UNIVERSITY of WASHINGTON TACOMA

Synthesis of Substituted Quinazolinones with Applications Toward Chagas Disease

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INTRODUCTION

Chagas disease is a parasitic infection that impacts millions in South America regions, predominantly those of lower socioeconomic status as Trypanosoma cruzi (*T. cruzi*) proliferates in areas of poorer living conditions. Benznidazole and nifurtimox are currently the only two medications which serve as treatments for Chagas disease. In spite of the high bioavailability and effective treatment, limitations arise with usage such as toxic side effects and requirement of long regimes. Therefore, greater research is necessary for better health in both populations of endemic and non-endemic regions. As the emergence of the parasite increases with migration into the US, Canada, Western Pacific, and parts of Europe from Central America it can become fatal for many. The quinazolinone core is classified as a privileged scaffold as the substitution of many functional groups have been found. Fortunately, amide substituted quinazolinones have demonstrated halts in the T. cruzi pathway of infection.

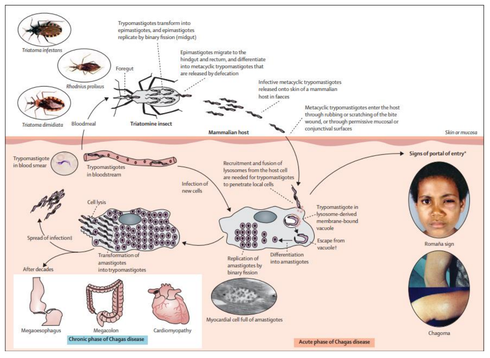
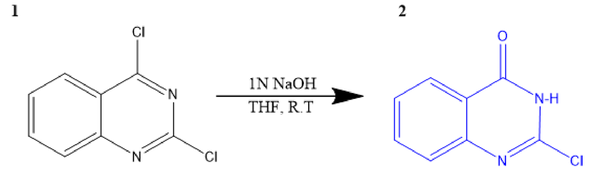


Figure 1. Life cycle of the *T. cruzi* infection pathway and associated symptoms of Chagas disease.

SYNTHESIS OF QUINAZOLINONE CORE



2-Chloroquinazolin-4(3H)-one. 2,4-Dichloroquinazolinone (1.0g, 5.0mmol) was dissolved in NaOH (10 ml, 10 mmol) and 10.5 ml tetrahydrofuran (THF) at room temperature. Reactants were combined in a round-bottom flask and stirred for 4 hours as the reaction mixture (RM) transitioned from cloudy-white to a clear solution. RM checked for full conversion through TLC verification. The TLC solvent system used was a 1:1 EA/Hex concentration. Once the reaction reached full conversion, the RM was chilled over an ice bath for 10 minutes and adjusted to pH 5 with acetic acid (AcOH). Solids were filtered and resembled a white powder. No purification techniques were utilized for the 2-Chloroquinazolin-4(3H)-one crude product.

OBJECTIVE

Goal #1: The long-term goal of the research project is to gain a better understanding on the synthesis of substituted quinazolinones in hopes of better treatments for Chagas disease.

Goal #2: Synthesize amide-substituted quinazolinones with insoluble carboxylic intermediates in high yields.

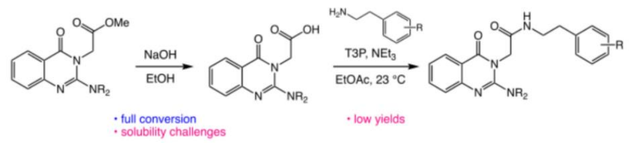
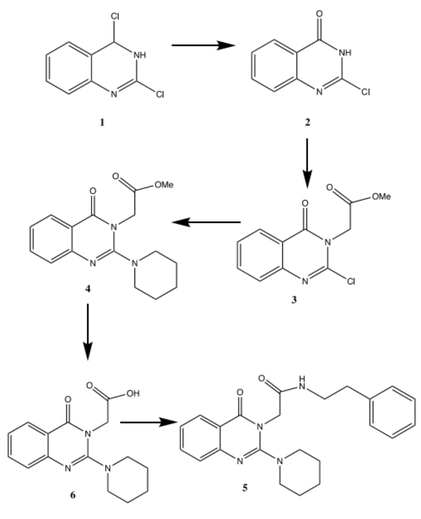


Figure 2. Current Focus for Optimization of Amide Formation. Amination followed by poor yields of the amidated product arose in Dr. Kim's prior research and served as the area of focus (Kim 2022). Our team aimed to try various extraction methods or manipulate coupling reagents to encourage higher yields.

METHODS

The synthetic route of the target compound contains a series of reactions. It begins with the hydrolysis reaction to form the quinazolinone core **2**. Second, alkylation yielding **3** then amination to replace the chlorine substituent **4**, followed by hydrolysis to yield the substituted carboxylic acid **5**, and lastly amidation to obtain the target compound **6**.

FORWARD SYNTHESIS



RESULTS

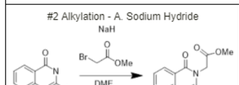
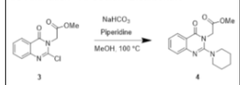
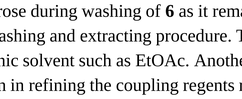
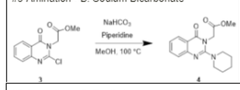
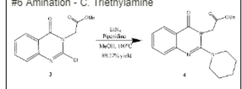
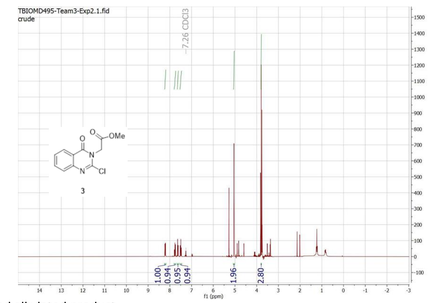
Experiment	Purification Methods	Yield	R_f (1:1 EA/Hex)
#1 Quinazolinone Synthesis 	Unpurified	126.7% (inflated pdt. yield)	0.66
#2 Alkylation - A. Sodium Hydride 	Flash column chromatography	83.8%	0.8
#3 Amination - A. Sodium Bicarbonate 	Reaction Abandoned	N/A.	0.9
#4 Alkylation - B. Sodium Carbonate 	Crude pdt.	28.5%	0.8
#5 Amination - B. Sodium Bicarbonate 	Column chromatography	9.2%	0.8
#6 Amination - C. Triethylamine 	Crude pdt.	89.3%	0.7

Table 1. The Experiments conducted. Over the span of 10 weeks 6 valuable experiments were conducted. Each experiment was analyzed by determining the percent yield and whether a purification method was used. Products were verified through TLC and H^1 NMR.

A. Crude alkylated product



B. Purified alkylated product

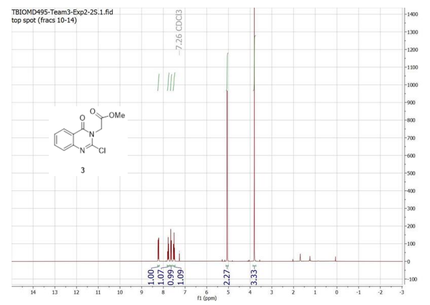


Figure 3. H^1 NMR of Alkylated Product. The purified product compares to the crude in **fig. 3A** as we see extra peaks populating around 1-2 ppm. In the crude product we expect to see these shifts in the presence of water or other solvents which reside around 1.5 ppm. The ^{1}H NMR data in **fig. 3B** for quinazolinone **3** confirmed expected signals of 1 ~8ppm for the benzene portion, signals of 2.27 at ~5ppm representative of protons between the ester and nitrogen, and 3.33 ~4ppm indicative of the protons from the methyl portion.

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- UW NMR manager for NMR product analysis.

DNDi Drugs for Neglected Diseases *initiative*

DNDi [2022]

CONCLUSION & FUTURE WORK

Previous researchers found low yields of the amide substituted quinazolinone **6** with the coupling reagents T3P, Net3 with EtOAc at 23ºC (Kim 2022). This may be due to the solubility challenges that arose during washing of **6** as it remained in the aqueous layer as opposed to the organic. Due to time constraints our team was unable to perform the amidation reaction. Although, we intended to manipulate the coupling reagents and utilize a more thorough washing and extracting procedure. The salting technique was considered, as it works to decrease the solubility to dissolve more of the compounds in the organic layer (Nichols 2016). Salting out can be used with NaCl or NH_4Cl followed by extraction with an organic solvent such as EtOAc. Another possibility would be changing the functional groups by using a less sterically hindered group such as dibutyl amine inplace of piperidine on the aminated quinazolinone **4.** Nonetheless, our team obtained valuable information in refining the coupling regents regarding quinazolinone synthesis. Future researchers can focus on other areas of synthesis rather than the regents such as NaH for the alkylation reaction instead of Na_2CO_3 to be able to focus more on the later stages of synthesis. Comparingly, in the amination reaction our researchers found that Et_3N produced a better yield than $NaHCO_3$, and therefore future researchers can focus more on the types of nitrogen-containing compounds they can attach to the core in place of piperidine. In general, our procedures can serve as a starting point so that more extensively substituted compounds can be synthesized to determine the bioactivity and their interactions in the body by biologists.

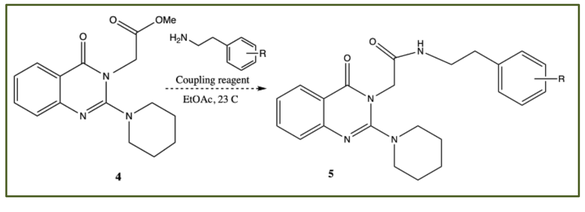

Figure 4. Future synthesis of compound 5