Synthesis of 4-Azidocoumarins and Their Use in Copper-Catalyzed Azide-Alkyne Cycloaddition reactions

Anthony J. Netsuri
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Synthesis of 4-Azidocoumarins and Their Use in Copper-Catalyzed Azide-Alkyne Cycloaddition Reactions

A Thesis Presented to
The Faculty of the Chemistry Program
The City College of New York

In (Partial) Fulfillment
of the Requirements for the Degree
Master of Science

by
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January 2013
APPROVED FOR THE CHEMISTRY PROGRAM

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ABSTRACT

SYNTHESIS OF 4-AZIDOCOUMARINS AND THEIR USE IN COPPER-CATALYZED AZIDE-ALKYNE CYCLOADDITION REACTIONS

By

Anthony J. Nesturi

Triazole-containing compounds have shown great biological activity ranging from antiviral, antibacterial, to anticancer, to name a few. Coumarin derivatives have also shown interesting biological activities. The combination of these bioactive compounds appears to have great promise for new and future medicines. In this work, various 4-azido-coumarins were synthesized via the transformation of the 4-hydroxy derivatives to 4-benzotriazolyloxy coumarins by reaction with the peptide coupling agent (benzotriazol-1-yloxy)tris-(dimethylamino)phosphonium hexafluorophosphate (BOP), and 1,8-diazabicycloundec-7-ene (DBU) as the base, in tetrahydrofuran (THF) solvent. The 4-benzotriazolyloxy coumarins were converted to the 4-azidocoumarins by reaction with sodium azide (NaN₃), and the overall process was simplified to a one-pot, two-step process. The methodology reduced the need for additional manipulations and purifications. Using the 4-azidocoumarins thus prepared, the 1,4-disubstituted-1,2,3 triazoles were synthesized via a copper-catalyzed alkyne-azide cycloaddition (CuAAC) reaction. CuAAC reactions of the azides were optimized by screening of various copper catalysts.
and copper tetrakis acetonitrile hexafluorophosphate ([Cu(CH$_3$CN)$_4$]PF$_6$) gave the best results in the experiments performed. Once the reaction conditions were optimized, various 4-(1,2,3-triazolyl)coumarins were synthesized in good to excellent yields.
INTRODUCTION

The triazole moiety, as the name indicates, contains three nitrogen atoms in a five-membered ring. As illustrated in Figure 1, there are two isomeric forms of triazoles, namely the 1,2,3 and the 1,2,4 isomers.

![Figure 1. The two isomers of triazoles.](image)

Compounds containing these five-membered rings exhibit a wide range of bioactivity. For example, the triazole containing compound in Figure 2 has shown a wide spectrum of antifungal activity. This compound is a potent antifungal with the ability to competitively inhibit the enzyme activity of lanosterol 14α-demethylase (CYP51) in the demethylation step involved in the biosynthesis of ergosterol. Ergosterol is an important constituent of the cell membrane of fungi. This antifungal property makes the mentioned compound a good agent to fight against the Candida species. The antifungal effect was observed with a minimal concentration range of 0.0625 to 1.0000 μg/mL, with 80% inhibition of growth of the various fungal pathogens tested.
Figure 2. An antifungal compound containing the 1,2,4-triazole motif.

Pharmaceutical companies like AstraZeneca have used triazole compounds in some of their antibacterial drugs. The utilizations of oxazolidinone moieties in their antibacterial compounds produced great results against gram-positive bacteria such as: methicillin-susceptible Staphylococcus aureus, penicillin-susceptible Streptococcus pneumoniae, and Haemophilus influenzae. The disadvantage of the oxazolidinone-derived product was its inhibitory effect on Monamine Oxidase-A (MAO-A) activity, which leads to severe hypertensive problems when the medication was used.³ To resolve this complication with the oxazolidinone compound Reck et al. attached a 1,2,3-triazole group, and the resulting compound (Figure 3) reduced the hypertensive problem produced by the use of the medication. The use of the 1,2,3-triazole did not appear to affect the antibacterial potency.³
Figure 3. An oxazolidinone antibacterial compound with a 1,2,3-triazole, which reduced MAO-A inhibition.

Triazole compounds have shown beneficial results in the fight against diseases that have been difficult to eradicate. Kumar et al. provided strong evidence that quinoline triazolyl sugar compounds attacks *Mycobacterium tuberculosis* H37Rv. They suggested the use of this compound as an effective anti-tuberculosis agent. From the various synthesized quinoline triazolyl sugar compounds. The compound in Figure 4 showed the most promise, where concentrations of 5 μg/mL and 25 μg/mL resulted in 76.41% and 78.37% reduction in relative light units in the luciferase mycobacteriophage test, respectively. In this test a plasmid is inserted into the bacterium by the phage virus, which contains luciferase and ampicillin resistant genes. The ampicillin resistance promotes the expression of the plasmid and with it luciferase is synthesized by the mycobacteria, resulting in the observed light, which is quantified to relative light units observed from the control and compound treated bacterium. The decreasing amount of light observed is correlated with the anti-tuberculosis activity of the quinoline triazolyl sugar compound.
Figure 4. Triazole containing compound as an anti-tuberculosis agent.

The bioactivity benefits of the triazole moiety contribute vastly in the creation of new bioactive compounds, and the added advantage that other bioactive groups attached to it could lead to a great improvement in activity or in the identification of novel activity. For example, the antiviral compound illustrated in Figure 5 contains a coumarin and a triazole moiety. This compound performs very well through dual action as a protease and reverse transcriptase inhibitor. The compound provides a 92% inhibition of reverse transcriptase with IC$_{50}$ at 5.56 μM and 61% inhibition of protease activity with IC$_{50}$ at 27.06 μM. The combined benefit gained by coumarin and triazole makes this compound a very promising antiviral agent.

Figure 5. Very potent reverse transcriptase and protease inhibitor.
The compound discussed above (Figure 5) is related to the subject of this thesis, in that it contains 1,2,3-triazole linked to a coumarin moiety. The red colored motif in Figure 5 is the coumarin and the blue is the 1,2,3-triazole ring. Coumarin is in fact a benzopyrone and the name comes from the French word for tonka bean “coumarou”. Other natural sources, particularly plants such as *Murraya siamensis*\(^6\) *Toddalia asiatica*\(^7\) and *Calophyllum lanigerum*,\(^8\) and Australian ascidians of the *Didemnum* species\(^9\) are a few sources of various coumarin compounds. Many coumarins have fascinating and significant physiological as well as biological properties. For example, the coumarins isolated by Ito *et al.* show anti-tumor activities in the Epstein-Barr virus-activation assay.\(^6\) This test measures the capability of the coumarin compounds to inhibit the ability of the Epstein-Barr virus to cause cancer in otherwise healthy cells.

Chen *et al.* have studied the use of coumarin derivatives as anticancer agents.\(^10\) In this study an *in vitro* analysis of the compounds showed that 7-(6-chloropyridin-2-ylthio)-4-methyl-2H-chromen-2-one was a highly potent compound with \(GI_{50}\) values ranging from 0.92 to 2.11 mM for differing cell lines (Figure 6.). The compound promoted apoptosis and showed growth inhibitory activity in cancer cells and only weak cytotoxicity to normal cells.\(^10\)
In a skin cancer study performed by Banerjee et al., 4-methyl-7-hydroxycoumarin showed great ability in preventing the progression of skin cancer when rats were exposed to 7,12-dimethylbenz[a]anthracene (DMBA). A summary of the results from the publication of Banerjee et al., illustrated in a graph, is shown in Figure 7. The data clearly show that the coumarin compound acted as an inhibitor of DMBA-mediated tumorigenesis. The authors determined that the coumarin compound prevented the cancer cells from entering the S phase of the DNA replication, ultimately leading to the reduction the tumor formation in mice.

Figure 7. Structures of DMBA and 4-methyl-7-hydroxycoumarin. Shown to the right is the effect of 4-methyl-7-hydroxycoumarin on the progression of skin cancer in mice (graph taken from reference 11).
Further studies on breast cancer have shown that high levels of estrogen synthesis are associated with the formation of tumors. The aromatase and sulfatase pathways involved in the synthesis of the estrogen, are linked to the formation of tumors. Studies in the chemotherapy of breast cancer have shown that coumarin analogs and their 7-hydroxycoumarin lower the activity of the sulfatase and aromatase enzymes. This action inhibits the synthesis of estrogen, which has been linked to one third of breast cancer cases. Figure 8 shows the structures of the two coumarin derivatives which are known to possess this novel anticancer activity.

![Chemical Structures](image)

**Figure 8.** Aromatase and sulfatase activity inhibiting coumarin compounds.

Compounds that combine the coumarin and triazole moieties have been reported in many publications. Other examples 3-(1,2,3-triazolyl)coumarins and 4-(1,2,3-triazolyl)coumarins have been previously synthesized and studied for their fluorescent properties and for their use in protein or DNA labeling. The labeling of protein and DNA by this metal-catalyzed cycloaddition greatly facilitated this process. These reactions can be run in some part of aqueous solvent, making it possible to incorporate fluorophores in great yield into water
soluble protein without compromising their structural integrity. The CuAAC method has provided various oligonucleotides that are synthesized under mild conditions and easily incorporated in DNA.

**Figure 9.** Shown to the left is a protein labeled with a fluorescent coumarin probe (figure taken from reference 14). Shown to the right is thymine labeled with a 3-(1,2,3-triazoyl)-coumarin fluorescent probe for DNA labeling (redrawn from reference 15).

The 1,2,4-triazolyl coumarin shown in Figure 10 has shown good antibacterial and antifungal properties. The antibacterial properties were comparable to streptomycin against *Bacillus subtilis*, *Escherichia coli*, and *Pseudomonas aeruginos*. The compound’s antifungal activity is similar to that of fluconazole against fungal cultures of *Aspergillus niger* and *Aspergillus flavus*.

**Figure 10.** Potent antibacterial triazole-coumarin conjugate.
A 1,2,3-triazole containing two coumarin moieties (Figure 11) presented significant antimycobacterial activity as against *Mycobacterium tuberculosis*, similar to streptomycin. This compound is also active against gram-positive bacteria, however, it was not as effective when compared to streptomycin and fluconazole.

![Chemical structure](image)

**Figure 11.** Antibacterial, antiviral, and antitubercular 1,4-disubstituted bis-coumarin triazole derivative.

As can be seen from the foregoing, not only are triazoles and coumarins independently important biologically, but a combination of the two moieties also leads to powerful biologically valuable compounds. Thus, we were highly interested in combining the biological properties of the triazolyl moiety with that of coumarin to generate a novel class of “triazolyl coumarins”. We chose to prepare C-4 triazolyl coumarins with the general structure shown in Figure 12.
Figure 12. C-4 triazolyl coumarins that were selected for synthesis.

Two possible retrosynthetic analyses of the structural prototype depicted in Figure 12 are shown in Scheme 1. In one approach (shown to the right in Scheme 1), a leaving group at the C-4 position of coumarin can potentially be displaced with a triazole. This however, could result in the formation of N1 and N2 regioisomeric products. In addition, the generality and ease of such a displacement with a triazole as nucleophile is unknown.

Scheme 1. Two possible approaches to the synthesis of C-4 triazolyl coumarins

As shown to the left of Scheme 1, an alternate approach is a dipolar cycloaddition of a C-4 azidocoumarin derivative with an alkyne. This appeared to be a methodology that could be more readily accomplished, with complete control over regiochemical questions. Specifically, the coumarin would be attached to
the N1 of the triazole and the triazolyl substituent would be positioned at the C-4 of the coumarin. Further, a variety of approaches are available for the synthesis of terminal alkynes, e.g. Sonogashira reactions of aryl halides, Corey-Fuchs olefination of aldehydes followed by reaction with a strong base, and the Bestmann-Ohira alkynylation of aldehydes, which provide ready access to almost any alkyne.

This leaves only the development of a general method for the synthesis of 4-azidocoumarins. A potential procedure for accomplishing this, on the basis of chemistry developed in our laboratories, is described later in this chapter.

*Synthesis of 1,2,3-Triazoles*

The classical method for the synthesis of the triazole ring is via a Huisgen ligation, which is a 2,3-dipolar cycloaddition between an alkyne and an azide. This reaction involves the 4π electrons of the zwitterionic azide, the *dipole*, and the 2π electrons of a neutral dipolarophile, the alkyne, resulting in a five-membered ring. The reaction occurs by the interaction of the HOMO of the dipole with the LUMO of the dipolarophile. Figure 10 shows three different types of interactions of the dipole-dipolarophile electron densities. The reaction types categorized by Huisgen are as follows.

(a) Type 1: is a normal electron-demand type of process wherein the high-lying HOMO (ψ2) of the dipole interacts with the LUMO (ψL) of dipolarophile.
(b) Type 2: due to the similar energy gap, either the HOMO of the dipole interacts with the LUMO of the dipolarophile, or the LUMO of the dipole can interact with the HOMO of the dipolarophile.

(c) Type 3: is the inverse-electron demand type of reaction, where the low-lying LUMO of the dipole ($\psi_3$) interacts with the HOMO ($\psi_A$) of the dipolarophile.

**Figure 13.** HOMO and LUMO interactions between the dipole and dipolarophile (redrawn from ref 18).

One major problem with the otherwise highly atom-economical Huisgen ligation is the propensity for the formation of regioisomeric triazoles via the cycloaddition. Thus, under thermal conditions, both 1,4- and 1,5-disubstituted-1,2,3-triazoles are formed in the Huisgen ligation (Scheme 2).

**Scheme 2.** Formation of regioisomeric products in the Huisgen ligation

\[
R - \equiv - + N\equiv N^+ N^+ \xrightarrow{\Delta} N\equiv N\equiv N - R^1 + N\equiv N\equiv N - R^1
\]
In 2002, Meldal and colleagues, as well as Fokin, Sharpless, and colleagues, independently demonstrated a metal-catalyzed version of the Huisgen ligation, that was highly regioselective. In the copper-catalyzed reaction, the use of a copper(I) catalyst largely eliminated the formation of the 1,5-disubstituted 1,2,3-triazoles in the reaction of azides with alkynes. The general term “click chemistry” was coined by Sharpless and coworkers due to the fact that high-energy groups (azide and alkyne) combine to form a stable, lower energy compound. The copper-catalyzed reaction of azides with alkynes is now called CuAAC (Copper (I) catalyzed Alkyne-Azide Cycloaddition). Generally, the CuAAC reactions utilize an in-situ reduction of copper(II) to copper(I), or a copper(I) salt, and organic solvent-soluble copper salts such as $[\text{Cu(CH}_3\text{CN)}_4]\text{PF}_6$ can also be used.

The proposed mechanism for the CuAAC reactions postulated by Fokin et al. is shown in Figure 14. The alkyne in the reaction reversibly coordinates through a weak $\pi$ bond with the copper catalyst. In the following step another molecule of copper catalyst forms a $\sigma$ bond with the alkyne. The azide coordinates with the $\pi$ bound copper, and this is followed by a nucleophilic attack at the N3 of the azide resulting in a C–N bond. The dinuclear copper intermediate proceeds rapidly to the formation of the triazolide by the oxidation of one of copper atoms with the stabilizing effect of the other copper atom. Finally, detachment of the copper catalyst is facilitated by a proton donor, forming the
desired 1,4-disubstituted 1,2,3-triazole. The catalytic cycle proceeds until the depletion of the reactants.

**Figure 14.** Catalytic cycle proposed by Fokin et al. for Cu(I) azide alkyne cycloaddition (ref 21).

**Synthesis of 4-Azidocoumarins**

A variety of 4-hydroxycoumarins are commercially available or are easily synthesized. The C-4 hydroxyl group in these compounds can be converted to a leaving group and then subjected to displacement with azide ion to yield 4-azidocoumarins. A variety of leaving groups are known at the C-4 position of coumarins (Scheme 3), and commonly the chloride and sulfonate derivatives of coumarin can be prepared.
In fact, both types of reactive coumarin derivatives have been used for the synthesis of a variety of 4-azidocoumarin derivatives. However, these approaches require two discrete steps, namely synthesis of the reactive coumarin derivative and a subsequent displacement with azide ion. Furthermore the halogenation procedure involves the reflux of 4-hydroxycoumarin in POCl₃ and the formation of a modest product yield with a byproduct formation that requires azeotropic distillation to finally separated the products.

Research in Prof. Lakshman’s laboratories and from chemists at Weyth, has led to the identification of a novel method for the activation of amide linkages. Specifically, Lakshman and Bae have demonstrated that reaction of inosine and deoxyinosine with the peptide-coupling agent
(1H-benzotriazol-1-yl)oxy)tris(dimethylamino)phosphoniumhexafluorophosphate (BOP) leads to the formation of $O_6^\beta$-(benzotriazol-1-yl) nucleoside analogues (Scheme 5).²⁸

**Scheme 5.** Synthesis of $O_6^\beta$-(benzotriazol-1-yl) nucleoside derivatives

Subsequently, researchers at Weyth have demonstrated similar results.²⁹ Importantly, in their work they have generated 4-aminoquinazoline by activation of 4(3H)-quinazolinone using BOP. What should also be noted is that both inosine and 4-hydroxyquinazoline can be subjected to activation with BOP and nucleophilic displacement in a one-pot process as proposed by the authors.²⁹

**Scheme 6.** Synthesis of 4-aminoquinazoline

These benzotriazolyl intermediates are reactive towards nucleophilic displacement. Notably, of relevance to the chemistry herein, Singh and
Lakshman have prepared azido nucleosides from the $O^6$-(benzotriazol-1-yl) nucleosides in a two-step operation as shown in Scheme 7.\textsuperscript{30}

**Scheme 7.** $O^6$-(benzotriazol-1-yl) nucleosides and their use in the synthesis of 1,2,3-triazole nucleosides

We therefore reasoned that reaction of 4-hydroxycoumarin with BOP and a suitable base, in a suitable solvent, should yield the benzotriazolyl ether of the coumarin. Whether or not this intermediate is isolable can only be determined only upon performing the reaction. Secondly, on the basis of prior results, it should be eminently possible to synthesize 4-azidocoumarins in a two-step, one-pot procedure as shown in Scheme 8.
Scheme 8. A potential, simple one-pot method for the synthesis of 4-azidocoumarins

Deprotonation of the hydroxycoumarin by a base should yield a resonance-stabilized enolate. This species can react at the phosphorus atom of BOP, with loss of the benzotriazolyloxy anion, as has been demonstrated previously for nucleosides and pyrimidinones.\textsuperscript{28,29} The formed benzotriazolyloxy anion can then displace hexamethylphosphoramide (HMPA), via an addition-elimination pathway, leading to the potentially isolable C-4 benzotriazolyloxy coumarin derivative. This species can be converted to the azido coumarin, via another addition-elimination reaction. This step can either be performed on the isolated C-4 benzotriazolyloxy coumarin or via an \textit{in situ} displacement, subsequent to formation of C-4 benzotriazolyloxy coumarin.
Once the 4-azidocoumarins become available, they can be used in CuAAC reactions with various alkynes, leading to C-4 triazolylcoumarins. As stated in the introduction, triazoles and coumarins are both physiologically very important. Thus, development of synthetic methodology for combining these two structural motifs into a new class of 4-(4-substituted-1H-1,2,3-triazolyl)coumarins could result in compounds that are inherently biologically active. We not only anticipate an understanding of the chemistry involved and an evaluation of the broad utility of the method, but we also expect to evaluate any potential anti-cancer and antiviral properties of the newly developed compounds (via collaborations) as well as mechanistic questions and other properties.
RESULTS AND DISCUSSION

As described in the introduction the azidization of 4-hydroxycoumarin was performed by a two-step one-pot reaction. Briefly, 4-hydroxycoumarin was converted to its benzotriazolyl ether by reaction with (benzotriazol-1-yloxy)tris(dimethylamino)phosphoniumhexafluorophosphate (BOP) in the presence of a base, and the newly synthesized benzotriazolyl ether was then converted to the azide. The reaction and the results are summarized in Table 1. For the first step of the reaction tetrahydrofuran (THF) and 1,2-dimethoxyethane (DME) were evaluated as solvents with 1,8-diazabicycloundec-7-ene (DBU) as the base.
Table 1. The results of the various attempts in the azidization of 4-hydroxycoumarin

<table>
<thead>
<tr>
<th>Entry</th>
<th>Solvent 1</th>
<th>Solvent 2</th>
<th>Additive</th>
<th>Amount of Additive</th>
<th>Result, Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>THF</td>
<td>None</td>
<td>18-Cr-6</td>
<td>10 mol%</td>
<td>95% Isolated yield Step 1 reaction time 2 hours Step 2 reaction time 24 hours</td>
</tr>
<tr>
<td>2</td>
<td>THF</td>
<td>None</td>
<td>18-Cr-6</td>
<td>5 mol%</td>
<td>80% Isolated yield Step 1 reaction time 2 hours Step 2 reaction time 72 hours</td>
</tr>
<tr>
<td>3</td>
<td>THF</td>
<td>None</td>
<td>Diglyme</td>
<td>20 mol%</td>
<td>Formation of the benzotriazoyl ether occurred but azidation was not observed</td>
</tr>
<tr>
<td>4</td>
<td>DME</td>
<td>None</td>
<td>None</td>
<td>None</td>
<td>Rapid formation of the benzotriazoyl ether occurred but azidation was not observed</td>
</tr>
<tr>
<td>5</td>
<td>DME</td>
<td>Diglyme</td>
<td>None</td>
<td>None</td>
<td>80% Isolated yield Step 1 reaction time 1 hours Step 2 reaction time 24 hours</td>
</tr>
<tr>
<td>6</td>
<td>THF</td>
<td>EtOH</td>
<td>None</td>
<td>None</td>
<td>90% Isolated yield Step 1 reaction time 2 hours Step 2 reaction time 24 hours</td>
</tr>
</tbody>
</table>

The azidization step was performed after formation of the coumarin benzotriazoyl ether and this occurs by an addition-elimination reaction with azide anion as the nucleophile. In the first attempt 10 mol% of 18-crown-6 (18-Cr-6) was used as shown in entry 1 in Table 1 18-Cr-6 ether is a phase-transfer catalyst,
which facilitates the solubility of sodium azide in organic solvents by solvation of the sodium cation (Na⁺). However, crown ethers are relatively expensive compounds and in the process of purification they can contaminate the product in the case of an inefficient separation. However, this reaction proceeded well and gave 4-azidocoumarin in 95% isolated yield. In attempt to reduce the amount of crown ether, 5 mol% of 18-Cr-6 ether was used in a subsequent reaction (entry 2 of Table 1), but this resulted in significantly slower reaction (step 2 took 72 hours) and a product yield of 80%. In entry 3, the same solvent system as in entry 2 was used, but 20 mol% of bis(2-methoxyethyl) ether was added to the azidation step (step 2) after the formation of the benzotriazolyl ether. This effort did not prove fruitful in the azidization step. Other attempts were directed towards removal of 18-Cr-6 from the azidization step via the use of other dry solvents. DME and diglyme were used in an attempt to replace the crown ether due to their potential cation-chelating abilities. In entry 4, use of DME provided a very fast reaction leading to the benzotriazolyl ether formation (1 hour for step 1). However, the azidization step did not occur in this solvent in a 24 hour period. In entry 5 the benzotriazolyl ether formation was performed in DME solvent, the solvent was evaporated and diglyme was added as a solvent for the azide-forming step. The step 2 of the reaction was run for 24 hours and yielded 80% of the isolated product. Although diglyme was reasonable, its high boiling point and miscibility with organic solvent such as CH₂Cl₂ made it tedious to remove from
the reaction mixture. For example in our workup we employed EtOAc as the organic solvent for extraction, and 300 mL solution containing of brine and water mixture at 3:7 respectively for separation. Other difficulties could be present with the solubility of the substituted coumarins in EtOAc. To remedy these difficulties another option was evaluated. A web search led to the understanding that sodium azide is soluble in ethanol at 0.316 g/100 mL (at 16 °C).32 Thus, in entry 6 the formation of the benzotriazole ether was conducted in THF, then an equal volume of anhydrous ethanol was added followed by the addition of sodium azide into the reaction mixture. With these changes, the azide forming reaction was complete within 24 h and gave a 90% isolated yield of 4-azidocoumarin.
Table 2. Results of the azidation reactions of various coumarins.\(^a\)

<table>
<thead>
<tr>
<th>Entry</th>
<th>RX#</th>
<th>R(^1)</th>
<th>R(^2)</th>
<th>R(^3)</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>74B</td>
<td>H</td>
<td>H</td>
<td>H</td>
<td>95%</td>
</tr>
<tr>
<td>2</td>
<td>84B</td>
<td>Br</td>
<td>H</td>
<td>H</td>
<td>93%</td>
</tr>
<tr>
<td>3</td>
<td>85B</td>
<td>Cl</td>
<td>H</td>
<td>H</td>
<td>99%</td>
</tr>
<tr>
<td>4</td>
<td>86B</td>
<td>Cl</td>
<td>H</td>
<td>Cl</td>
<td>80%</td>
</tr>
<tr>
<td>5</td>
<td>88B</td>
<td>Me</td>
<td>H</td>
<td>H</td>
<td>71%</td>
</tr>
<tr>
<td>6</td>
<td>89B</td>
<td>H</td>
<td>Me</td>
<td>H</td>
<td>72%</td>
</tr>
<tr>
<td>7</td>
<td>90B</td>
<td>H</td>
<td>MeO</td>
<td>H</td>
<td>86%</td>
</tr>
</tbody>
</table>

\(^a\)The benzotriazole ether step in all reactions was conducted for 2 hours at room temperature followed by overnight azidization.

The method developed and described above for the azidization of the 4-hydroxycoumarin (Table 1) provided us with a protocol for the formation of other substituted 4-azidocoumarin derivatives. The results are summarized in Table 2, where the various 4-hydroxycoumarins in entries 1 to 7 have gave good to excellent yields of the corresponding azido derivatives, ranging from 71–99%. Of particular importance is the need for effective mixing during the course of these reactions. Specifically, great care and consideration should be given to the azidization step. Inadequate mixing gave poor yields, approximately 50%, when reactions were performed in screw cap vials. The impedance to good mixing was
caused by the formation of an insoluble gelatinous texture of the reaction mixture in the azidization step. Use of round bottom flasks provided superior results. The six azidocoumarins synthesized were obtained in good yields after column chromatography and were then used in the azide-alkyne click reactions.

Initially, the click reactions were performed by using the copper sulfate (CuSO$_4$)/sodium ascorbate as the catalyst system originally reported by Sharpless and coworkers (entries 1 through 8).$^{21}$ In this method CuSO$_4$, is reduced in situ by sodium ascorbate. However, the solvent system employed for entries 1–4 in Table 3 was CH$_2$Cl$_2$/H$_2$O, the reactions were performed at room temperature and at 50 °C with no substantial differences observed by TLC (incomplete reactions with only about 50% product formation). The solvent system in entries 5 and 6 consisted of an ethanol–water mixture, and no product formation was observed. Similarly, use of t-butanol and water as the solvent system with CuSO$_4$/sodium ascorbate at room temperature or at 50 °C did not result in product formation (entries 7 and 8). Next, the catalytic system was changed from CuSO$_4$/sodium ascorbate to copper(II) chloride (CuCl$_2$)/sodium ascorbate as shown in entries 9 and 10. Reactions were conducted both at room temperature and at 50 °C, and the solvent system was also changed to THF/H$_2$O. At the elevated temperature of 50 °C a small amount of product formation (ca 10 %) was observed by TLC. Due to the observed no to low conversions with the inorganic copper-catalyzed reactions, the use of organic copper(I) salts were evaluated. In entry 11,
effectiveness of copper(I) thiophene-2-carboxylate in CH$_2$Cl$_2$/CH$_3$OH was evaluated 50 °C. This reaction gave very good yield (entry 11). Nonetheless, we encountered the formation of a small amount of the isomeric 1,5-triazole as shown by NMR in Figure 15 (small peak at 6.35 ppm).

![NMR spectrum](image)

**Figure 15.** NMR spectrum of copper(I) thiophene-2-carboxylate catalyzed reaction with products of 1,4 and traces of 1,5-(1,2,3-triazoyl)-coumarin (circled by the oval).

A NOESY experiment of the 1,5-triazoyl isomer was performed and the data is illustrated in Figure 16. The data shows that there is no spatial correlation observed between the triazoyl and the vinyl protons. This supports the 1,5-disubstitution pattern. In this isomer the vinyl proton not spatially proximal to any other proton, especially the vinyl proton.
Figure 16. NOESY spectrum in CDCl$_3$ of 4-(5-phenyl-1H-1,2,3-triazol-1-yl)-coumarin showing no correlation with any protons and especially the vinyl proton.

Similar to the 1,5-disubstituted triazolyl coumarin, a NOESY experiment was conducted on the 1,4-triazoyl isomer and this is shown in Figure 17. In this spectrum a clear interaction of the vinyl proton at 6.35 ppm with the triazoyl proton at 7.99 ppm is seen, and this is indicated by the lines.
The use of copper(I)-thiophene-2-carboxylate presented the possibility to use another organic Cu catalyst, namely tetrakis (acetonitrile) copper(I) hexafluorophosphate \([\text{Cu(CH}_3\text{CN})_4]\text{PF}_6\) under the same reaction conditions as with Cu(I) thiophene-2-carboxylate (entry 12). A reaction with \([\text{Cu(CH}_3\text{CN})_4]\text{PF}_6\) resulted in a product yield of 95%, and the NMR showed...
formation of only the 1,4-triazole regioisomer. In entry 13, the exact reaction conditions used by Qian Wang et al.\textsuperscript{13} were attempted for our reaction. The reactions reported by Wang et al. involve the CuAAC reactions of 3-azidocoumarin derivatives with 5% CuSO\textsubscript{4}/10% sodium ascorbate in ethanol and water.\textsuperscript{13} Although they report a single example of the use of 5% CuSO\textsubscript{4}/10% sodium ascorbate in ethanol and water, use of these conditions for the CuAAC reaction of 4-azidocoumarin and 1-ethynylbenzene did not effectuate any product formation in our hands (as evaluated by NMR, Figure 18).

![Figure 18](image)

**Figure 18.** Crude mixture of a reaction between 4-azidocoumarin and 1-ethynylbenzene with 5% CuSO\textsubscript{4}/10% sodium ascorbate as the catalyst in ethanol and water after workup showing no formation of triazole.

In entry 14, a reaction with copper(I) iodide as catalyst in acetonitrile and water was performed at 50 °C, and only a low extent of product formation was
observed (ca 10–25%), as observed by TLC. Due to the low conversion observed in entry 14, the solvent system was evaporated and replaced with CH₂Cl₂/CH₃OH (entry 15). Still, this was not effective either. Use of copper(I) chloride in CH₂Cl₂/CH₃OH again showed low conversion to product (ca 10–25%), and at 50 °C (entry 16) no significant change was observed. The low yielding reaction prompted another change of the solvent system (entry 17), and in CH₃CN/CH₃OH at 50 °C, as a light improvement was observed but conversion still remained (ca 20–30%), as observed by TLC.

In summary, [Cu(CH₃CN)₄]PF₆ proved to be the best among the catalytic systems tested in Table 3. Product formation occurred in high yield and no trace of the isomeric 1,5-triazole was observed by NMR. Reactions were performed in CH₂Cl₂/CH₃OH, which offered good solubility for all the azidocoumarin as well as the catalyst. Addition of CH₃OH was used to ensure the presence of a proton source for the recycling of the catalyst.
Table 3. Various conditions tested for the CuAAC reaction of 4-azidocoumarin with phenylacetylene

<table>
<thead>
<tr>
<th>Entry</th>
<th>Catalyst</th>
<th>Solvent System</th>
<th>Condition</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>5 mol % CuSO₄ 10 mol % sodium ascorbate</td>
<td>DCM 0.33 mL H₂O 0.3 mL</td>
<td>rt, 24 h</td>
<td>~40-50%</td>
</tr>
<tr>
<td>2</td>
<td>5 mol % CuSO₄ 10 mol % sodium ascorbate</td>
<td>DCM 0.33 mL H₂O 0.3 mL</td>
<td>50 °C, 24 h</td>
<td>~40-50%</td>
</tr>
<tr>
<td>3ᵇ</td>
<td>5 mol % CuSO₄ 10 mol % sodium ascorbate</td>
<td>DCM 0.66 mL H₂O 0.6 mL</td>
<td>rt, 24 h</td>
<td>~40-50%</td>
</tr>
<tr>
<td>4ᵇ</td>
<td>12 mol % CuSO₄ 24 mol % sodium ascorbate</td>
<td>Ethanol 0.66 mL H₂O 0.6 mL</td>
<td>rt, 24 h</td>
<td>No Product</td>
</tr>
<tr>
<td>5ᵇ</td>
<td>12 mol % CuSO₄ 24 mol % sodium ascorbate</td>
<td>Ethanol 0.66 mL H₂O 0.6 mL</td>
<td>50 °C, 24 h</td>
<td>No Product</td>
</tr>
<tr>
<td>6ᵇ</td>
<td>12 mol % CuSO₄ 24 mol % sodium ascorbate</td>
<td>t-Butanol 0.66 mL H₂O 0.6 mL</td>
<td>rt, 24 h</td>
<td>No Product</td>
</tr>
<tr>
<td>7ᵇ</td>
<td>12 mol % CuSO₄ 24 mol % sodium ascorbate</td>
<td>t-Butanol 0.66 mL H₂O 0.6 mL</td>
<td>50 °C, 24 h</td>
<td>No Product</td>
</tr>
<tr>
<td>8ᵇ</td>
<td>33.5 mol % CuCl₂ 6.7 mol % Sodium Ascorbate</td>
<td>THF 0.53 mL H₂O 0.12 mL</td>
<td>rt, 24 h</td>
<td>No Product</td>
</tr>
<tr>
<td>9ᵇ</td>
<td>33.5 mol % CuCl₂ 6.7 mol % sodium ascorbate</td>
<td>THF 0.53 mL H₂O 0.12 mL</td>
<td>50 °C, 24 h</td>
<td>~10-25%</td>
</tr>
<tr>
<td>10ᵇ</td>
<td>10 mol % Copper(I)-thiophene-2-carboxylate</td>
<td>CH₂Cl₂ 0.55 mL CH₃OH 0.12 mL</td>
<td>50 °C, 24 h</td>
<td>100% conversion²</td>
</tr>
<tr>
<td>11ᵇ</td>
<td>10 mol % [(CH₃CN)₄Cu]PF₆</td>
<td>CH₂Cl₂ 0.55 mL CH₃OH 0.12 mL</td>
<td>50 °C, 24 h</td>
<td>95% Yield³</td>
</tr>
<tr>
<td>12ᵇ</td>
<td>20 mol % CuCl 20 mol % sodium ascorbate</td>
<td>Ethanol 2.5 mL H₂O 2.5 mL</td>
<td>rt in dark, 24 h</td>
<td>No product⁴</td>
</tr>
<tr>
<td>14ᵇ</td>
<td>20 mol % CuI</td>
<td>CH₂CN 0.47 mL H₂O 0.15 mL</td>
<td>50 °C, 24 h</td>
<td>~10-25%</td>
</tr>
<tr>
<td>15ᵇ</td>
<td>20 mol % CuI</td>
<td>DCM 0.55 mL CH₃OH 0.12 mL</td>
<td>50 °C, 24 h</td>
<td>~10-25%</td>
</tr>
<tr>
<td>16ᵇ</td>
<td>20 mol % CuCl</td>
<td>DCM 0.55 mL CH₃OH 0.12 mL</td>
<td>50 °C, 24 h</td>
<td>~10-25%</td>
</tr>
<tr>
<td>17ᵇ</td>
<td>20 mol % CuCl</td>
<td>CH₂CN 0.55 mL CH₃OH 0.12 mL</td>
<td>50 °C, 24 h</td>
<td>~20-30%</td>
</tr>
</tbody>
</table>

ᵃReaction were run at 0.1 mmol scale. ᵃᵇReaction were run at 0.2 mmol scale. ᵃᶜNot isolated yield TLC only and crude mixture ¹H NMR confirmed. ᵃᵈIsolated yield. ᵃᵉAnalysed by ¹H NMR, and reaction performed based on literature procedure from reference 7.
The reaction conditions involving the use of [(CH3CN)4Cu]PF6 performed very well on all five alkynes that were tested in reactions with 4-azidocoumarin (Table 4), with yields in the range of 72–95%. The only notable exceptions are shown in entries 3 and 4, involving the reactions of 1-ethynyl-4-methoxybenzene and 3-ethynylpyridine. The reaction of 1-ethynyl-4-methoxybenzene was incomplete after 24 hours at 50 °C. To remedy this problem the reaction was re-run at 80 °C for 24 hours, which resulted in a product yield of 80%. The reaction of 3-ethynyl-pyridine went to completion at room temperature with a product yield of 95%.

**Table 4.** Alkynes used and conditions, and product yields in the CuAAC reactions of 4-azidocoumarin

<table>
<thead>
<tr>
<th>Entry</th>
<th>Alkyne</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td><img src="image" alt="Alkyne 1" /></td>
<td>95%</td>
</tr>
<tr>
<td>2</td>
<td><img src="image" alt="Alkyne 2" /></td>
<td>87%</td>
</tr>
<tr>
<td>3</td>
<td><img src="image" alt="Alkyne 3" /></td>
<td>80%&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>4</td>
<td><img src="image" alt="Alkyne 4" /></td>
<td>95%&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>Procedure: 0.2 mmol reaction in 0.55 mL of CH2Cl2 and 0.12 mL of CH3OH at 50 °C for 24 h with 10 mol % of [Cu(CH3CN)4]PF6. <sup>b</sup>Reaction was performed at 80 °C. <sup>c</sup>Reaction was performed at room temperature.
The reaction conditions used for the conversions of 4-azidocoumarin to the 4-(1,2,3)-triazolyl coumarins shown in Table 4 were then tested with the six substituted 4-azidocoumarins, and the results are listed in Table 5. The overall reactivity of the substituted coumarin compounds remained unchanged. As reported for the reaction of 4-azidocoumarin with 1-ethynyl-4-methoxybenzene, a similar reactivity problem was encountered in the reaction of 7-methoxycoumarin with 1-ethynyl-2-methyl-4-methoxybenzene (entry 4). By TLC analysis, this reaction showed only a low extent of product formation. When the reaction temperature was raised to 80 °C for another 24 hours, TLC showed formation of new uncharacterized materials indicating possible decomposition.
Table 5. Various alkynes and azidocoumarin combinations in diverse CuAAC reactions

<table>
<thead>
<tr>
<th>Entry</th>
<th>R</th>
<th>R¹</th>
<th>R²</th>
<th>R³</th>
<th>Yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td></td>
<td>Br</td>
<td>H</td>
<td>H</td>
<td>67%</td>
</tr>
<tr>
<td>2</td>
<td></td>
<td>Me</td>
<td>H</td>
<td>H</td>
<td>86%</td>
</tr>
<tr>
<td>3</td>
<td></td>
<td>H</td>
<td>Me</td>
<td>H</td>
<td>89%</td>
</tr>
<tr>
<td>4</td>
<td></td>
<td>H</td>
<td>MeO</td>
<td>H</td>
<td>10-20%</td>
</tr>
</tbody>
</table>

Procedure: 0.2 mmol reaction in 0.55 mL of CH₂Cl₂ and 0.12 mL of CH₂Cl₂ at 50 °C over 24 h using 10 mol % of [Cu(CH₃CN)₄]PF₆. Product formation is tracked by TLC only.

Further optimization of at least some of these reactions is necessary to improve yields. In this aspect the use of tris(3-hydroxypropyltriazolylmethyl)amine and the use of 2,6-lutidine has been described by Sharpless et al., however, further experiments are needed to see if such additives will provide an improvement in the yields.
EXPERIMENTAL SECTION

General Experimental Considerations

Thin layer chromatography was performed on aluminum foil-backed TLC plates of 200 μm thickness and were visualized under short wavelength (265 nm) and/or long wavelength (354 nm) ultraviolet light. Column chromatographic purifications were performed on 200–300 mesh silica gel. Tetrahydrofuran (THF) solvent was distilled over lithium aluminum hydride (LiAlH₄) then stored over sodium metal (Na). It was then refluxed for at least one hour over sodium and distilled prior to use. CH₂Cl₂ was distilled over CaCl₂, and redistilled prior to use. Ethyl acetate (EtOAc) was distilled prior to use. All other reagents were obtained from commercial sources and were used without further purification. Proton NMR spectra were recorded at 500 MHz in CDCl₃ and are referenced to the residual solvent resonance. Carbon NMR spectra were recorded at 125 MHz in CDCl₃ and are referenced to the residual solvent resonance.

General procedure for the conversion of 4-hydroxycoumarin derivatives to the intermediate O₄-(1,2,3-benzotriazolyl) derivatives, and then to the 4-azidocoumarins.

The conversions of various 4-hydroxycoumarin derivatives (R₁=H, R₂=H, R₃=H) to the intermediate O₄-(1,2,3-benzotriazolyl) derivatives and then the azides were conducted in a two-step, one-pot process. As a representative
procedure, into a 200 mL oven-dried round bottom flask equipped with a stirring bar, was placed 4-hydroxycoumarin (1.00 g, 5.09 mmol) in THF (50 mL) to give a 0.1 M solution. 1H-Benzo[triazol-1-yloxy]tris(dimethylamino)phosphoniumhexafluorophosphate (BOP, 2.65 g, 6.00 mmol, 1.2 equiv) was added and the mixture was stirred at 25 °C for 10 minutes. At this stage a white solid persists at the bottom of the flask. Next DBU (0.91 mL, 6.04 mmol, 1.2 equiv) was added at which point the mixture became homogenous, and turned to a slight yellow solution. The mixture was stirred at room temperature until completion, generally 2 h, although the course of the reaction was intermittently followed by TLC (SiO₂/30% EtOAc in hexanes) until it ended. The reaction mixture was added NaN₃ (0.66 g, 10.0 mmol, 2 equiv) followed by the addition of EtOH (200 proof, 50 mL). The mixture was allowed to stir for 24 h at room temperature, and the course of product formation was intermittently monitored by TLC (SiO₂/30% EtOAc in hexanes). Upon completion of the reaction, the mixture was partitioned between CH₂Cl₂ and brine. The organic layer was separated, dried over anhydrous Na₂SO₄, and transferred into a round bottom flask. To this 200–300 mesh silica gel was added and the solvent was evaporated under reduced pressure. The silica gel with the adsorbed reaction mixture was loaded onto a 200–300 mesh silica gel column and eluted with a mixture of EtOAc in hexanes. Fractions containing pure material were combined, evaporated under reduced pressure, and vacuum
dried. All azidocoumarins were synthesized in a similar manner and any deviations from this procedure are noted under the individual compound headings.
4-Azidocoumarin (1). Chromatography using 10–20% EtOAc in hexanes gave 889 mg (95% yield from a 5 mmol reaction) of compound 1 as a light yellow solid. R_f (SiO_2/30% EtOAc in hexanes) = 0.37. Melting point = 155–157 °C dec (lit. 154–155 °C). ^1H NMR (500 MHz, CDCl_3): δ 7.72 (d, 1H, Ar-H, J = 7.8 Hz), 7.60 (t, 1H, Ar-H, J = 8.3 Hz), 7.35 (d, 1H, Ar-H, J = 8.3 Hz), 7.30 (t, 1H, Ar-H, J = 7.6 Hz), 6.14 (s, 1H, =C-H). ^13C NMR (125 MHz, CDCl_3): δ 160.5, 153.6, 153.4, 133.2, 124.3, 123.4, 116.8, 114.8, 100.2. HRMS (ESI/TOF): calcd for C_9H_6N_3O_2 [M + H]^+ 188.0455, found 188.0451.

4-Azido-6-chlorocoumarin (2). Chromatography using gradient elution with 10–20% EtOAc in hexanes gave 219 mg (99% yield from a 1 mmol reaction) of compound 2 as a white solid. R_f(SiO_2/30% EtOAc in hexanes) = 0.44. Melting point = 169–171 °C dec. ^1H NMR (500 MHz, CDCl_3): δ 7.69 (d, 1H, Ar-H, J = 2.4 Hz), 7.35 (d, 1H, Ar-H, J = 8.8 Hz), 7.29 (d, 1H, Ar-H, J = 8.8 Hz), 6.16 (s, 1H, =C-H). ^13C NMR (125 MHz, CDCl_3): δ 158.6, 152.1, 147.9, 133.12, 129.6, 122.9, 121.6, 116.78, 101.3. HRMS (ESI/TOF): calcd for C_9H_5ClN_3O_2 [M + H]^+ 222.0065, found 222.0062.
4-Azido-6-bromocoumarin (3). Chromatography using gradient elution with 15–20% EtOAc in hexanes gave 247 mg (93% yield from a 1 mmol reaction) of compound 3 as a white solid. $R_f$ (SiO$_2$/30% EtOAc in hexanes) = 0.48. Melting point = 171–172 °C dec. $^1$H (500 MHz, CDCl$_3$): $\delta$ 7.85 (d, 1H, Ar-H, $J = 2.4$ Hz), 7.68 (dd, 1H, Ar-H, $J = 2.2$, 9.0 Hz), 7.24 (d, 1H, Ar-H, $J = 8.8$ Hz), 6.15 (s, 1H, =C-H). $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta$ 160.2, 152.7, 152.6, 136.3, 126.3, 119.0, 117.4, 116.6, 101.2. HRMS (ESI/TOF): calcd for C$_9$H$_5$BrN$_3$O$_2$ [M + H]$^+$ 265.9560, found 265.9563.

4-Azido-6-methylcoumarin (4). Chromatography using gradient elution with 10–20% EtOAc in hexanes gave 143 mg (71% yield from a 1 mmol reaction) of compound 4 as a white crystalline solid. $R_f$ (SiO$_2$/30% EtOAc in hexanes) = 0.57. Melting point = 171–172 °C dec. (lit.$^{33}$ 140–142 °C). $^1$H (500 MHz, CDCl$_3$): $\delta$ 7.44 (s, 1H, Ar-H), 7.35 (d, 1H, Ar-H, $J = 8.3$ Hz), 7.18 (d, 1H, Ar-H, $J = 8.3$ Hz), 6.05 (s, 1H, =C-H), 2.37 (s, 3H, CH$_3$). $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta$ 160.7, 153.2, 151.6, 134.1, 134.1, 122.9, 116.6, 114.4, 100.0, 20.8. HRMS (ESI/TOF): calcd for C$_{10}$H$_8$N$_3$O$_2$ [M + H]$^+$ 202.0611, found 202.0633.
**4-Azido-7-methylcoumarin (5).** Chromatography using gradient elution with 10–20% EtOAc in hexanes gave 145 mg (72% yield from a 1 mmol reaction) of compound 5 as a white solid. 

\[ R_f(\text{SiO}_2/30\% \text{ EtOAc in hexanes}) = 0.46. \]

Melting point = 160–162 °C dec. (lit.\textsuperscript{33} 155–156 °C). \( ^1\text{H} (500 \text{ MHz, CDCl}_3): \delta 7.58 (d, 1\text{H, Ar-H, } J = 8.3 \text{ Hz}), 7.14 (s, 1\text{H, Ar-H, } J = 7.1 \text{ Hz}), 7.10 (d, 1\text{H, Ar-H, } J = 8.3 \text{ Hz}), 6.06 (s, 1\text{H, } =\text{C-H}), 2.46 (s, 3\text{H, CH}_3). \]

\( ^{13}\text{C NMR (125 MHz, CDCl}_3): \delta 160.8, 153.6, 153.4, 144.6, 125.5, 122.9, 116.9, 112.3, 99.0, 21.7. \)

HRMS (ESI/TOF): calcd for C\textsubscript{10}H\textsubscript{8}N\textsubscript{3}O\textsubscript{2} [M + H\textsuperscript{+}] 202.0611, found 202.0637.

**4-Azido-7-methoxycoumarin (6).** Chromatography using gradient elution with 10–20% EtOAc in hexanes gave 187 mg (86% yield from a 1 mmol reaction) of compound 6 as a light red solid.

\[ R_f(\text{SiO}_2/30\% \text{ EtOAc in hexanes}) = 0.40. \]

Melting point = 173–175 °C dec. (lit.\textsuperscript{33} 178–180 °C). \( ^1\text{H} (500 \text{ MHz, CDCl}_3): \delta 7.60 (d, 1\text{H, Ar-H, } J = 8.8 \text{ Hz}), 6.85 (dd, 1\text{H, Ar-H, } J = 2.0, 9.0 \text{ Hz}), 6.82 (d, 1\text{H, Ar-H, } J = 2.4 \text{ Hz}), 5.98 (s, 1\text{H, } =\text{C-H}), 3.88 (s, 3\text{H, OCH}_3). \]

\( ^{13}\text{C NMR (125 MHz, CDCl}_3): \delta 163.9, 161.1, 155.5, 153.6, 124.4, 112.5, 108.2, 100.5, 97.0, 55.7. \)

HRMS (ESI/TOF): calcd for C\textsubscript{10}H\textsubscript{8}N\textsubscript{3}O\textsubscript{3} [M + H\textsuperscript{+}] 218.0560, found 218.0564.
4-Azido-6,8-dichlorocoumarin (7). Chromatography using gradient elution with 15–20% EtOAc in hexanes gave 205 mg (80% yield from a 1 mmol reaction) of compound 7 as a white solid. R f (SiO2/30% EtOAc in hexanes) = 0.53. Melting point = 171–172 °C dec. 1H(500 MHz, CDCl3): δ 7.64 (d, 1H, Ar-H, J = 2.5 Hz), 7.62 (d, 1H, Ar-H, J = 2.5), 6.19 (s, 1H, =C-H). 13C NMR (125 MHz, CDCl3): 158.9, 152.5, 148.3, 133.5, 129.9, 123.3, 121.9, 117.1, 101.6. HRMS (ESI/TOF): calcd for C9H4Cl2N3O2 [M + H]⁺ 255.9675, found 255.9675.

General procedure for the azide-alkyne ligation reactions leading to the formation of 4-(4-substituted-1H-1,2,3-triazol-1-yl)-coumarin derivatives.

The CuAAC reactions of various 4-azidocoumarin derivatives (R1 = H, R2 = H, R3 = H) as follows. As a representative procedure, into a long (6 inch) screw-cap vial equipped with a stirring bar, were placed 4-azidocoumarin (37.4 mg, 0.2 mmol, 1 equiv) and CH2Cl2 (0.55 mL) and CH3OH (0.12 mL) and [Cu(CH3CN)4]PF6 (7.5 mg, 0.02 mmol, 0.10 equiv). The mixture was stirred at room temperature to dissolve the reactants, then phenylacetylene (26 µL, 0.24 mmol, 1.2 equiv) was pipetted into the reaction mixture. The vial was capped and placed in a sand bath maintained at 50 °C, and left stirring overnight. TLC (SiO2/30% EtOAc in hexanes) showed complete reaction of the 4-azidocoumarin after 24 h. The mixture was partitioned between CH2Cl2 and
brine. The organic layer was separated, dried over anhydrous Na$_2$SO$_4$, and transferred to a round bottom flask. To this 200–300 mesh silica gel was added and the solvent was evaporated under reduced pressure. The silica gel with the adsorbed reaction mixture was loaded onto a 200–300 mesh silica gel column and eluted with a mixture of EtOAc in hexanes. Fractions containing pure material were combined, evaporated under reduced pressure, and vacuum dried. All 4-triazolyl coumarins were synthesized in a similar manner and any deviations from this procedure are noted under the individual compound headings.
4-(4-Phenyl-1H-1,2,3-triazol-1-yl)-coumarin (8). Chromatography using gradient elution with 25-35% EtOAc in hexanes gave 55 mg (95% yield from a 0.2 mmol reaction) of compound 8 as a light yellow solid. Rf (SiO2/30% EtOAc in hexanes) = 0.20. Melting point = 178–180 °C (lit.\textsuperscript{13} 166–168 °C). \textsuperscript{1}H (500 MHz, CDCl\textsubscript{3}): δ 8.22 (s, 1H, triazolyl-H), 7.98 (d, 1H, Ar-H, J = 8.3 Hz), 7.93 (d, 2H, Ar-H, J = 7.3 Hz), 7.68 (t, 1H, Ar-H, J = 7.8 Hz), 7.49 (m, 3H, Ar-H, J = 7.7 Hz), 7.41 (m, 2H, Ar-H), 6.62 (s, 1H, =C-H). \textsuperscript{13}C NMR (125 MHz, CDCl\textsubscript{3}): δ 159.7, 154.2, 148.5, 146.6, 133.5, 129.1, 129.1, 129.0, 126.0, 125.7, 125.0, 120.3, 117.5, 114.3, 109.6. HRMS (ESI/TOF): calcd for C\textsubscript{17}H\textsubscript{12}N\textsubscript{3}O\textsubscript{2} [M + H]\textsuperscript{+} 290.0924, found 290.0934.

4-(4-(p-Tolyl)-1H-1,2,3-triazol-1-yl)-coumarin (9). Chromatography using gradient elution with 20-30% EtOAc in hexanes to yield 53 mg (87% yield from a 0.2 mmol reaction) of compound 9 as a white solid. Rf (SiO2/30% EtOAc in hexanes) = 0.28. Melting point = 198–199 °C dec (lit\textsuperscript{13} 181–183 °C). \textsuperscript{1}H (500 MHz, CDCl\textsubscript{3}): δ 8.20 (s, 1H, triazolyl-H), 8.00 (d, 1H, Ar-H, J = 8.3 Hz), 7.82 (d, 2H J = 7.8Hz), 7.69 (t, 1H, Ar-H, J = 7.8 Hz), 7.48 (d, 1H, Ar-H, J = 8.3 Hz), 7.38 (t, 1H, Ar-H, J = 7.6 Hz), 7.30 (d, 2H, Ar-H, J = 7.8 Hz), 6.61 (s, 1H, =C-H), 2.42 (s, 3H, CH\textsubscript{3}). \textsuperscript{13}C NMR (125 MHz, CDCl\textsubscript{3}): δ 159.8, 154.3, 148.7, 146.7, 139.2, 133.5, 129.8, 126.2, 125.9, 125.7, 125.0, 119.8, 117.5,
4-(4-(4-Methoxyphenyl)-1H-1,2,3-triazol-1-yl)-coumarin (10). The reaction conditions for this compound were the same in all aspects except that the reaction was conducted at 80 °C. Chromatography using gradient elution with 30–40% EtOAc in hexanes to yield 51 mg (72% yield from a 0.2 mmol reaction) of compound 10 as a white solid. \( R_f \) SiO\(_2\)/30% EtOAc in hexanes = 0.21. Melting point = 196–198 °C dec (lit\(^{13}\) 183–185 °C). \(^1\)H (500 MHz, CDCl\(_3\)): \( \delta \) 8.14 (s, 1H, triazolyl-H), 8.02 (d, 1H, Ar-H, \( J = 7.8 \) Hz), 7.87 (d, 2H, Ar-H, \( J = 8.3 \) Hz), 7.70 (t, 1H, Ar-H, \( J = 7.8 \) Hz), 7.49 (d, 1H, Ar-H, \( J = 8.3 \) Hz), 7.39 (t, 1H, Ar-H, \( J = 7.6 \) Hz), 7.03 (d, 2H, Ar-H, \( J = 8.8 \) Hz), 6.62 (s, 1H, =C-H), 3.88 (s, 3H, -OCH\(_3\)). \(^{13}\)C NMR (125 MHz, CDCl\(_3\)): \( \delta \) 160.3, 159.8, 154.3, 148.5, 146.7, 133.5, 129.4, 127.4, 125.8, 125.0, 121.6, 119.2, 117.6, 114.5, 109.4, 55.4. HRMS (ESI/TOF): calcd for C\(_{18}\)H\(_{14}\)N\(_3\)O\(_3\) [M + H]\(^+\) 320.1030, found 320.1039.
4-(4-(Pyridin-3-yl)-1H-1,2,3-triazol-1-yl)-coumarin (11). The reaction conditions for this compound were the same in all aspects except that the reaction was conducted at room temperature (25 °C). Chromatography using gradient elution with 20–30% EtOAc in hexanes to yield 55 mg (95% yield from a 0.2 mmol reaction) of compound 11 as a white solid. \( R_f \) (SiO\(_2\)/30% EtOAc in hexanes) = 0.28. Melting point = 145–146 °C. \(^1\)H (500 MHz, CDCl\(_3\)): \( \delta \) 8.62 (m, 2H, Ar-H and triazolyl-H), 8.26 (d, 1H, Ar-H, \( J = 8.3 \) Hz), 7.92 (dd, 1H, Ar-H, \( J = 1.5, 8.3 \) Hz), 7.85 (t, 1H, Ar-H, \( J = 1.7, 7.7 \) Hz), 7.68 (t, 1H, Ar-H, \( J = 1.3, 7.8 \) Hz), 7.48 (dd, 1H, Ar-H, \( J = 1.0, 8.3 \) Hz), 7.37 (t, 1H, Ar-H, \( J = 7.7 \) Hz), 7.32 (ddd, 1H, Ar-H, \( J = 1.2, 4.9, 7.6 \) Hz), 6.67 (s, 1H, =C-H). \(^13\)C NMR (125 MHz, CDCl\(_3\)): \( \delta \) 159.6, 154.2, 149.6, 148.9, 148.7, 146.6, 137.2, 133.5, 125.3, 125.0, 123.6, 122.9, 120.6, 117.5, 114.2, 110.0. HRMS (ESI/TOF): calcd for C\(_{16}\)H\(_{12}\)N\(_4\)O\(_2\) [M + H]\(^+\) 291.0877, found 291.0888.

4-(4-(Thiophen-3-yl)-1H-1,2,3-triazol-1-yl)coumarin (12). Chromatography using gradient elution with 15-20% EtOAc in hexanes gave 53 mg (89% yield from a 0.2 mmol reaction) of compound 12 as a white solid. \( R_f \) (SiO\(_2\)/30% EtOAc in hexanes) = 0.33. Melting point = 185–187 °C dec. \(^1\)H (500 MHz, CDCl\(_3\)): \( \delta \) 8.13 (s, 1H, triazolyl-H), 7.98 (d, 1H, Ar-H, \( J = 8.3 \) Hz), 7.87 (d, 1H, thienyl-H, \( J = 2.0 \) Hz), 7.70 (t, 1H Ar-H, \( J = 7.8 \) Hz), 7.55 (d, 1H, thienyl-H,
$J = 5.1 \text{ Hz}$), 7.48 (m, 2H, Ar-H and thienyl-H), 7.39 (t, 1H, Ar-H, $J = 7.8 \text{ Hz}$), 6.62 (s, 1H, =C-H). $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta$ 159.7, 154.3, 146.7, 144.7, 133.6, 130.1, 127.0, 125.7, 125.7, 125.1, 122.7, 119.9, 117.6, 114.4, 109.6.

HRMS (ESI/TOF): calcd for C$_{15}$H$_{10}$N$_3$O$_2$S [M + H]$^+$ 296.0488, found 296.0488.

6-Bromo-4-(4-(2,4,5-trimethylphenyl)-1H-1,2,3-triazol-1-yl)coumarin (13).

Chromatography using gradient elution with 15–25% EtOAc in hexanes to yield 55 mg (67% yield from a 0.2 mmol reaction) of compound 13 as a pale yellow solid. $R_f$ (SiO$_2$/30% EtOAc in hexanes) = 0.40.

Melting point = 206–207 °C dec. $^1$H (500 MHz, CDCl$_3$): $\delta$ 8.29 (d, 1H, Ar-H, $J = 2.4 \text{ Hz}$), 8.08 (s, 1H, triazolyl-H), 7.77 (dd, 1H, Ar-H, $J = 2.2$, 8.8 Hz), 7.67 (s, 1H, Ar-H), 7.36 (d, 1H, Ar-H, $J = 8.8\text{Hz}$), 7.11 (s, 1H, Ar-H), 6.63 (s, 1H, =C-H), 2.49 (s, 3H, CH$_3$), 2.32 (s, 3H, CH$_3$), 2.30 (s, 3H, CH$_3$). $^{13}$C NMR (125 MHz, CDCl$_3$): $\delta$ 159.1, 153.2, 148.2, 145.5, 137.7, 136.4, 134.6, 132.8, 132.5, 130.0, 128.6, 125.4, 121.6, 119.2, 117.9, 115.8, 109.7, 20.9, 19.5, 19.2. HRMS (ESI/TOF): calcd for C$_{20}$H$_{17}$BrN$_3$O$_2$ [M + H]$^+$ 410.0499, found 410.0485.
6-Methyl-4-(4-(thiophen-3-yl)-1H-1,2,3-triazol-1-yl)coumarin  (14).

Chromatography using gradient elution with 15–20% EtOAc in hexanes gave 55 mg (89% yield from a 0.2 mmol reaction) of compound 14 as a white solid. R_f (SiO_2/30% EtOAc in hexanes) = 0.54. Melting point = 209–210 °C. 

\[ ^1H \text{ (500 MHz, CDCl}_3\text{): } \delta \ 8.11 \text{ (s, 1H, triazolyl-H), 7.87 (d, 1H, thienyl-H, } J = 2.9 \text{ Hz), 7.73 (s, 1H, Ar-H), 7.56 (dd, 1H, thienyl-H, } J = 1.2, 5.1 \text{ Hz), 7.46 (m, 2H, Ar-H, thienyl-H), 7.38 (d, 1H, Ar-H, } J = 8.3 \text{ Hz), 6.58 (s, 1H, } =\text{C-H), 2.42 (s, 3H, -CH}_3\text{).} \]

\[ ^{13}C \text{ NMR (125 MHz, CDCl}_3\text{): } \delta \ 160.0, 152.5, 146.7, 144.6, 135.7, 135.1, 134.7, 130.1, 127.2, 125.6, 125.2, 122.6, 119.9, 117.3, 114.0, 109.6, 21.1. \]

HRMS (ESI/TOF): calcd for C_{16}H_{12}N_{3}O_{2}S [M + H]^+ 310.0645, found 310.0653.

4-(4-(Cyclohex-1-en-1-yl)-1H-1,2,3-triazol-1-yl)-7-methylcoumarin  (15).

Chromatography using gradient elution with 15–20% EtOAc in hexanes gave 53 mg (86% yield from a 0.2 mmol reaction) of compound 15 as a white solid. R_f (SiO_2/30% EtOAc in hexanes) = 0.53. Melting point = 208–210 °C. 

\[ ^1H \text{ (500 MHz, CDCl}_3\text{): } \delta \ 7.84 \text{ (m, 2H, triazolyl-H, Ar-H), 7.23 (s, 1H, Ar-H), 7.15 (d, 1H, Ar-H, } J = 8.3 \text{ Hz), 6.72 (m, 1H, } =\text{C-H), 6.45 (s, 1H, } =\text{C-H), 2.48 (s, 3H, CH}_3\text{), 2.42 (m, 2H, CH}_2\text{), 2.25 (m, 2H, CH}_2\text{), 1.80 (m, 2H, CH}_2\text{), 1.70 (m, 2H, CH}_2\text{).} \]

\[ ^{13}C \text{ NMR (125 MHz, CDCl}_3\text{): } \delta \ 160.2, 154.3, \]
149.9, 146.8, 145.1, 127.3, 126.1, 125.9, 125.4, 118.7, 117.5, 111.7, 107.6, 26.2, 25.3, 22.2, 21.9, 21.7. HRMS (ESI/TOF): calcd for C_{18}H_{18}N_{3}O_{2} [M + H]^+ 308.1394, found 308.1397.
CONCLUSION

A successfully and safe two step one pot process for the azidization of 4-hydroxycoumarin was demonstrated in this thesis. The process involved the initially transformation of 4-hydroxycoumarin to its 4-benzotriazolyloxy coumarin with the used of inexpensive reagents like BOP, DBU and THF. Using this reaction scheme the formation of the benzotriazole ether bond was followed by addition elimination with sodium azide providing the azidization of the compounds.

The copper catalyzed alkyne azide cycloaddition were optimized by screening various copper catalysts and tetrakis(acetonitrile)copper(I) hexafluorophosphate, which contributed to better results in this reaction. Once the reaction was optimized, various 1,2,3-triazoles were synthesized with good to excellent yields with the formation of solely 1,4 substituted 1,2,3 triazoyl coumarin compounds. As previously stated in this paper, other authors have worked in the synthesis of leaving groups in the 4 position of coumarin compounds. However, synthesis of leaving groups such as 4-chloro and 4-sulfonate do not offer the versatility and ease of the two step one pot reaction that we have presented in this thesis.
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Pulse Sequence: s2pul
Solvent: CDCl3
Ambient temperature
Operator: mkl
File: Anthony_Azidocoumarin
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.892 Sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1. 499.7707095 MHz
DATA PROCESSING
FT size 32768
Total time 0 min, 15 sec
Pulse Sequence: a2pul
Solvent: CDC13
Temp. 23.4 °C / 296.6 K
Operator: mkI
File: Anthony_4aridocoumarin_carbon
INOVa-500 *trig*

Pulse 45.0 degrees
Acq. time 1.300 sec
width 25000.0 Hz
3304 repetitions
OBSERVE C13, 125.6674149 MHz
DECOUPLES H1, 499.7720084 MHz
Power 39 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 10 hr, 54 min, 41 sec
Pulse sequence: 92pul  
Solvent: CDCl₃  
Temp. 25.0 °C / 298.1 K  
Operator: mkl  
File: Anthony_RX851B_proton  
INOVA-500 "riga"

Pulse 24.7 degrees  
Acq. time 1.892 sec  
Width 8000.0 Hz  
8 repetitions  
OBSERVE H1, 499.7707065 MHz  
DATA PROCESSING  
FT size 32768  
Total time 0 min, 15 sec
Pulse Sequence: a2pul
Solvent: CDC13
Temp. 25.0 C / 298.1 K
Operator: mkl
File: AnthacyxRx851e_carbon
INOVA-500 "trigp"

Pulse 45.0 degrees
Acq. time 1.100 sec
Width 25000.0 Hz
1780 repetitions
OBSERVE C13, 125.667414[9] MHz
DECOUPLE H1, 459.77300384 MHz
Power 39 dB continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 5 hr, 27 min, 23 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 25.0 °C / 298.1 K
Operator: barbara
File: Anthony_RX84_02
INOVA.500 "riga"

Relax. delay 2.000 sec
Pulse 30.6 degrees
Acq. time 1.892 sec
width 8000.0 Hz
32 repetitions
OBSERVE MHz, 499.7707095 MHz
DATA PROCESSING
FT size 32768
Total time 2 min, 4 sec

![Chemical Structure](image)
Pulse Sequence: s2pul
Solvent: CDC13
Ambient temperature
Operator: ml
File: Anthony_RX04E_C13_01
INOVA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
40000 repetitions
OBSERVE C13, 125.6674149 MHz
DECOPLE WL 499.7730084 MHz
Power 39 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
PT size 65536
Total time 14 hr, 32 min, 55 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Ambient temperature
Operator: mkl
File: Anthony_RX48_01
INOVA-500 "riga"

Pulso 34.7 degrees
Acq. time 1.892 sec
Width 8000.0 Hz
8 repetitions
Observe H1, 499.7707302 MHz
DATA PROCESSING
FT size 32768
Total time 0 min, 15 sec
Pulse Sequence: a2pul
Solvent: CDCl3
Temp. 20.0 C / 293.1 K
Operator: mkI
File: Anthony RH88C Cl3
INOVA-500 *riga*

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
1720 repetitions

OBSRVR  C13, 125.6674149 MHz
DECOUPLE  H1, 499.7730084 MHz
Power 39 dB
continuously on
WALTZ-15 modulated

DATA PROCESSING
Line broadening 0.5 Hz
FT size 55536
Total time 290 hr, 58 min, 27 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Ambient temperature
Operator: mkl
file: Anthony_RX88_01
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.852 sec
Width 8000.0 Hz
9 repetitions
OBSERVE H1, 499.7707095 MHz
DATA PROCESSING
FT Size 32768
Total time 0 min, 15 sec
Pulse Sequence: s2pul

Solvent: CDCl3
Temp. 29.0°C / 293.1 K
Operator: skl
File: Anthony_RX05B_C13
INOMA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
640 repetitions

OBSERVE C12, 125.6674149 MHz
DECOUPLE H1, 499.7730084 MHz
Power 39 dB
continuously on
WALTZ-16 modulated

DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 290 hr, 58 min, 27 sec
Pulse Sequence: s2pul

solvent: CDCl3
Ambient temperature
Operator: mk1
File: Anthony_RX90B_proton
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.892 sec
Width 8000.0 Hz
8 repetitions
OBSERVE  H1, 499.7707095 MHz
DATA PROCESSING
PT size 32768
Total time 0 min, 15 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Ambient temperature
Operator: msli
File: Anthony_EX90B_Carbon
INOVA-500 "Riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
3584 repetitions
OBSERVE C13, 125.6674149 MHz
DECouple H1, 499.7730094 MHz
Power 39 db
CONTINUOSLY ON
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 14 hr, 0 min, 15 sec
Pulse Sequence: e2pul
Solvent: CDCl3
Ambient temperature
Operator: barbara
File: Anthony_RX06_02
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.892 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.7707326 MHz
DATA PROCESSING
FT size 32768
Total time 0 min, 15 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Ambient temperature
Operator: mk1
File: Anthony_EX86B_C13_01
INOVA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
3584 repetitions
OBSERVE C13, 125.6674149 MHz
DROUPE HL, 499.7730084 MHz
Power 35 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
PT spin 65536
Total time 14 hr, 32 min, 55 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 25.0°C / 298.1 K
Operator: mkl
File: Anthony_RX66C_Proton
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.892 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.7707095 MHz
DATA PROCESSING
FT size 32768
Total time 0 min, 15 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 25.0 C / 298.1 K
Operator: mkl
File: Anthony_RX66_Carbon
INOVA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
764 repetitions

OBSERVE C13, 125.6674149 MHz
DECCUPLE H1, 499.7730084 MHz
Power 39 db
continuously on

WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 9 hr, 27 min, 23 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Ambient temperature
Operator: mkl
File: Anthony_RX76C_proton
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.892 sec
Width 8000.0 Hz
0 repetitions
OBSERVE H1, 499.7707095 MHz
DATA PROCESSING
FT size 32768
Total time 0 min, 15 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Ambient temperature
Operator: mkl
File: Anthony_RX76C_Carbon
INOVA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25600.0 Hz
1600 repetitions
OBSERVE C13, 125.6674149 MHz
DECOUPLE H1, 499.7730084 MHz
Power 39 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 34 min, 55 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 25.0°C / 298.1 K
Operator: mk1
File: Anthony_RX77_01
INOVA-500

Relax. delay 2.000 sec
Pulse 38.6 degrees
Acq. time 1.892 sec
Width 9000.0 Hz
32 repetitions
OBSERVE H1 499.7707055 MHz
DATA PROCESSING
PT size 32768
Total time 2 min, 4 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 23.4 °C / 296.6 K
Operator: mkl
File: Anthony_RX77_carbon2
INOVA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
47440 repetitions
OBSERVE C13, 125.6674459 MHz
DECOUPLE H1, 499.7730084 MHz
Power 39 dB
continuously on
WALTZ-16 modulated

DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 145 hr, 29 min, 13 sec
Pulse Sequence: s2pul

Solvent: CDCl3
Ambient temperature
Operator: barbara
File: Anthony_RX87_01
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.892 sec
Width 8000.0 Hz
8 repetitions
Observe H1, 499.7707095 MHz
Data processing
FT size 32768
Total time 0 min, 15 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 20.0 C / 293.1 K
Operator: mk1
File: Anthony_BK97_C13
INOVA-500 "rign"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
1856 repetitions
OBSERVE C13, 125.6674149 MHz
DECOUPLE H1, 499.7730084 MHz
Power 39 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 290 hr, 58 min, 27 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Ambient temperature
Operator: mk1
File: Anthony_RX78C_proton
INOVA-500 “Riga”

Pulse 31.7 degrees
Acq. time 1.892 sec
Width 8000.0 Hz
8 repetitions
OBSERVK H1, 499.7707095 MHz
DATA PROCESSING
FT size 32768
Total time 0 min, 15 sec
Pulse Sequence: e2pul

Solvent: CDCl3
Temp. 25.0 °C / 298.1 K
Operator: mk1
File: Anthony_RX78C_Carbon3
INOVA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
6580 repetitions
OBSERVE 13C, 125.6674499 MHz
DECUPLE 1H, 499.7730084 MHz
Power 39 dB
continuously on
WALTZ-16 modulated

DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 21 hr, 49 min, 23 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 25.0 C / 298.1 K
Operator: mk1
File: Anthony_RX84C_proton3
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.902 sec
Width 8000.0 Hz
8 repetitions
OBSERVE H1, 499.7707095 MHz
DATA PROCESSING
FT size 32768
Total time 0 min, 15 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 25.0 C / 298.1 K
Operator: mk1
File: Anthony_RX06C_Carbon
INOVA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
3244 repetitions
OBSERVE C13, 125.6674149 MHz
DECOUPLE H1, 495.7730084 MHz
Power 39 dB
continuously on
WALTS-16 Modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 9 hr, 27 min, 23 sec
Pulse Sequence: s2pul
Solvent: CDCl3
Temp. 25.0 C / 298.1 K
Operator: mkl
File: Anthony_RX88C_Proton_Clean
INOV-500 "riga"

Relax. delay 2.000 sec
Pulse 38.6 degrees
Acq. time 1.992 sec
Width 8000.0 Hz
32 repetitions
OBSERVE H1, 499.7707095 MHz
DATA PROCESSING
PT size 32768
Total time 2 min, 4 sec
STANDARD CARBON PARAMETERS

Pulse Sequence: a2pul
Solvent: CDCl3
Temp. 25.0 °C / 298.1 K
Operator: mik
File: anthoxy-RX06C_Carbon2
INOV-500 'riga'

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
49376 repetitions
OBSERVE C13, 125.6674149 MHz
DECOUPLE H1, 499.7730084 MHz
Power 30 dB
continuously on
WALTZ-16 modulated
DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 21 hr, 49 min, 23 sec

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![Chemical Structure Image]

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ppm
Pulse Sequence: s2pul

Solvent: CDCl3
Ambient temperature
Operator: mkl
File: Anthony_RX89C_F1
INOVA-500 "riga"

Pulse 34.7 degrees
Acq. time 1.892 sec
Width 8000.0 Hz
0 repetitions
OBSERVE H1, 499.7707095 MHz
DATA PROCESSING
FT size 32768
Total time 0 min, 15 sec
Pulse sequence: s2pul

Solvent: CDCl3
Temp. 15.0 °C / 288.1 K
Operator: mkl
File: Anthony_rx99c_c13
INOVA-500 "riga"

Pulse 45.0 degrees
Acq. time 1.300 sec
Width 25000.0 Hz
4000 repetitions
OBSERVE C13, 125.6674149 MHz
DECOUPLE H1, 499.7730084 MHz
Power 29 dB
continuously on
WALTZ-16 modulated

DATA PROCESSING
Line broadening 0.5 Hz
FT size 65536
Total time 1 hr, 27 min, 17 sec
ANTHODI-TRIAZONE-NODY

Pulse Sequence: NOESY
Solvent: CDCl3
Temp, 25.0 C / 298.1 K
Operator: mkl
File: Anthony_NOESY_1,5-1-28-14
INOVA-500 "riga"

Relax. delay 1.000 sec
Mixing 0.400 sec
Acq. time 0.171 sec
Width 5074.6 Hz
2D Width 5074.6 Hz
48 repetitions
2 x 240 increments
Observe 49, 77, 72, 28 MHz
DATA PROCESSING
Gauss apodization 0.079 sec
F1 DATA PROCESSING
Gauss apodization 0.031 sec
FT size 2048 x 2048
Total time 10 hr, 18 min, 46 sec
Pulse Sequence: NOESY
Solvent: CDCl3
Temp. 25.0 °C / 298.1 K
Operator: mk
File: ANTHONI-1_4_TRIAZOLE_major_NOESY
INOVA-500 "Figm"

Relax. delay 1.000 sec
Mixing 0.400 sec
Acc. time 0.171 sec
Width 5974.6 Hz
2D Width 5974.6 Hz
32 repetitions
2 x 256 increments
OBSERVE Hi 459.7707226 MHz
DATA PROCESSING
Gauss apodization 0.079 sec
F1 DATA PROCESSING
Gauss apodization 0.031 sec
F1 size 2048 x 2048
Total time 7 hr, 20 min, 33 sec