UNIVERSITY OF CALGARY | Program for Undergraduate Research Experience (PURE)

FINAL REPORT AND REFLECTION

SYNTHESIS OF 1) DAP CORE ANION DETECTORS AND 2) HALOGENTED CYANINE DYES

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Project 1:

Introduction

Anions have various roles of importance in biological systems and industrial processes and can also be unwanted resultants of various processes, especially in industry. Anions can be detected via several approaches, but a popularized way is with a small molecule that can act as a chemodosimeter which, via chemical pathways, interacts with anions in selective manners to elicit measurable responses. Certain chemodosimeters rely on anion-π interactions, where the electron deficient small molecule interacts with an anion resulting in an electron transfer (ET) or anion-induced polarization that radicalizes the anion detector, often resulting in a physical change (i.e. colour change).[1] These radicalized anion detectors can sometime undergo further radicalization, causing additional physical changes. This mechanism of multiple points of radicalization can be useful for determining quantities of anion in a sample. A specific anion-π interaction of interest is that of anion-diazapyrene (DAP) interactions which can be modified to elicit measurable responses to specifec anions. Changing the π-acidity of the DAP through different substitutions on the aromatic core and use of various linkers can adjust the strength and extent of interactions between anions and DAP molecules.[2] This in turn allows for the alteration of its selectivity so that it can be used to detect specific anions, or specific subsets of anions. The alteration of DAP for the purpose of detecting anions is therefore of potential great use in industrial and biological atmospheres as it can be used as a detection method for biproducts via a physical change when in the presence of specific DAP complexes. The research involved in this project focused on the substitution of N-methyl-2,7-diazapyrene to different aromatic cores for the purpose of anion detection. If time allowed, the synthesized anion detector compounds would have been tested with various anions via NMR titrations to determine sensitivity and potential selectivity.

Experimental

General Information: The $N_{2(g)}$ system for reactions was maintained using a Schlenk apparatus, cooled with dry ice. All ^{1}H NMR were obtained using the Bruker AVIII-400.

N,N'-dimethyl-1,4,5,8-naphthalenetetracarboxylic diimide (NDI) (1): In a 25mL round bottom flask (RBF), with a stir bar, 1,4,5,8-naphthalenetetracarboxylic dianhydride (NDA) (2.003g, 7.5mmol) and 40% methylamine (60mL) was refluxed at 120℃ for 24 hours. The mixture was returned to room temperature (RT, 20℃) and the purple-pink precipitate (1.793g, 6.1mmol, 82%) was vacuum filtered, washing with H_2O and methanol (MeOH) until washings were colourless.^{1}H NMR ($CDCl_3$) δ 8.78 (s, 4H), 3.61 (s, 6H)

1,3,6,8-tetrahydro-2,7-dimethylbenzol-3,8-phenanthroline (2): A 250mL RBF with a stir bar was flame dried and introduced to an N_2 environment, maintained throughout the reaction. $LiAlH_4$ (2.420g, 27.5mmol) and $AlCl_3$ (1.309g, 9.8mmol) was added to the RBF. Via cannula transfer, dry tetrahydrofuran (THF) (120mL) was added to the RBF and stirred, followed by small additions of **1** (1.793g, 6.1mmol). The RBF was fitted with a condenser and refluxed, under N_2, at 140℃ for 18 hours. The reaction mixture was poured, portion wise, into a cooled 500mL Erlenmeyer containing 130mL THF and 25g ice. The resulting brown precipitate was filtrated, washing with THF and ether, and allowed to dry for 4 hours. The dried brown solid was ground with a mortar and pestle, and

transferred to a 250mL RBF, with a stir bar, and 120mL of $CHCl_3$. This mixture was refluxed, no longer under N_2, for 18hrs and vacuum filtered. The brown solid was returned to the 250mL RBF and refluxed with an additional 120mL of $CHCl_3$ for 5 hours and then vacuum filtered. The filtrates were combined and dried via rotary evaporation to yield a dark brown powder which was recrystallized using minimal volumes of chlorobenzene to yield a fluffy yellow-gray powder (0.662g, 2.8mmol, 46%). ^{1}H NMR ($CDCl_3$) δ 7.12 (s, 4H), 3.88 (s, 8H), 2.57 (s, 6H)

2,7-diazapyrene (DAP) (3): In a 25mL RBF, **2** (0.620g, 2.6mmol) and selenium (1.222g, 15.5mmol) were combined and mixed. The RBF was placed in a heating mantle surrounded with sand, glass wool and covered in tin foil and then heated for 18hours at ~400℃. The black solid was extracted via manual scraping and 6M HCl washings and transferred to a 250mL Erlenmeyer flask, with a stir bar, and heated. The washings were vacuum filtrated, and the filtrate was then subsequently vacuum filtrated a second time to remove any residual selenium. To the 2nd filtrate, with cooling, $KOH_{(s)}$ was added until a creamy tan precipitate (0.364g, 1.8mmol, 68%) crashed out of solution which was then vacuum filtered and washed with H_2O until washings were colourless. ^{1}H NMR ($CDCl_3$) δ 9.52 (s, 4H), 8.23 (s, 4H)

N-methyl-2,7-diazapyrenium iodide (4): In a 50mL RBF, with a stir bar, **3** (301mg, 1.5mmol) was dissolved in $CHCl_3$ (30mL) and capped with a rubber septum. Iodomethane (0.91mL, 14.6mmol) was added through the septum, dropwise, and stirred at RT for 24 hours. The yellow precipitate was collected via vacuum filtration and the filtrate returned to the RBF. To the filtrate, additional iodomethane (0.91mL, 14.6mmol) was added and the solution stirred for 24 hours. The golden yellow precipitate was collected via vacuum filtration and, after confirmation of its identity via ^{1}H NMR, combined with the previous yellow solid (448mg, 1.3mmol, 88%). ^{1}H NMR (D_2O) δ 9.62 (s, 2H) 9.48 (s, 2H), 8.42 (d, 2H), 8.34 (d, 2H), 4.87 (s, 3H)

o-1,2-benzyl-bis(7-methyl-2,7-diazapyrenium) dibromide diiodide (5): In a 25mL RBF, with a stir bar, **4** (50mg, 0.14mmol), α,α'-dibromo-o-xylene (18mg, 0.07mmol), and tetrabutylammonium iodide (TBAI) (27mg, 0.07mmol) were combined followed by acetonitrile (ACN) (10mL). The mixture was then refluxed at 105℃ for 70 hours. The yellow orange precipitate (40mg, 0.04mmol, 29%) was collected via vacuum filtration, while still hot, washing with hot ACN. ^{1}H NMR (D_2O) δ 10.02 (d, 8H), 8.74 (m, 8H), 7.62 (s, 2H), 7.41 (s, 2H), 6.66 (s, 4H), 4.90 (s, 6H)

2-benzyl-7-methyl-2,7-diazapyrenium bromide iodide (6): In a 25mL RBF, with a stir bar, **4** (112mg, 0.32mmol), benzyl bromide (0.38mL, 3.2mmol), and TBAI (143mg, 0.39mmol) were combined followed by ACN (15mL). The mixture was then refluxed at 120℃ for 70 hours. The light orange precipitate (129mg, 0.25mmol, 77%) was collected via vacuum filtration, while still hot, washing with hot ACN. ^{1}H NMR (D_2O) δ 10.13 (s, 2H), 10.04 (s, 2H), 8.86 (d, 2H), 8.82 (d, 2H), 7.66 (m, 2H), 7.56 (m, 3H), 6.41 (s, 2H), 4.97 (s, 3H)

m-1,3-benzyl-bis(7-methyl-2,7-diazapyrenium) dibromide diiodide (7): In a 25mL RBF, with a stir bar, **4** (151mg, 0.44mmol), α,α'-dibromo-m-xylene (54mg, 0.20mmol), and TBAI (77mg, 0.21mmol) were combined followed by ACN (30mL). The mixture was then refluxed at 105℃ for 136 hours. The red precipitate (151mg, 0.16mmol, 36%) was collected via vacuum filtration, while still hot, washing with hot ACN. ^{1}H NMR (D_2O) δ 10.15 (s, 4H) 10.05 (s, 4H) 8.86 (d, 4H) 8.82 (d, 4H) 7.95 (s, 1H) 7.73 (d, 2H) 7.66 (t, 1H) 6.45 (s, 4H) 4.98 (s, 8H)

o-1,2-benzyl-bis(7-methyl-2,7-diazapyrenium) tetrahexafluorophosphate (8): In a 5mL RBF, **5** (33mg, 0.034mmol), ammonium hexafluorophosphate (96mg, 0.59mmol) and H_2O (2mL) were combined and stirred at RT for 48 hours. The yellow orange precipitate (25mg, 0.02mmol, 64%) was collected via vacuum filtration, washing with H_2O. ^1H NMR (DMSO) δ 10.36 (s, 8H), 8.92 (s, 8H), 7.61 (d, 4H), 6.70 (s, 4H), 4.92 (s, 6H)

Project 2:

Experimental

General Information: The $N_{2(g)}$ system for reactions was maintained using a Schlenk apparatus, cooled with dry ice. All ^1H and ^{13}C NMR were obtained using the Bruker AVIII-400.

Dimethyl 3-hydroxyglutarate (1): In a 50mL RBF, dimethyl-1,3-acetonedicarboxylate (4.020g, 23mmol) was dissolved in MeOH (20mL) and cooled to 0℃. To this cooled mixture, $NaBH_4$ (290mg, 7.7mmol) was added portion wise and the mixture allowed to return to RT and subsequently stirred for 16 hours. The product was worked up with H_2O, ethyl acetate (EtOAc) and brine, and then dried over $MgSO_4$. The solution was then dried via rotary evaporation to yield a pale-yellow oil-like substance (2.624g, 14.9mmol, 65%). ^1H NMR ($CDCl_3$) δ 4.45 (m, 1H), 3.71 (s, 6H), 3.37 (d, 1H), 2.57 (d, 4H)

1,3-bis(2-benzimidazole)-2-propanol (2): In a 50mL RBF, **1** (2.624g, 14.9mmol) and o-phenylenediamine (3.220g, 29.8mmol) was dissolved in 6M HCl (25mL). The mixture was refluxed at 120℃ for 18 hours and then cooled to 0℃. The pH of the cooled solution was then adjusted to 14 using 5M NaOH, causing a yellow-tan precipitate to crash out of solution. This yellow-tan precipitate (2.463g, 8.4mmol, 57%) was collected via vacuum filtration and washed with H_2O. ^1H NMR (DMSO) δ 7.48 (m, 4H) 7.11 (m, 4H) 4.52 (m, 1H) 3.01 (dd, 4H).

1,3-bis(1-ethyl-2-benzimidazole)-2-propanol (3): In a 25mL RBF, **2** (0.300g, 1mmol) was suspended in dimethylformamide (DMF) (9mL). To the RBF, powdered KOH (1.335g, 2.4mmol) was added and dissolved over 30 mins, stirring at RT. To the RBF, ethyl iodide (0.17mL, 2.1mmol) was added and stirred at RT for 16 hours. The mixture was worked up in EtOAc and brine and then dried over $MgSO_4$. The solution was then dried via rotary evaporation. The compound was then purified via a silica column using 30:1 ($CHCl_3$:MeOH) with 1% triethylamine as an eluent system to isolate a brown oil-like substance that solidified into a tan powder (0.273g, 0.78mmol, 76%). ^1H NMR (DMSO) δ 7.54 (m, 4H), 7.18 (m, 4H), 5.44 (b, 1H), 4.64 (p, 1H), 4.30 (q, 4H), 3.32 (dd, 2H), 3.11 (dd, 2H), 1.32 (t, 6H). ^{13}C NMR (DMSO) δ 152.59, 142.34, 134.44, 121.52, 121.24, 118.32, 109.91, 68.48, 37.88, 31.14, 14.94. *m/z* calcd. for $C_{21}H_{25}N_4O$ $[M+H]^+$: 349.2023, found: 349.2031.

5,6-dibromo-2-methylbenzimidazole (4): In a 50mL RBF, 2-methylbenzimidazole (1.501g, 11mmol) was suspended in concentrated acetic acid (20mL). The mixture was brought to 60℃ and stirred, followed by portion wise additions of N-bromosuccinimide (NBS) (4.248g, 24mmol), and then stirred for 18 hours at 60℃. The mixture was cooled to RT and stirred for 2 hours until white precipitate crashed out of solution. The precipitate was collected via vacuum filtration, washing with H_2O. The white precipitate was suspended in 0.1M NaOH and sonicated for 10 minutes to remove persistent acetic acid, and then vacuum filtrated to collect (1.650g, 5.7mmol, 50%). ^1H NMR (DMSO) δ 12.49 (s, 1H), 7.89 (d, 2H), 2.48 (s, 3H)

4,7-dibromo-2-methylbenzimidazole (5): In a 10mL RBF, 1,2-diamine-3,6-dibromobenzne (0.501g, 1.9mmol) and acetic anhydride (0.20mL, 2.1mmol) were combined in 6M HCl (5mL) and refluxed at 120℃ for 20 hours. The pH of the mixture was adjusted to ~8 and worked up with EtOAc and brine, then dried over $MgSO_4$ followed by drying via rotary evaporation resulting in a brown white solid. This was purified via a silica column, using an eluent system of 5:1 (EtOAc:Hexanes), the relevant fractions were then combined and dried via rotary evaporation yielding a light orange solid (150mg, 0.52mmol, 27%). ^{1}H NMR (CD_3OD) δ 8.52 (s, 1H), 8.42 (s, 1H), 4.67 (t, 2H), 4.51 (q, 2H), 2.95 (t, 4H), 2.31 (p, 2H), 1.49 (t, 3H).

5,6-dibromo-1-ethyl-2-methylbenzimidazole (6): In a 10mL RBF, **4** (0.720g, 2.5mmol) was dissolved in DMF (4mL) followed by powdered KOH (302mg, 5.4mmol) and stirred for 2 hours at RT. To the mixture, ethyl iodide (0.22mL, 2.7mmol) was added slowly and the mixture stirred for 16 hours at RT. The mixture was then worked up with EtOAc and brine, and then dried over $MgSO_4$, the resulting solution was dried via rotary evaporation to yield a yellow fluffy powder (623mg, 2mmol, 79%). ^{1}H NMR (CD_3OD) δ 7.90 (s, 1H), 7.85 (s, 1H), 4.26 (q, 2H), 2.60 (s, 3H), 1.38 (t, 3H). ^{13}C NMR (CD_3OD) δ 154.25, 142.01, 134.74, 121.96, 116.75, 116.55, 114.30, 38.51, 13.53, 11.94. *m/z* calcd. for $C_{10}H_{10}Br_2N_2$ $[M+H]^+$: 318.9263, found: 318.9262.

5,6-dibromo-1-ethyl-2-methyl-3-(3'-sulfopropyl)-benzimidazole (7): In a 5mL RBF, **6** (200mg, 0.63mmol) and 1,3-propanesultone (60µL, 0.68mmol) were mixed in chlorobenzene (2mL) and refluxed for 19 hours. The white precipitate (218mg, 50mmol, 79%) was isolated via vacuum filtration, washing with diethyl ether. ^{1}H NMR (CD_3OD) δ 8.52 (s, 1H), 8.42 (s, 1H) 4.67 (t, 2H), 4.51 (q, 2H), 2.95 (m, 4H), 2.30 (m, 2H), 1.49 (t, 3H).

Sodium methoxide (8): To a flame dried 25mL RBF, dry MeOH (10mL) was kept under $N_{2(g)}$. $Na_{(s)}$ (196mg, 8.5mmol) was added in small portions to the mixture, re-establishing $N_{2(g)}$ environment with each addition to yield a colourless mixture (10mL, 0.8525M).

Sodium 1-ethyl-2-[3-[1-ethyl-1,3-dihydro-3-(3-sulfopropyl)-5,6-dibromo-benzimidazol-2-ylidene]-1-propen-1-yl]-3-(3-sulfopropyl)-5,6-dibromo-benzimidazolium (9): In a flame dried 25mL RBF, **7** (350mg, 0.80mmol) was suspended in dry ethanol (7mL). Iodoform (188mg, 0.48mmol) was added to the mixture and then stirred at RT for 15 minutes. **8** (2.3mL, 1.96mmol) was added and the mixture was then heated at 60℃ for 10 minutes and then returned to RT and stirred for 17 hours. The iridescent dark pink precipitate was collected via vacuum filtration, washing with ether. **Note* The dark precipitate was a crude mix of starting material, expected product and solvent signals, time leftover in the research term did not allow for purification and proper characterization.*

Conclusion

Over the course of the research term, work was split between two projects, anion sensing and dye synthesis. Due to the time required for synthesis in both projects, the only characterization that was feasible, with the given time, was NMR data and some MS data for the projects. However, with more time, NMR titrations could be explored for determining the DAP based compounds selectivity towards specific anions and potential use in industrial or biological settings, as well as synthesizing other DAP substituted compounds with different linkers. Additionally, further time could be put into synthesizing more dyes, as only one was synthesized and not even purified due to time restrictions.

References

1. Schottel B.L.; Chifotides H.T.; Dunbar K.R. *Chem. Soc. Rev.* **2008**, 37, 68-83.
2. Ramírez-Zatarain S.D., et al. *Supramol. Chem.* **2017**, 29(6), 446-454.

Experience Gained

As this is the first experience I have had with research and chemistry outside of an undergraduate academic setting, the knowledge that I have gained has been pivotal towards building myself as a chemist and researcher. Firstly, I have been introduced to various synthesis techniques and general lab procedures that otherwise I would have been inexperienced in, including $LiAlH_4$ reductions, column chromatography, ion exchanges, solid state reactions and much more. I was exposed to, and able to work with, characterization techniques, including proton and carbon NMR. The time spent in the lab was invaluable as it built my technical skills as a chemist. I learned the value in purity of compounds, especially when used in subsequent multi-step synthesis pathways. I was able to learn to communicate effectively with my team, which will surely be reflected in future endeavours and research. During the entire research term, I also was able to develop my time management skills and organizational skills as there was multiple points where I was running multiple experiments at the same time and was needing to monitor and work up these in similar time frames. I have enjoyed my time so much with the Sutherland group that I am continuining my summer research into the fall 2019 semester via a 402 program and hope to work with their group further in the future.

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