Development of a High-Intensity Parallel Photoreactor for High Throughput Screening

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1.0 General information.

1.1 General reagents and methods.
All reagents were used as purchased from commercial suppliers. Solvents for reactions were purchased from Sigma Aldrich, anhydrous, sure-seal quality, and used with no further purification. All reactions were set up and sealed inside an MBraun glovebox operating with a constant N_2-purge (oxygen typically <5 ppm). Aryl bromide **5** was synthesized as described in literature.[1]

Analytical chemicals and reagents: Dimethyl sulfoxide (99.9%, Fisher certified ACS), acetonitrile (Fisher Optima LC/MS Grade) and water (Fisher Optima LC/MS Grade) were obtained from Fisher Scientific (Waltham, MA, USA). Trifluoroacetic acid (≥99.0%), acetic acid and methanol were obtained from Sigma-Aldrich (St. Louis, MO, USA).

1.2 UPLC-MS analysis.
Reactions were monitored using a Waters Acquity UPLC I-Class system (Waters Corp., Milford, MA, USA) equipped with a binary pump, a FTN sampler, column manager, a photodiode array detector, SQD detector 2 with electrospray ionization (ESI) source in the positive mode and MassLynx software. Separations were performed on a Waters CORTECS UPLC C18 column (50 × 2.1 mm, 1.6 μm).

Conditions: Mobile phase A = 0.1% TFA in H_2O and B = 20% MeOH in MeCN.
Gradient:

Time (min)	Flow (mL/min)	% A	% B
0.00	0.700	95	5
1.70	0.700	10	90
1.95	0.700	10	90
1.96	0.700	95	5
2.00	0.700	95	5

Table S1. Parameters of 2-minute UPLC analytical method.

Flow rate 0.7 mL/min; column temperature = 45 ºC; injection volume = 1 μL; UV scan = 210 – 500 nM. Acetonitrile (HPLC grade), 0.1% TFA in H_2O (HPLC grade). 20% MeOH in MeCN solution was prepared by mixing 800 mL of MeOH with 3200 mL MeCN. High throughput data analysis was done with Virscidian Analytical Studio™ software. Conversion to product was analyzed by UPLC-UV. Area percent of product, remaining starting material and side product peaks at 210 nm were calculated.

2.0 Photochemistry high-throughput experimentation workflow.

2.1 Reaction and source plates.
Corning 3657 384-well microplates (95 μL-wells, round bottom, non-treated clear polypropylene) were used as source plates for stock solutions. Advantage™ 384-well plates (Analytical Sales, Cat. No. 38120, polypropylene, 120 μL-wells, flat bottom, clear) were used as analytical plates on

UPLC-MS. Corning 1536-well plates (Corning Echo^{TM} qualified, Cat. No. 3730, Cyclic Olefin-Copolymer COC, 12.5 µL-wells, flat bottom, clear) and 384-well plates (UV-Star Plate, F-Bottom, µClear, CLEAR, Cycloolefin, 110 µL, catalogue # 781801) were used as reaction plates.

2.2 Robotics dosing.

The reactions in this work were dosed with the SPT Labtech Mosquito® LV robot and quenched using the Thermo Matrix 2x2 Platemate robot.

2.2.1 Mosquito® LV robotics dosing.

Dosing of reaction components into the reaction 1536-well plates in reactions with low-volatility solvents was accomplished in the glovebox using a Mosquito® LV HTS ("Mosquito® LV") liquid handling robot (Figure S1, SPT Labtech, 4.5 mm pitch tip spool) with no special modifications and using the SPT Labtech native software.

2.2.2. Thermo matrix Platemate robotics dosing.

For reaction quenching and preparation of analytical plates, dosing of reaction components into 384-well plates was accomplished in the glovebox using a Matrix 2x2 Platemate ("Matrix") liquid handling robot from Thermo Scientific with no special modifications and using the ControlMate native software. This robot enables faster dosing with simultaneous 384-tip additions (see Fig. S1). For all experiments in this work, 30 µL tips in disposable magazines from Thermo Scientific (Catalog # 5316) were used.
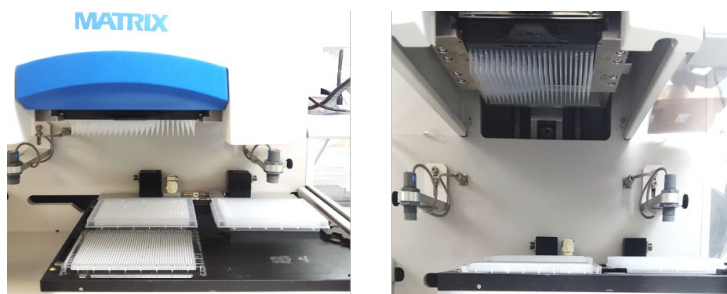
Figure S1. Versatile dosing modes of the Thermo Matrix Platemate liquid handling robot with 384-tip dosing.

2.3 Photoredox reaction plate set-up.

We developed a platform for parallel plate-based photoredox chemistry. The photochemical reaction plate used in the photoredox reactions described below is shown in Figure S2. The acrylic bottom enables light penetration, while providing a solid bottom for the 1536-well plate to rest. The reaction plate was sealed with Peelable Aluminum RT seal (Agilent Technologies, Catalog #24214-001) using a Velocity 11 PlateLoc Thermal Plate Sealer (Catalog #23480) and capped using the aluminum sealing block shown in Figure S3 below. This set-up provides a high-quality seal that prevents solvent loss.
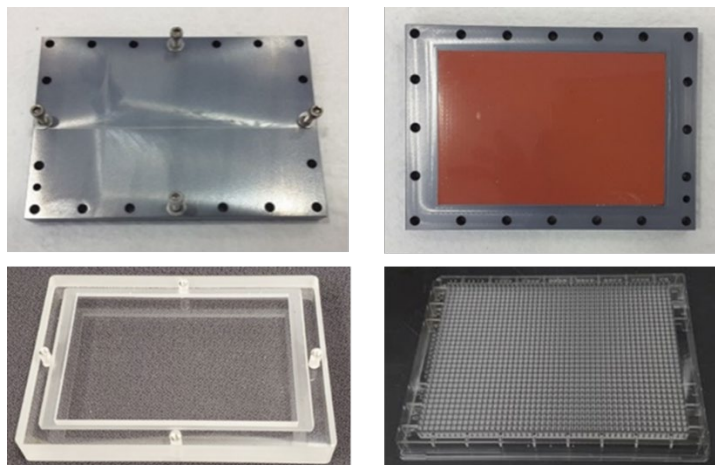
Figure S2. Left to right, top then bottom: Aluminum top for nanomole-scale chemistry, aluminum top underside with silicone rubber mat, acrylic bottom for photoredox reactions that enable efficient light penetration and COC 1536-well plate.

2.4 Description of photoreactors studied.

2.4.1 Kessil lamp photoreactor.

In this first generation photoreactor developed for parallel nano-mole scale photoredox reactions[1], the sealed nano-photoredox plate is placed inside a vacuum oven (Fisher Scientific, Isotemp vacuum oven, Model 281A) lined with reflective aluminum foil on the interior for maximal light exposure. The plate sits on a borosilicate crystalizing dish to suspend it from the bottom of the oven. This approach eliminates edge effects since light is reflected all around instead of focusing directly on the bottom of the plate. The set-up is run under N_2 purge with a slight vacuum. A H150W tuna blue Kessil lamp (P/N: H150-blue, S/N: L4C3DG0006, 24 VDC, 1.5 A, 34 W) placed outside the vacuum oven was used to illuminate the reaction and the temperature of the reactions was maintained at 55 ºC, as shown in Figure S3. At the maximum applied power, the optical intensity at the reaction surface was measured at 0.8 mW/cm^2 (Table S2).

Figure S3. Kessil Lamp Photoreactor. Left to right. (A) Interior set-up of vacuum oven. (B) Irradiation with Kessil lamp.

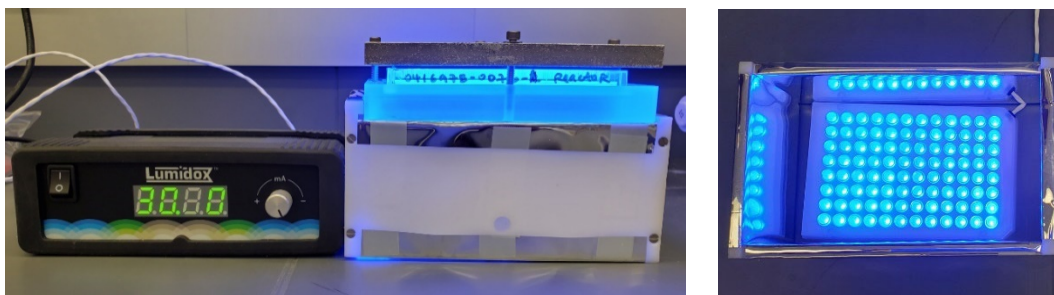### 2.4.2 Lumidox® I photoreactor.

Figure S4. Lumidox® I Photoreactor. Left to right: Controller set up and 96-membered array of blue LEDs.

Control and comparison studies with the new photoreactor were done using the commercially available parallel photoreactor from Analytical Sales & Services, Inc. This reactor set-up includes a controller (LUMCON, 9 VDC, 2.3 Amp, Fuse 1.6 A, 250 V) and a blue LED array (LUM96B, Lumidox® LED array, blue 470 nm, 96-well format) (Figure S4). At the maximum applied power, the optical intensity at the reaction surface was measured at 16 mW/cm^2 (Table S2).

2.4.3 BPR200 photoreactor.
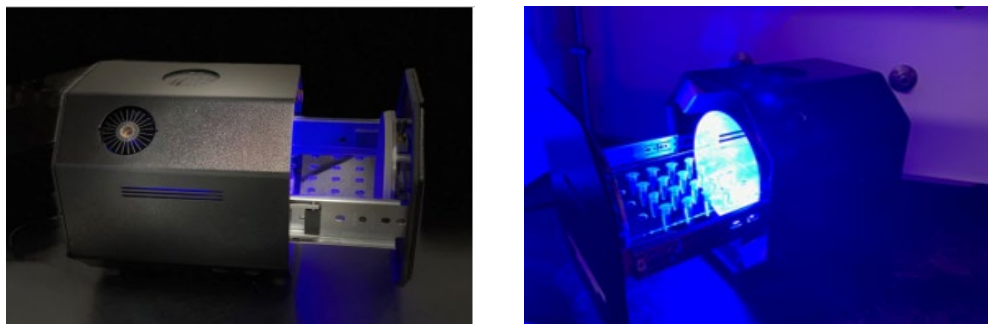
Figure S5. BPR200 photoreactor, available from Efficiency Aggregators.

Control and comparison studies with the new photoreactor were done using the commercially available parallel photoreactor from Efficiency Aggregators (Catalog number NC1558343). This reactor set-up allows for interchangeable wavelengths, multiplex photoredox reactions, covalent labeling of biomolecules, and is compatible for use in living cells. Manufacturer: Efficiency Aggregators LLC EBPR20080W. At the maximum applied power, the optical intensity at the reaction surface was measured at 8.3 mW/cm^2 (Table S2).

2.4.4 New high-intensity parallel photoreactor.

A 6 x 8 array of hexagonal quad CREE XTE LED boards (Kiwi Lighting) was used to provide high intensity 450 nm illumination. The LEDs were powered by a 0-60 V 0-8 A DC Laboratory Power Supply (BK Precision 9111). This allowed for adjustment and monitoring of applied electrical power. To reduce edge effects, the overall size of the LED array was larger than the standard SBS well plate footprint. The final configuration consisted of a polycarbonate diffuser (Makrolon Lumen XT LC3 0.060") placed 20 mm above the light source, with the plate placed 6

mm above the diffuser. A 120 mm fan mounted underneath the LED array heatsink provided forced convection room temperature air which passed first over the heatsink, and then around the illuminated plate, before exiting the top of the instrument. A blue light attenuating shield which was included for operator safety. The entire set-up is shown in Figure S6. At an applied power of 143 W (35.83 V @ 4 A), the optical intensity at the diffuser surface was measured at 188 mW/cm^2 (Table S2).
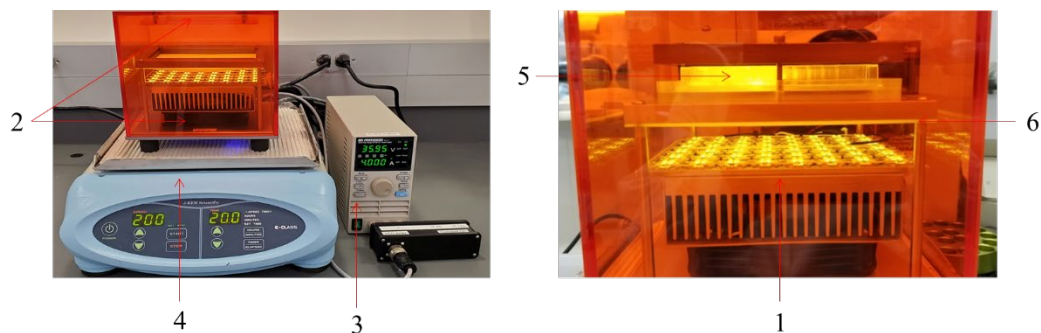
Figure S6. New parallel photoreactor. (1) Light source: 48 4-LED-clusters (1 W for each bulb, 192 W maximum energy output) (2) Efficient heat removal: fan in the bottom, air vent on the top (3) Controller that enables tunable light intensity (4) Smaller footprint makes gentle agitation (shaking) on a J-KEM MaxQ 2000 shaker possible (5) Compatible with COC reaction plates and glass vials, from nano- to milli-mole scale (6) Layer of diffuser for uniform light distribution

Photoreactor	Actual Voltage (V)	Actual Current (A)	Calc Wattage	Measured Optical Power (W)	Sensor Diameter (mm)	Optical Intensity (mW/cm^2)
New Parallel Photoreactor	30.98	0.125	3.873	0.024	26.000	4.5
	31.62	0.25	7.905	0.052	26.000	9.8
	32.32	0.5	16.160	0.120	26.000	22.6
	33.19	1	33.190	0.252	26.000	47.5
	34.3	2	68.600	0.505	26.000	95.1
	35.02	3	105.060	0.744	26.000	140.1
	35.83	4	143.320	0.998	26.000	187.9
Kessil Lamp	Maximum	Maximum	Maximum	0.004	26.000	0.8
BPR200	Maximum	Maximum	Maximum	0.044	26.000	8.3
Lumidox® 1	Maximum	Maximum	Maximum	0.086	26.000	16.2
Lumidox® 2	Maximum	Maximum	Maximum	0.900	26.000	169.5

Table S2. Optical power measurements and intensity calculations across parallel photoreactors. Optical power was measured using a thermal sensor power photometer (Thorlabs, Catalog number PM160T-HP).

3.0 Chemistry experimentation.

3.1 Photoreactor validation and intensity studies.
The goal of this experiment was to compare and contrast the effect of light intensity, hot spots and edge effects on the sp^2-sp^3 photoredox decarboxylative C–C coupling using the commercial photoreactors and the new parallel photoreactor.
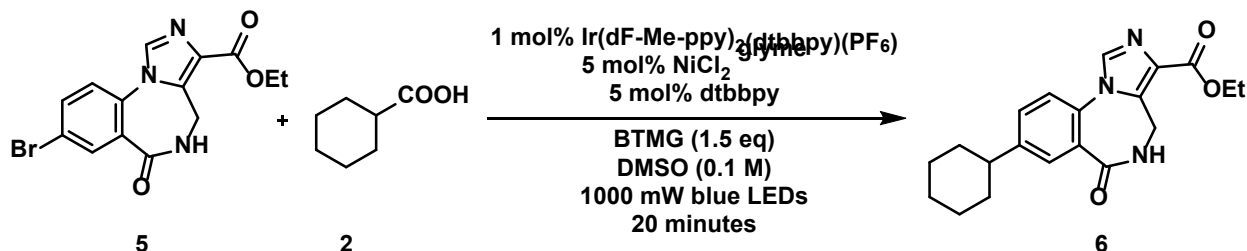
3.1.1 Chemistry set-up (100 nmol scale, 1 µL volume).
A stock solution containing all of the reaction components was made as follows: Aryl bromide **5**[3] (1 equiv), cyclohexanecarboxylic acid **2** (1.5 equiv), NiCl$_2$ glyme (0.05 equiv), 4,4′-Di-tert-butyl-2,2′-dipyridyl (0.05 equiv), Ir(dF-Meppy$_2$)(dtbbpy)(PF$_6$) (0.01 equiv) and BTMG (1.5 equiv) in 0.1 M DMSO. This stock solution was dispensed to a 384-well source plate, which was placed onto the Mosquito® LV deck. The Mosquito® LV robot was used to dose all combined reaction components in 1 µL aliquots into 384 wells of a 1536-well plate across every other row and column. Once the dosing was completed, the 1536-well plate was heat sealed, placed on the acrylic bottom and capped with the aluminum top to allow light penetration and set to react for 20 minutes. Four similar plates were prepared using the above method and placed on the following reaction settings:

Plate 1) New high-intensity parallel photoreactor at 1000 mW
Plate 2) Lumidox® commercial photoreactor from Analytical Sales at full intensity power
Plate 3) Kessil blue lamp in a positive nitrogen chamber at full intensity power
Plate 4) BPR200 photoreactor from Efficiency Aggregators at full intensity power

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
1	Stock 1																							
2	Stock 1																							
3	Stock 1																							
4	Stock 1																							
5	Stock 1																							
6	Stock 1																							
7	Stock 1																							
8	Stock 1																							
9	Stock 1																							
10	Stock 1																							
11	Stock 1																							
12	Stock 1																							
13	Stock 1																							
14	Stock 1																							
15	Stock 1																							
16	Stock 1																							
uL	70																							
nL	1000																							

3.1.2 Reaction work-up.

After reaction, each of the reaction plates was placed on the Matrix liquid handling robot together with a 384-well analytical plate containing 101.5 µL of DMSO stock solution of acetic acid (1%). From the 384-well analytical plate, the Matrix removed 4 µL into the 1536-well reaction plate. After which, 2.5 µL of the resulting quenched and diluted reaction plate was sampled back into the analytical plate, equivalent to a 200-fold dilution. The 384-well plate was then heat-sealed and shaken.

3.1.3 Analytical.

The quenched analytical plate was subjected to UPLC-MS analysis using a 2-minute method. The samples were carefully curated using Virscidian Analytical Studio™ (cutting overlapping peak shoulders, re-assigning misassigned peaks, etc.) to provide high quality UV data. Catalyst, ligand, carboxylic acid- and base-derived peaks and solvents were removed from these analyses to minimize their impact on area percent measurements. The LC area percent (LCAP) at UV 210 nm of product was used to determine assay yields. The amount of starting material remaining was also tabulated as LCAP. These were extracted from Analytical Studio to Excel.

3.1.4 Results and discussion.

Source	Product 6 LCAP	Unreacted 5 LCAP	Protodehalogenation side product 11 LCAP
Kessil Lamp	2.3 ± 1.1	84.1	3.2
Lumidox® Reactor	7.4 ± 1.0	70.7	7.2
BPR200	18.1 ± 1.5	45.2	16.1
New Parallel Photoreactor	43.9 ± 0.4	0	30.1

Table S3. Average product **6**, unreacted **5** and protodehalogenation side-product **11** LCAPs across 384 reactions.

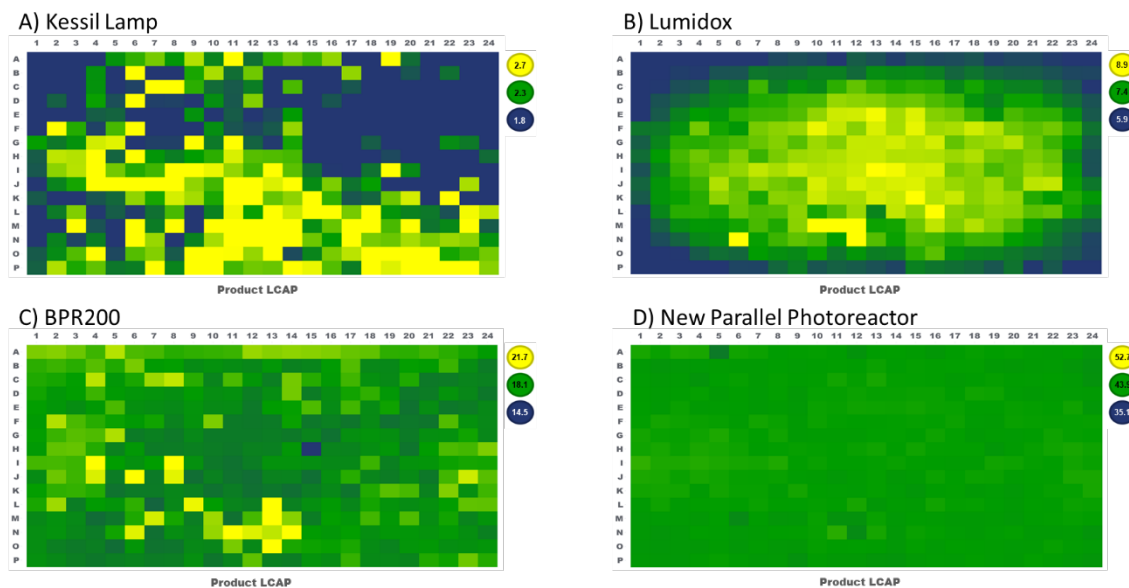A) Kessil Lamp   B) Lumidox   C) BPR200   D) New Parallel Photoreactor

Figure S7. Heat maps of average product LCAP ± 20% illustrating edge effects and hot spots for the various photoreactor designs. A) Kessil Lamp B) Lumidox® I reactor C) BPR200 and D) New Parallel Photoreactor

In comparison to the Kessil lamp and commercial photoreactors, the new parallel photoreactor generated the highest amount of desired product with minimal to no hot spots and edge effects. Refer to "Full Dataset 3.1.4. Photoreactor validation and intensity studies" file attached for detailed LCAP across each well.

3.2 Intensity studies using the new photoreactor.

The goal of this experiment was to investigate the effect of light intensity on reactivity using three
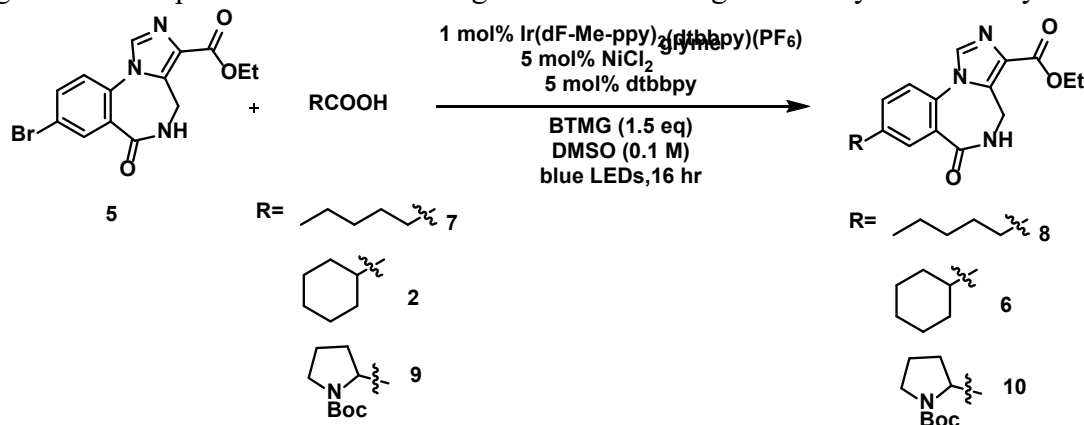
different classes of carboxylic acids.

3.2.1 Chemistry set-up (100 nmol scale, 1 μL volume).

Stock solutions containing all of the reaction components were made as follows:

Stock 1) Aryl bromide **5** (1 equiv), *n*-hexanoic acid **7** (1.5 equiv), $NiCl_2$ glyme (0.05 equiv), 4,4′-Di-tert-butyl-2,2′-dipyridyl (0.05 equiv), Ir(dF-Meppy$_2$)(dtbbpy)(PF$_6$) (0.01 equiv) and BTMG (1.5 equiv) in 0.1 M DMSO.

Stock 2) Aryl bromide **5** (1 equiv), cyclohexanecarboxylic acid **2** (1.5 equiv), $NiCl_2$ glyme (0.05 equiv), 4,4′-Di-tert-butyl-2,2′-dipyridyl (0.05 equiv), Ir(dF-Meppy$_2$)(dtbbpy)(PF$_6$) (0.01 equiv) and BTMG (1.5 equiv) in 0.1 M DMSO.

Stock 3) Aryl bromide **5** (1 equiv), *N*-Boc-L-proline **9** (1.5 equiv), $NiCl_2$ glyme (0.05 equiv), 4,4′-Di-tert-butyl-2,2′-dipyridyl (0.05 equiv), Ir(dF-Meppy$_2$)(dtbbpy)(PF$_6$) (0.01 equiv) and BTMG (1.5 equiv) in 0.1 M DMSO.

These stock solutions were dispensed into a 384-well source plate (see source plate map below), which was placed onto the Mosquito® LV deck. The Mosquito® LV robot was used to dose each stock solution containing the combined reaction components as 1 μL aliquots into a 1536-well plate. Once the dosing was completed, the 1536-well plate was heat sealed, placed on the acrylic bottom and capped with the aluminum top to allow light penetration and set to react for 16 hours. Six similar plates were prepared using the above method and placed on the following light intensity

settings (light intensities are derived from measured optical power (mW) obtained with a 26 mm sensor photometer) using the parallel photoreactor.

Plate	Measured Optical Power (mW)	Light Intensity (mW/cm²)
1	31	5.8
2	63	11.7
3	125	23.5
4	250	47.1
5	500	94.2
6	1000	188

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
1	Stock 1	Stock 2	Stock 3																					
2	Stock 1	Stock 2	Stock 3																					
3	Stock 1	Stock 2	Stock 3																					
4	Stock 1	Stock 2	Stock 3																					
5	Stock 1	Stock 2	Stock 3																					
6	Stock 1	Stock 2	Stock 3																					
7	Stock 1	Stock 2	Stock 3																					
8	Stock 1	Stock 2	Stock 3																					
9	Stock 1	Stock 2	Stock 3																					
10	Stock 1	Stock 2	Stock 3																					
11	Stock 1	Stock 2	Stock 3																					
12	Stock 1	Stock 2	Stock 3																					
13	Stock 1	Stock 2	Stock 3																					
14	Stock 1	Stock 2	Stock 3																					
15	Stock 1	Stock 2	Stock 3																					
16	Stock 1	Stock 2	Stock 3																					
Dose Vol. (nL)	1000	1000	1000																					

3.2.2 Reaction work-up.
Refer to Section 3.1.2.

### 3.2.3 Analytical.
Refer to Section 3.1.3.

3.2.4 Results and discussion.
In general, higher light intensity favors increasing product formation, with cyclohexanecarboxylic acid and *N*-Boc L-proline giving better reactivity over *n*-hexanoic acid.
Refer to "Full Dataset 3.2.4. Intensity studies using the new parallel photoreactor" file attached for detailed LCAP across each well.

3.3 Dual metal catalyst loading studies.
The goal of this experiment was to investigate the effect of photocatalyst and metal loading ratios across three different classes of carboxylic acids using the optimal light intensity.
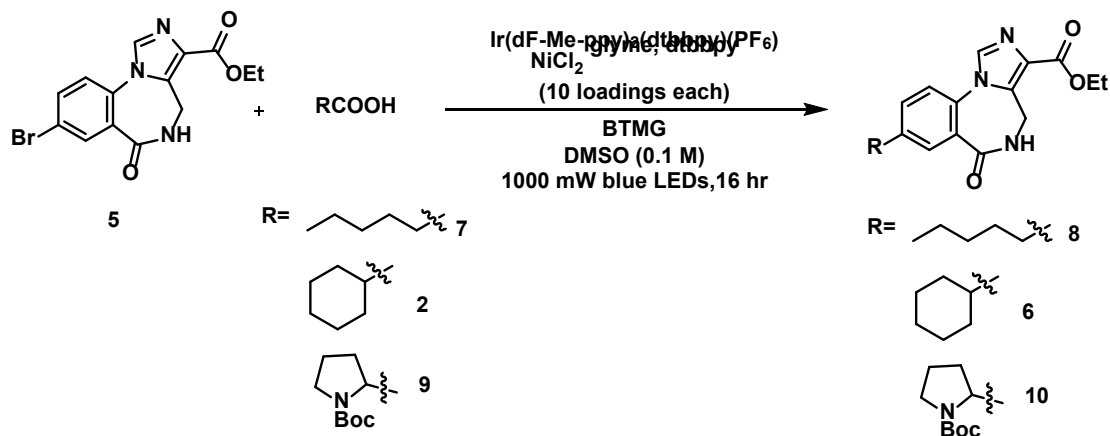
3.3.1 Chemistry set-up (100 nmol scale, 1 μL volume).

In the glovebox, stock solutions containing each of the reaction components were made as follows:

Ir(dF-Me-ppy2)(dtbbpy)(PF6) This stock solution (0.32 M in DMSO, 0.16 equiv) was diluted through nine serial dilutions, halving the concentration each dilution to make a total of ten stock solutions.

(1) 0.32 M in DMSO, 0.16 equiv, 16%
(2) 0.16 M in DMSO, 0.08 equiv, 8%
(3) 0.08 M in DMSO, 0.04 equiv, 4%
(4) 0.04 M in DMSO, 0.02 equiv, 2%
(5) 0.02 M in DMSO, 0.01 equiv, 1%
(6) 0.01 M in DMSO, 0.005 equiv, 0.5%
(7) 0.005 M in DMSO, 0.0025 equiv, 0.25%
(8) 0.0025 M in DMSO, 0.0013 equiv, 0.13%
(9) 0.0013 M in DMSO, 0.00063 equiv, 0.063%
(10) 0.00063 M in DMSO, 0.00031 equiv, 0.031%

NiCl2 glyme (0.08 M in DMSO, 0.2 equiv) and 4,4′-Di-tert-butyl-2,2′-dipyridyl (0.08 M in DMSO, 0.2 equiv) were pre-aged for 15 minutes. This stock solution was diluted through nine serial dilutions, halving the concentration each dilution to make a total of ten stock solutions.

(1) 0.08 M in DMSO, 0.2 equiv, 20%
(2) 0.04 M in DMSO, 0.1 equiv, 10%
(3) 0.02 M in DMSO, 0.05 equiv, 5%
(4) 0.01 M in DMSO, 0.025 equiv, 2.5%
(5) 0.005 M in DMSO, 0.0125 equiv, 1.25%
(6) 0.0025 M in DMSO, 0.00625 equiv, 0.625%
(7) 0.00125 M in DMSO, 0.00312 equiv, 0.312%
(8) 0.000625 M in DMSO, 0.0016 equiv, 0.156%
(9) 0.0003125 M in DMSO, 0.0008 equiv, 0.08%
(10) 0.000156 M in DMSO, 0.0004 equiv, 0.04%

Aryl bromide **5** (0.25 M in DMSO, 1 equiv)

n-Hexanoic acid **7** (0.5 M in DMSO, 1.5 equiv) and BTMG (0.67 M in DMSO, 1.5 equiv)

Cyclohexanecarboxylic acid **2** (0.5 M in DMSO, 1.5 equiv) and BTMG (0.67 M in DMSO, 1.5 equiv)

N-Boc-L-proline **9** (0.5 M in DMSO, 1.5 equiv) and BTMG (0.67 M in DMSO, 1.5 equiv)

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	
1																		
2																		
3																		
4	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(20%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
5	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(10%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
6	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(5%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
7	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(2.5%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
8	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(1.25%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
9	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(0.625%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
10	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(0.312%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
11	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(0.156%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
12	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(0.08%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
13	S1	DMSO	Ir(0.031%)	Ir(0.063%)	Ir(0.125%)	Ir(0.25%)	Ir(0.5%)	Ir(1%)	Ir(2%)	Ir(4%)	Ir(8%)	Ir(16%)	Ni(0.04%))	DMSO	S4/BTMG	S2/BTMG	S5/BTMG	
14																		
15																		
16																		
Dose Vol. (nL)	400	5						50					250	5		300		

These stock solutions were dispensed into a 384-well source plate (see source plate map above), which was placed onto the Mosquito® LV deck. The Mosquito® LV robot was used to dose each reagent/mixture component via multi-aspiration mode into a 1536-well plate. The dosing order was as follow: Aryl bromide **5** (400 nL), Ir photocatalyst (50 nL), Nickel-ligand mixture (250 nL) and carboxylic acid-BTMG mixture (300 nL).

3.3.2 Reaction work-up.
Refer to Section 3.1.2.

3.3.3 Analytical.
Refer to Section 3.1.3.

3.3.4 Results and discussion.
Refer to "Full Dataset 3.3.4. Dual metal catalyst loading study using the new parallel photoreactor" file attached for detailed LCAP across each well.

3.4 Reproducibility studies on a 1536-well plate.
The goals of this study were twofold: through setting up reactions in 384 replicates, we could investigate the reproducibility of the nano photoredox workflow. In addition, the material from these reactions could be combined and purified for downstream assays. Hits from all three classes of carboxylic acids were selected to be run in replicates.

3.4.1 Chemistry set-up (100 nmol scale, 1 μL volume).
Three stock solutions, one for each carboxylic acid, containing all of the reaction components were made as follows:

Stock 1) Aryl bromide **5** (1 equiv), *n*-hexanoic acid **7** (1.5 equiv), NiCl$_2$ glyme (0.05 equiv), 4,4′-Di-tert-butyl-2,2′-dipyridyl (0.05 equiv), Ir(dF-Meppy$_2$)(dtbbpy)(PF$_6$) (0.08 equiv) and BTMG (1.5 equiv) in 0.1 M DMSO.

Stock 2) Aryl bromide **5** (1 equiv), cyclohexanecarboxylic acid **2** (1.5 equiv), NiCl$_2$ glyme (0.05 equiv), 4,4′-Di-tert-butyl-2,2′-dipyridyl (0.05 equiv), Ir(dF-Meppy$_2$)(dtbbpy)(PF$_6$) (0.00125 equiv) and BTMG (1.5 equiv) in 0.1 M DMSO.

Stock 3) Aryl bromide **5** (1 equiv), *N*-Boc-L-proline acid **9** (1.5 equiv), NiCl$_2$ glyme (0.1 equiv), 4,4′-Di-tert-butyl-2,2′-dipyridyl (0.1 equiv), Ir(dF-Meppy$_2$)(dtbbpy)(PF$_6$) (0.00125 equiv) and BTMG (1.5 equiv) in 0.1 M DMSO.

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
1	Stock 1	Stock 2	Stock 3																					
2	Stock 1	Stock 2	Stock 3																					
3	Stock 1	Stock 2	Stock 3																					
4	Stock 1	Stock 2	Stock 3																					
5	Stock 1	Stock 2	Stock 3																					
6	Stock 1	Stock 2	Stock 3																					
7	Stock 1	Stock 2	Stock 3																					
8	Stock 1	Stock 2	Stock 3																					
9	Stock 1	Stock 2	Stock 3																					
10	Stock 1	Stock 2	Stock 3																					
11	Stock 1	Stock 2	Stock 3																					
12	Stock 1	Stock 2	Stock 3																					
13	Stock 1	Stock 2	Stock 3																					
14	Stock 1	Stock 2	Stock 3																					
15	Stock 1	Stock 2	Stock 3																					
16	Stock 1	Stock 2	Stock 3																					
Dose Vol. (nL)	1000	1000	1000																					

These stock solutions were dispensed to a 384-well source plate (see source plate map above), which was placed onto the Mosquito® LV deck. The Mosquito® LV robot was used to dose each stock solution in 1 μL aliquots into 384 wells of a 1536-well plate across every other row and column. Once the dosing was completed, the 1536-well plate was heat sealed, placed on the acrylic bottom and capped with the aluminum top to allow light penetration and set to react for 16 hours. Three plates were prepared using the above method. The first plate had 384 replicates of stock solution S1, irradiated at 500 mW. The second plate had 384 replicates of stock solution S2, irradiated at 500 mW. The third plate had 384 replicates of stock solution S3, irradiated at 250 mW. Light intensities are derived from measured optical power (mW) obtained with a 26 mm sensor photometer.

3.4.2. Reaction work-up
After 16 hr, each reaction plate was placed on the Matrix liquid handling robot together with a 384-well plate containing 24 μL of DMSO stock solution of acetic acid (1%). From the 384-well plate, the Matrix sampled 4 μL of the solution into the 1536-well reaction plate. After which, 5 μL mixture from the reaction plate was transferred back into the 384-well plate. This process was repeated three times, equivalent to a 25-fold dilution. Using a multi-channel pipette, the quenched reaction mixture was combined and transferred into a 40 mL vial. A total of three vials were prepared in this format, one for each carboxylic acid. Analytical samples were prepared by taking a 25 μL aliquot of the quenched mixture and diluting into 175 μL DMSO. The remaining samples were chromatographed using reverse phase C18 HPLC to afford the compounds (Table S4).

3.4.3 Analytical.
Refer to Section 3.1.3.

3.4.4 Results and discussion.

| Acid	Nanoscreen 0.1 μmol	384 Replicates 0.1 μmol	Isolated Yield (mass, %)
n-Hexanoic	26	34	1.5 mg (32%)
Cyclohexanecarboxylic	53	49	4.5 mg (65%)
N-Boc-L-proline	80	88	12.1 mg (79%)

Table S4. Reproducibility study results showing comparisons between nanoscreen, 384 nanoscale replicates and isolated yield.

4. Compound characterization

8
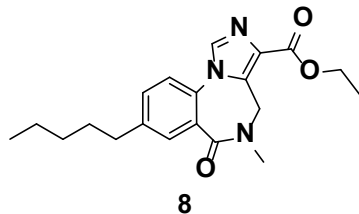
Colorless oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.38 (s, 1H), 7.72 (d, J = 2.0 Hz, 1H), 7.66 (d, J = 8.2 Hz, 1H), 7.59 (dd, J = 8.3, 2.1 Hz, 1H), 4.98 (s, 1H), 4.47 (s, 1H), 4.32 (s, 2H), 3.11 (s, 3H), 2.75 – 2.67 (m, 2H), 1.63 (p, J = 7.8 Hz, 2H), 1.40 – 1.29 (m, 7H), 0.89 (t, J = 7.0 Hz, 3H). ^{13}C NMR (126 MHz, DMSO-d_6) δ 162.78, 143.13, 136.81, 135.89, 133.06, 131.61, 130.17, 128.81, 123.23, 60.57, 42.50, 35.55, 34.79, 31.29, 30.83, 22.40, 14.70, 14.38. MS: 356.28 (M+1).


^{1}H NMR in DMSO-d6
Compound **8**

^{13}C NMR in DMSO-d6
Compound **8**
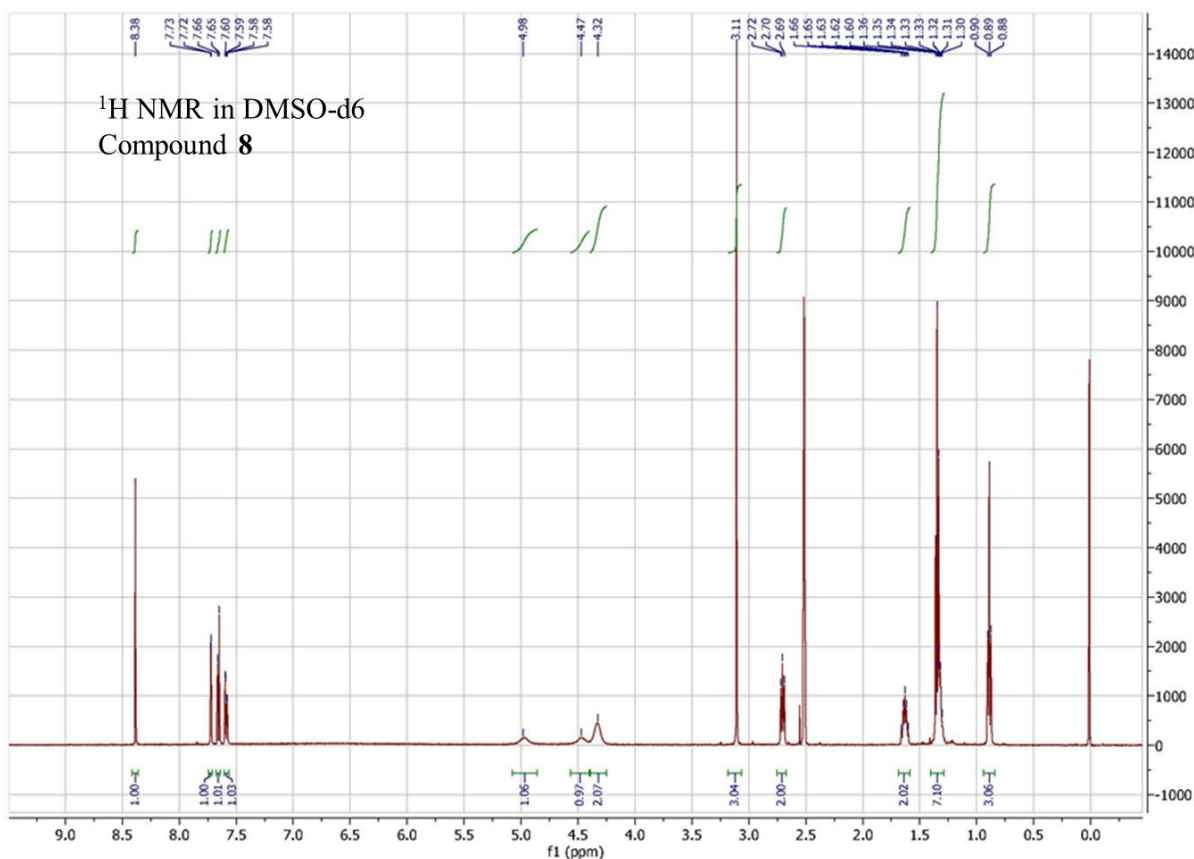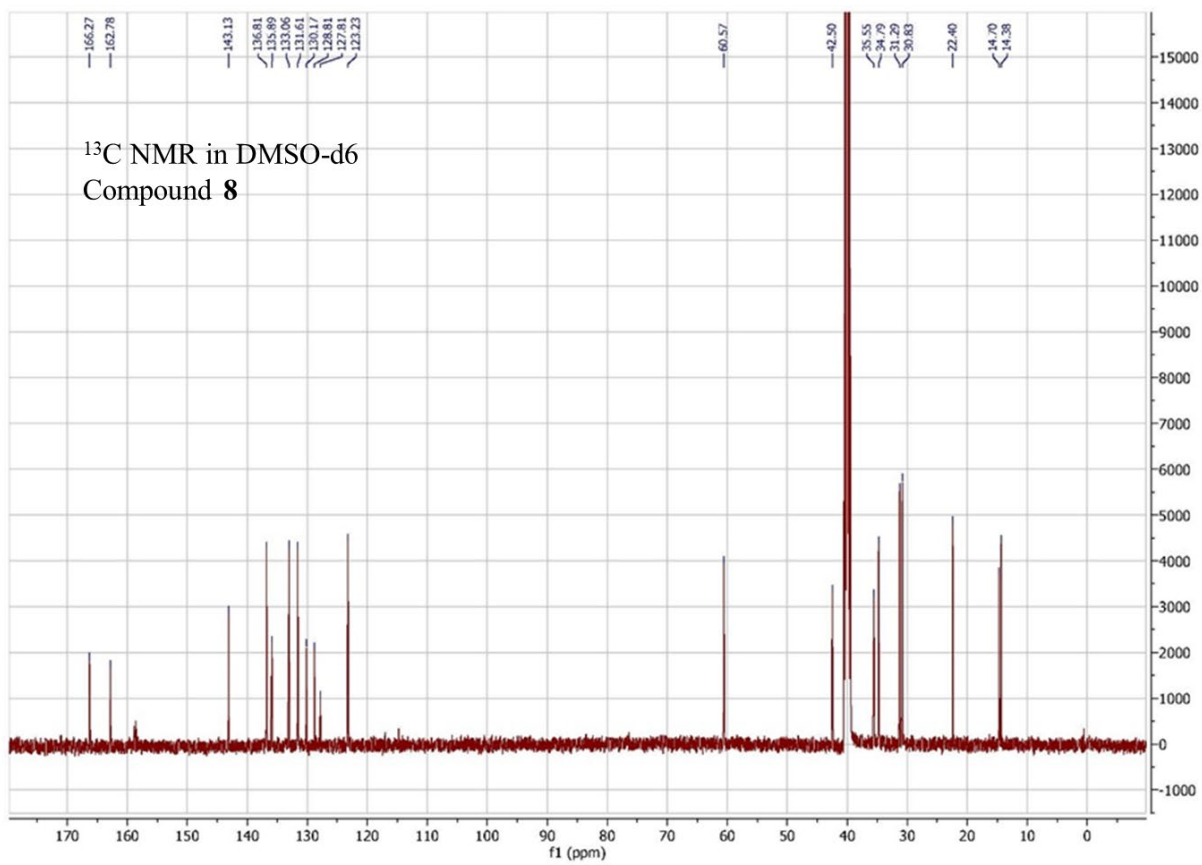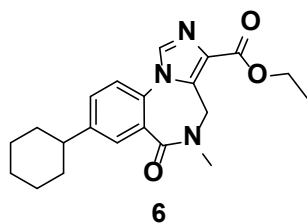
Colorless oil. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.35 (s, 1H), 7.75 (d, J = 2.0 Hz, 1H), 7.66 (d, J = 8.3 Hz, 1H), 7.62 (dd, J = 8.3, 2.0 Hz, 1H), 4.97 (s, 1H), 4.48 (s, 1H), 4.32 (s, 2H), 3.11 (s, 3H), 2.67 (m, 1H), 1.89 – 1.79 (m, 4H), 1.73 (d, J = 12.6 Hz, 1H), 1.53 – 1.37 (m, 4H), 1.35 (t, J = 7.1 Hz, 3H), 1.32 – 1.25 (m, 1H). ^{13}C NMR (126 MHz, DMSO-d_6) δ 166.32, 162.86, 148.05, 135.88, 131.60, 130.32, 130.05, 128.83, 123.29, 60.53, 43.54, 42.51, 35.58, 26.68, 25.90, 14.71. MS: 368.30 (M+1).

13C NMR in DMSO-d6
Compound **6**

10
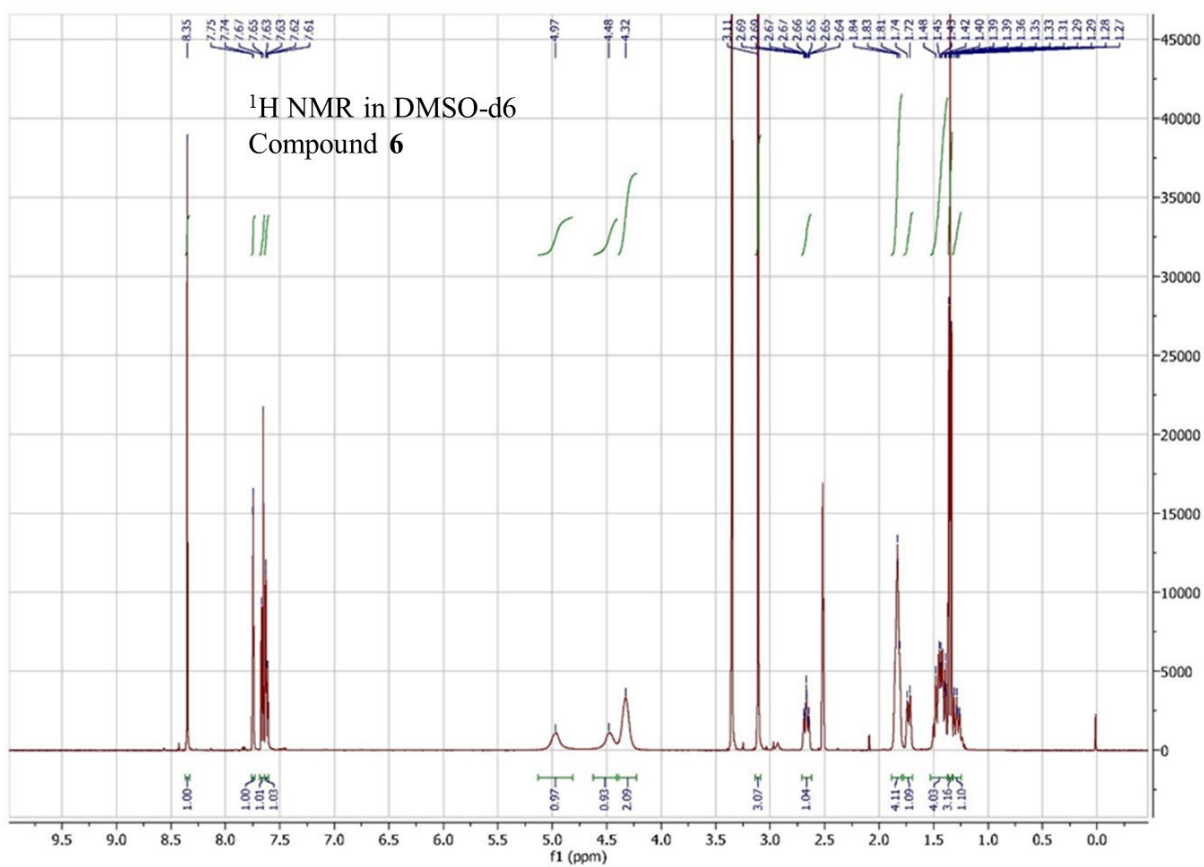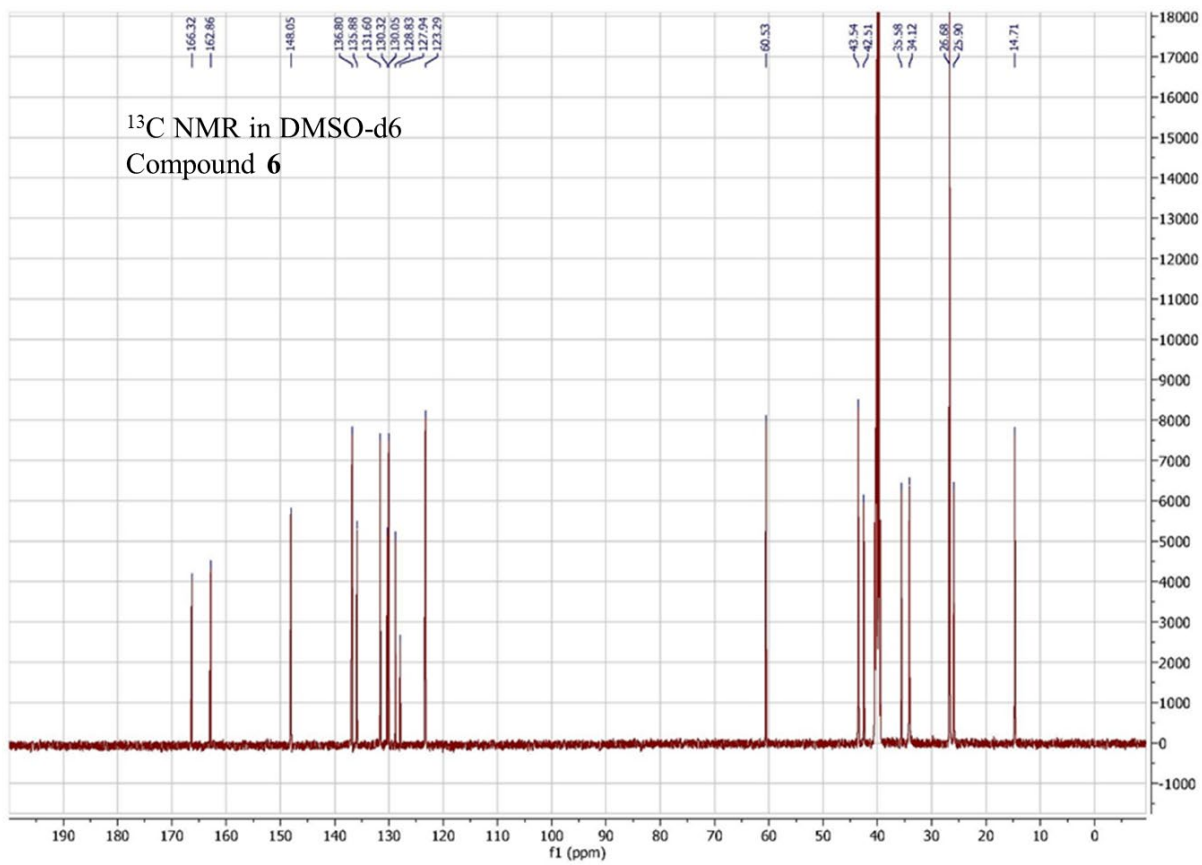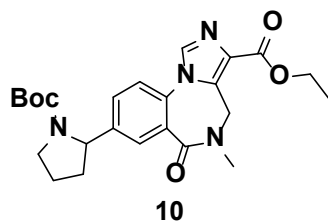
White solid. ¹H NMR (500 MHz, DMSO-d_6) δ 8.38 (d, J = 7.3 Hz, 1H), 7.76 – 7.66 (m, 2H), 7.56 (dd, J = 20.8, 7.5 Hz, 1H), 4.90 (m, 2H), 4.33 (s, 3H), 3.54 (m, 2H), 3.11 (s, 3H), 2.46 – 2.27 (m, 1H), 1.93 – 1.67 (m, 3H), 1.42 (s, 4H), 1.35 (t, J = 7.1 Hz, 3H), 1.15 (s, 5H). ¹³C NMR (126 MHz, DMSO-d_6) δ 166.18, 162.84, 136.85, 135.85, 130.59 (d, J = 54.7 Hz), 123.25, 79.17, 60.35 (d, J = 52.2 Hz), 42.53, 35.74 (d, J = 48.4 Hz), 34.81, 28.32, 23.53 (d, J = 26.5 Hz), 14.70. MS: 455.40 (M+1).

¹H NMR in DMSO-d6
Compound **10**

^{13}C NMR in DMSO-d6
Compound **10**
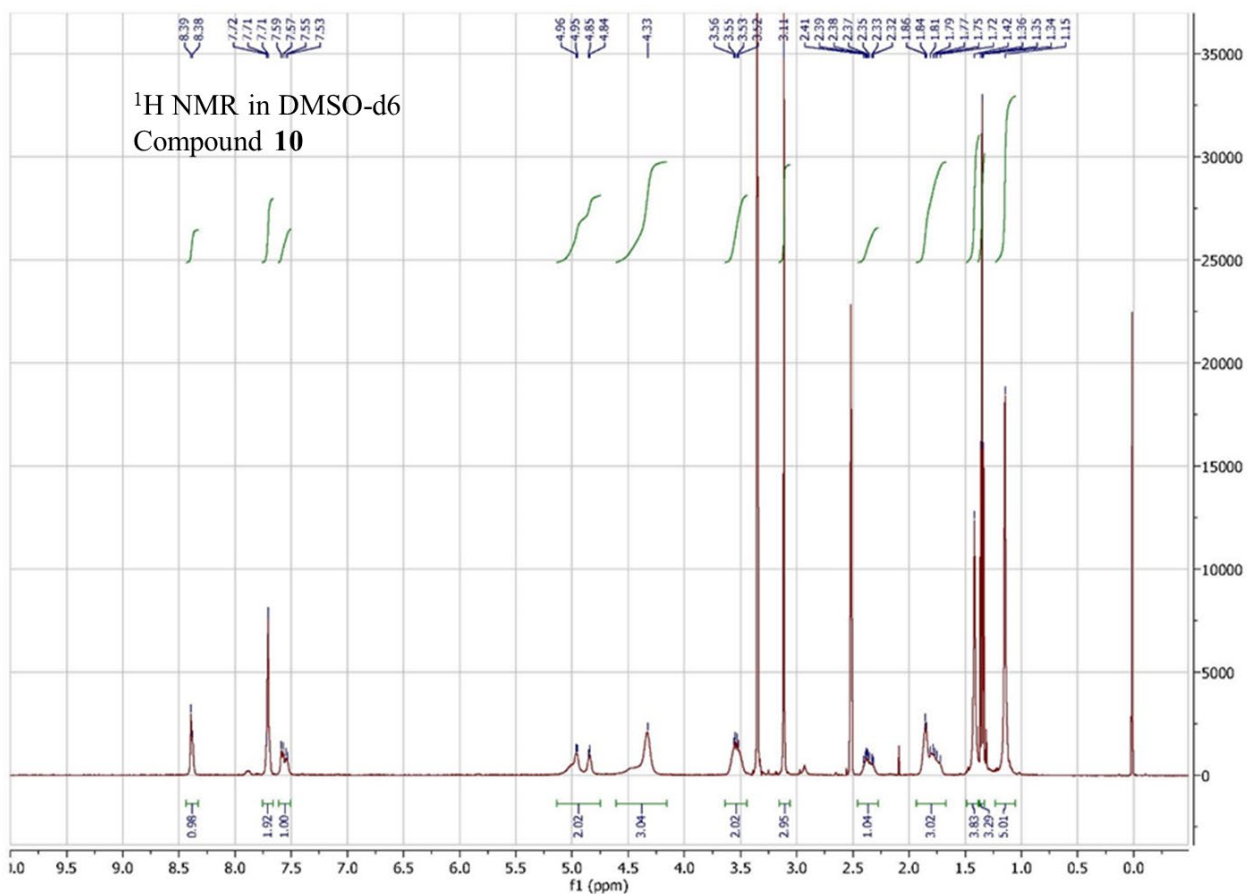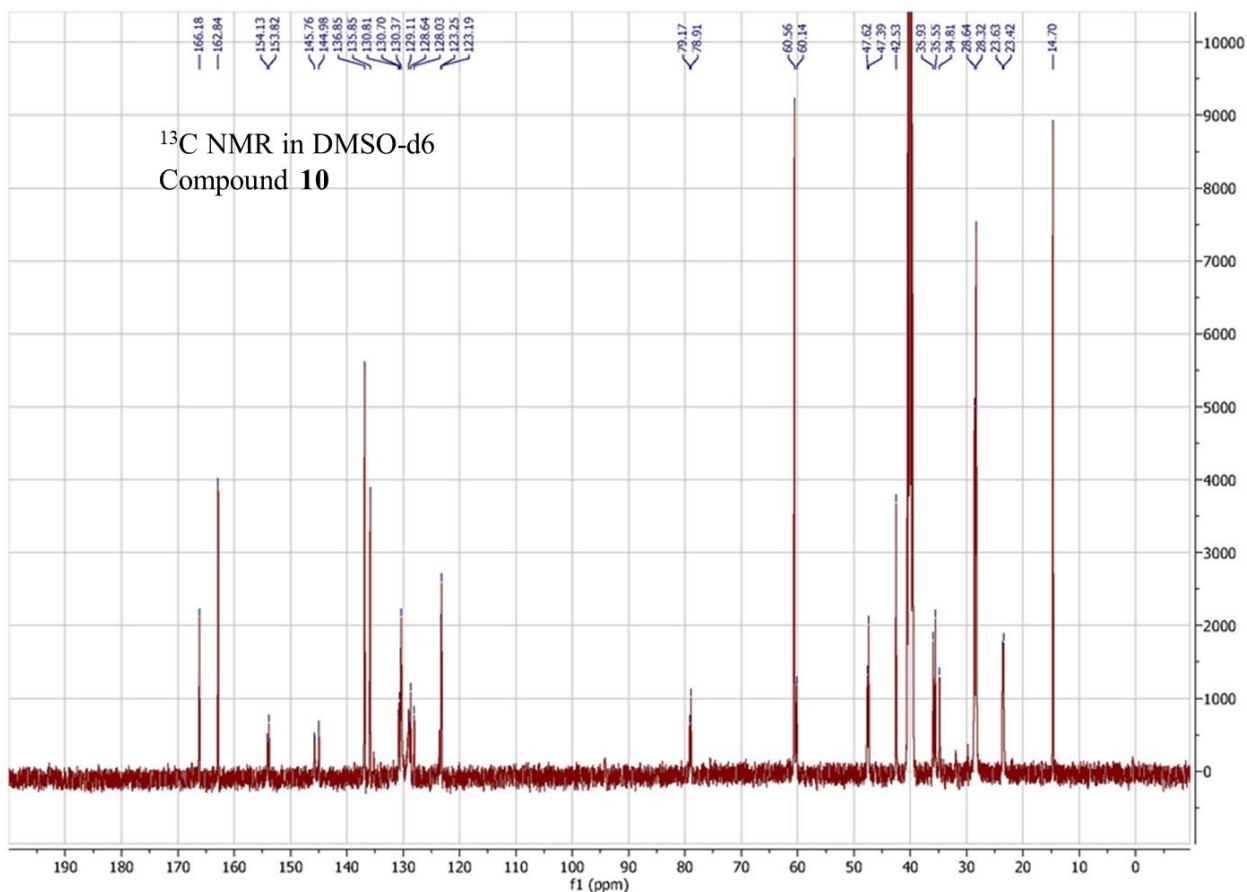
5. References

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