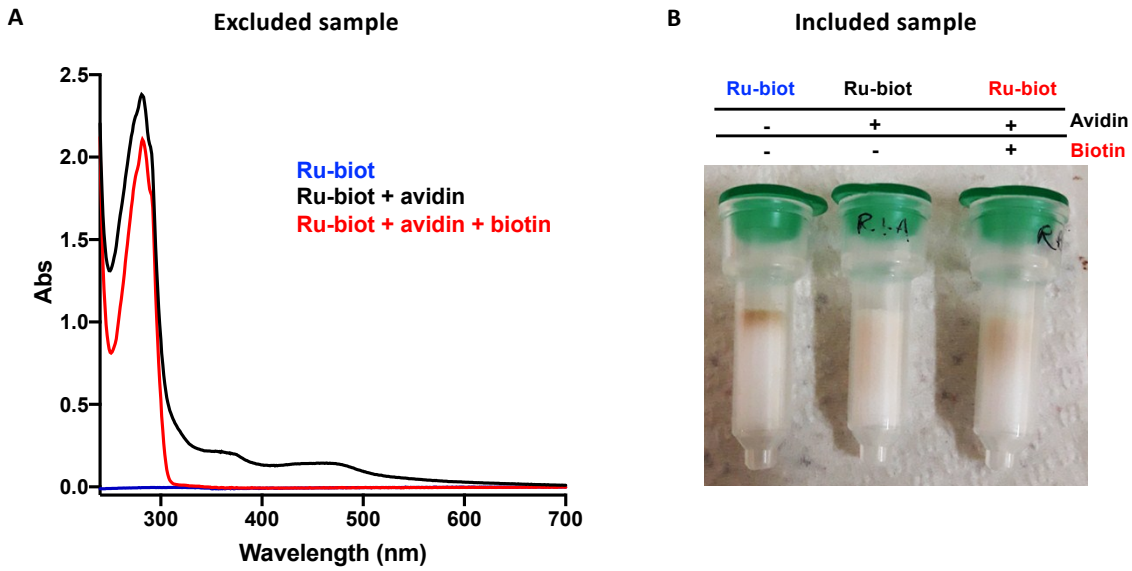


Fig. S7. Binding to avidin measured by using gel exclusion column. Panel A shows the electronic spectra of the excluded samples after applied onto a micro Biospin 6 column, the collected samples were from a mixture containing only Ru-biot (applied as 100 μM) (Blue trace), Ru-biot (100 μM) with avidin (100 μM) (black trace), and Ru-biot (100 μM) with avidin (100 μM) plus biotin (1 mM) (red trace). Panel B shows the resin with the included sample right after the centrifugation, where the color observed indicates the presence of Ru-biot.

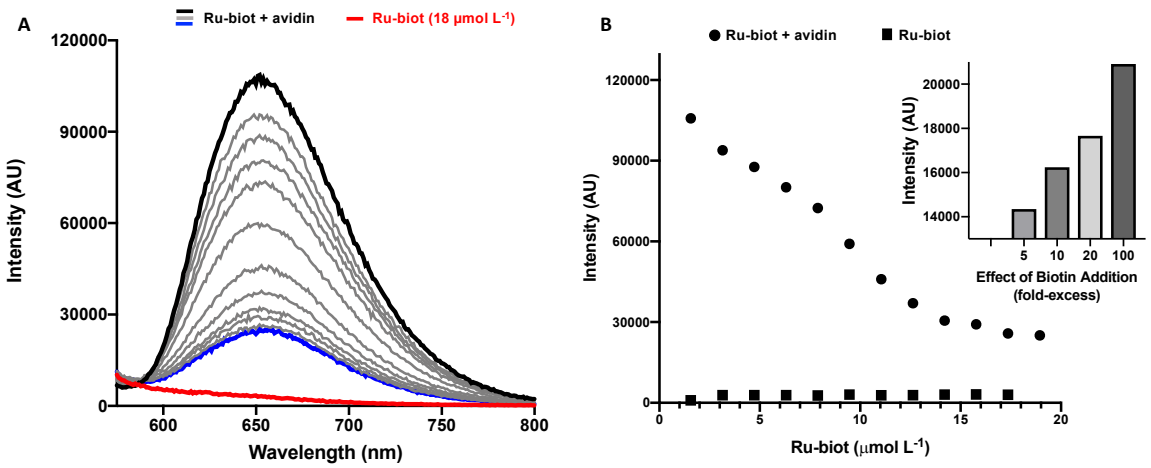


Fig. S8. Titration of aviding using **Ru-biot**. Panel A show the luminescence profile during addition of **Ru-biot** into a solution containing avidin (4 µM), black trace is the emission spectrum of 1.6 µM of **Ru-biot** added, while the grey traces are further additions until the last one (19 µM) (blue trace). The red trace is the luminescence spectrum for **Ru-biot** (18 µM) without avidin. Panel B shows two set of data, one for the titration of **Ru-biot** on a solution containing avidin (circle data) and another without avidin (squares) as monitored by luminescence (at 656 nm). Inset shows a bar plot of the changes in luminescence upon addition of an excess of biotin right after titration of avidin with **Ru-biot**.

Table S1. Calculated Electronic Absorption Transitions and Assignments for **Ru-biot.**

Exptl. λ (nm)	**Calc. λ (nm)**	***f***	**Major contribution**	**Character**	
600	540	0.0013	HOMO→LUMO (99%)	π(anth) → π*(dppz)	LLCT
550	527	0.1207	HOMO → L+1 (89%)	π(anth) → π*(bpy)	ILCT
457	430	0.2485	H-4 → L+1 (46%) H-3→L+2 (28%)	dπ (Ru) → π*(bpy-anth) dπ (Ru) → π*(bpy-biot)	MLCT
437	416	0.0916	H-4 → L+2 (23%) H-3 → L+3 (30%)	dπ (Ru) → π*(bpy-biot) dπ (Ru) → π*(dppz)	MLCT
368	337	0.1493	H-12 → LUMO (59%) H-9 → L+4 (21%)	π(dppz) → π*(dppz) π(dppz) → π*(dppz)	IL
268	293	0.2147	H-16 → L+2 (28%) H-15 → L+2 (29%)	π(biotin) → π*(bpy-biot) π(biotin) → π*(bpy-	IL

				biot)	
256	291	1.0885	H-12 → LUMO (13%) H-9 → L+4 (43%)	π(bpy) → π*(dppz) π(dppz) → π*(dppz)	IL