Synthesis of Novel Triazole-Based Antimalarial Small Molecules

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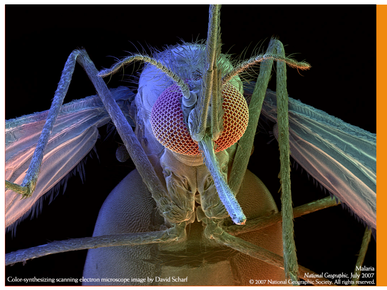
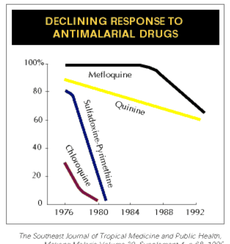


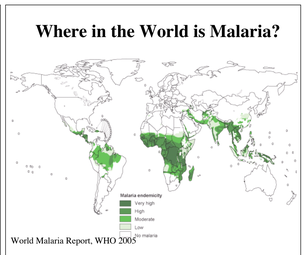
Introduction

Malaria:

Malaria has plagued humans for millennia, skillfully adapting itself for life as a parasite. Between 300 and 500 million people are infected each year with one of the four species of protozoan that cause malaria: *Plasmodium malariae, P. ovale, P. vivax, P. falciparum.* Despite the advances in modern medicine, malaria still kills 1 to 2 million people each year, most of whom are children. Moreover, malaria parasites have formed significant resistance to antimalarials in the past several decades (1).



Resistance to common antimalarials like quinine and chloroquine is increasing.



Malaria is present in over 100 countries, putting 3.2 billion people at risk of infection.

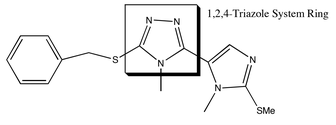
The Screening Program at the Broad:

The Broad Institute operates a high throughput screening program designed to discover novel compounds that are active against *P. falciparum*, the species of malaria responsible for 75% of all malaria cases. Over 80,000 compounds have been screened in the program's *in vitro* assay system. Of the compounds screened, nearly 200 were found to inhibit parasite growth and possess the necessary chemical scaffolds for drug design.

Active Compounds:

In collaboration with Genzyme, active compounds, or "hits," were screened at their Waltham site. Compounds must both be active against *P. falciparum* and have low cytotoxicity in human cell lines for drug development to continue. One compound that meets both of these requirements is a triazole-based molecule that had not previously been known to possess antimalarial attributes. To demonstrate the efficacy of the triazole-based compounds, seven similar commercially available compounds were screened. The results showed that these compounds require a 3-(imidazolyl)triazole motif for activity against *P. falciparum.*

The "Hit"

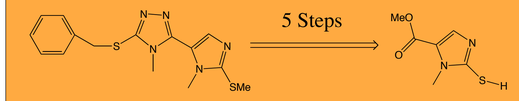


This project is designed to develop a synthetic pathway by which this molecule may be produced. Once established, this pathway will facilitate biological testing of this molecule and its analogs.

Results

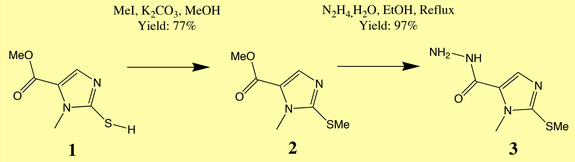
Retrosynthetic Analysis

Using the principle of retrosynthetic analysis, we worked backwards from the desired product and determined a possible synthetic pathway as well as a commercially available starting material.



Active Compound — Commercially Available Compound

The Synthetic Pathway: Steps 1 and 2



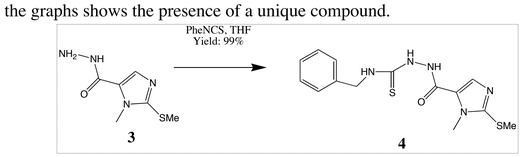
1. The first step of the synthesis was based on an established process for methylating a similar compound (2). LCMS and NMR analysis showed the reaction went to completion. Extraction in ethyl acetate isolated the product.
2. Using a method established in literature (3), **2** was reacted with hydrazine hydrate in refluxing ethanol. Excess hydrazine hydrate was evaporated.

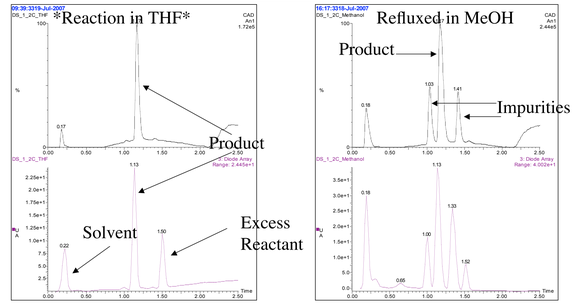
The Synthetic Pathway: Step 3 Optimization of Reaction Conditions

Although the first two reactions in the chemical pathway produced the product at high yield, the reaction from substance **3** to **4** and **5** required optimization of the process.

Synthesis of **4**

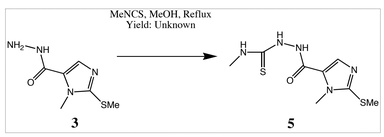
Four different reaction conditions were tried to optimize the production of **4**. Refluxing reactions were performed in EtOH, MeOH and DMF. The reaction that proved best was done in THF at room temperature. The graphs show two of the reactions, each analyzed with chromatography techniques. Each peak on the graphs shows the presence of a unique compound.

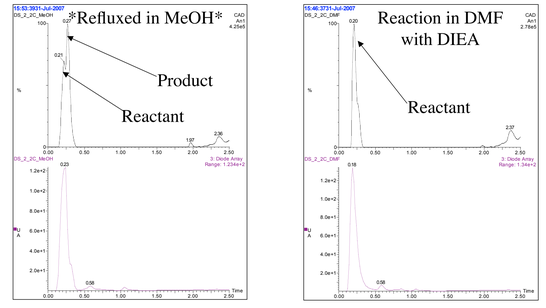




Synthesis of **5**

Four different reaction conditions were tried to optimize the production of **5**. Compound **3** was reacted in DMF with DIEA (an organic base) and in EtOH with KOH. It was also reacted without base in THF and refluxed in MeOH. The refluxing reaction proved most effective.

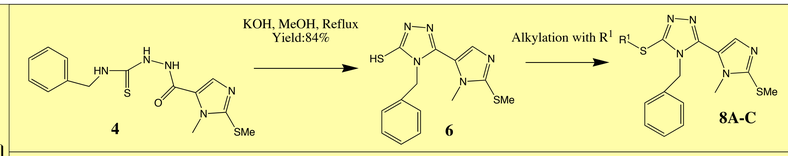


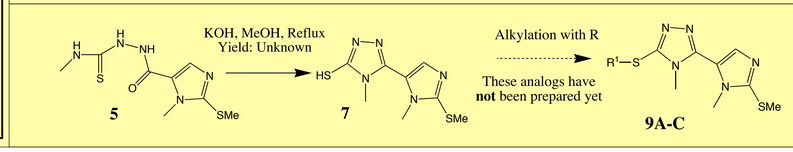


The Synthetic Pathway: Steps 4 and 5 Analog Synthesis

Variable Groups for 8A-C

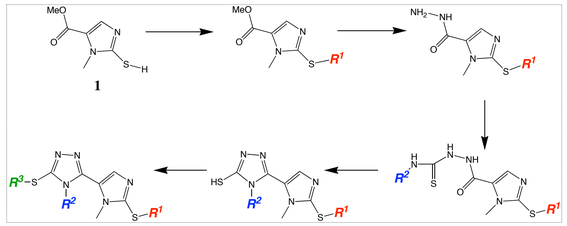
- **A:** R^1= **Me** (MeI, K_2CO_3, MeOH) Yield: 81%
- **B:** R^1= **CH_2Ph** (Benzyl-bromide, MeOH) Yield: Unknown
- **C:** R^1= (Bromomethylnaphthalene, MeOH) Yield: Unknown

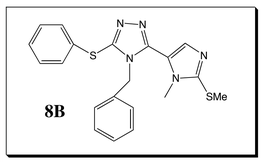




Conclusions

This project has successfully produced a viable pathway for production of the original "hit." Moreover, the synthetic pathway provides three points of diversity for the synthesis of analogs.





•Analogs **8A-C** have been successfully completed. Analog **8B** (pictured left) is pure, while the other two are undergoing purification.

•Analogs **9A-C** have not been synthesized yet, but with most of the methodology in place, their synthesis is imminent.

Future Research

•Once the analogs have been purified, the compounds will be put through *in vitro* assays in which their efficacy against *P. falciparum* will be measured.

•In parallel, Genzyme will conduct testing to assess the pharmacokinetic properties of the molecules.

•The structure activity relationships (SAR) of these molecules can then be analyzed.

•Synthesis of different analogs based on this data can begin.

•If efficacy warrants, then the molecules will continue toward drug development.

Literature Cited

1. *Kumar, Sanjai. Global Problem of Malaria, Biology of Malaria Parasites and Implications for Transfusion-Transmitted Malaria and Detection Methods. Center for Biologics Research and Review. Food and Drug Administration. Malaria Workshop. July 12, 2006.*
2. *O'Connell, John F. et al. "Convenient Synthesis of Methyl 1-Methyl-2,4-dibromo-5-imidazolecarboxylate." Department of Chemistry, University of California, Berkeley, CA 94720, USA. October 1988.*
3. *Sarhan, Abd El-Wareth A.O. "On the Synthesis and Reactions of Indole-2-carboxylic Acid Hydrazide." Monatshefte fiir Chemie 132, 753-763. 2001.*

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