Siena College logo TM

**Department of Chemistry and Biochemistry**

Phone 518-783-2440 • Fax 518-783-2986

www.siena.edu

515 Loudon Rd

Loudonville, NY 12211

ekolonko@siena.edu

August 13, 2018

Dr. Bing Wu, Review Editor

*Journal of Visualized Experiments*

1 Alewife Center Suite 200

Cambridge MA 02140

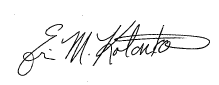
Dear Dr. Wu,

I am submitting a revised version of the manuscript JoVE58803 entitled "Synthesis of esters via a greener Steglich esterification in acetonitrile" for consideration for publication in the *Journal of Visualized Experiments*.

We appreciate the comments and suggestions from the peer and editorial reviews and have amended the manuscript to address the issues raised. A summary of the comments and the specific changes that we have made to the manuscript is provided on the accompanying pages.

We thank you for the opportunity to contribute a revision to the *Journal of Visualized Experiments*.

Sincerely,



Erin M. Kolonko

Assistant Professor

Department of Chemistry and Biochemistry

Siena College

ekolonko@siena.edu

(518)-782-6956

**Response to reviewer major and minor concerns for JoVE58803 “Synthesis of esters via a greener Steglich esterification in acetonitrile”**

**Editorial comments:**  
1. Please take this opportunity to thoroughly proofread the manuscript to ensure that there are no spelling or grammar issues.

*Response:* We have thoroughly proofread the manuscript for spelling and grammar issues, to the best of our knowledge.

2. Please rephrase the Long Abstract to more clearly state the goal of the protocol.

*Response:* We have added a specific statement of the goal to the Long Abstract. (Lines 37-38)

3. Please rephrase the Introduction to include a clear statement of the overall goal of this method.

*Response:* We have added a specific statement of the goal to the introduction. (Lines 51-53)

4. JoVE cannot publish manuscripts containing commercial language. This includes trademark symbols (™), registered symbols (®), and company names before an instrument or reagent. Please remove all commercial language from your manuscript and use generic terms instead. All commercial products should be sufficiently referenced in the Table of Materials and Reagents. You may use the generic term followed by “(see table of materials)” to draw the readers’ attention to specific commercial names. Examples of commercial sounding language in your manuscript are: Bruker AVANCE, Bruker Maxis Impact HD, etc.

*Response:* We have removed commercial language regarding instrumentation and replaced the information with generic terms.

5. Please revise the protocol (lines 96-105, etc.) to contain only action items that direct the reader to do something (e.g., “Do this,” “Ensure that,” etc.). The actions should be described in the imperative tense in complete sentences wherever possible. Avoid usage of phrases such as “could be,” “should be,” and “would be” throughout the Protocol. Any text that cannot be written in the imperative tense may be added as a “Note.” Please include all safety procedures and use of hoods, etc. However, notes should be used sparingly and actions should be described in the imperative tense wherever possible.

*Response:* We have revised the "General remarks" section to use only imperative tense. (Lines 105-109)

6. 2.1.1: Please mention how complete reaction is confirmed by TLC.

*Response:* We have added a line in the protocol note to more clearly explain when a complete reaction by TLC is determined. (Lines 122-124)

7. 3.1.5: Please add more details here about how to prepare an NMR sample and how to perform the NMR experiments. Alternatively, add references to published material specifying how to perform the protocol action.

*Response:* We have added a reference for preparing an NMR sample to the protocol. (Line 176)

8. Because Table 1 shows data from mass spectrometry, please mention in the protocol that mass spectrometry is performed on the products.

*Response:* We have added the recommended statement. (Line 173)

9. Lines 190-198: Please consider describing such modifications in the protocol.

*Response:* We have added Sections 4-5 to the protocol to describe the specific modifications to the protocol. (Lines 179-207)

10. References: Please do not abbreviate journal titles. Please include page numbers for all journal references.

*Response:* We have corrected the reference section as requested.

**Reviewer #2:**  
Major Concerns:  
If the major thrust of the paper is extending the reaction utility to ACN solvent, then I would like to see a better review of what has been done. I find the references somewhat lacking. The use of carbodiimide procedure published by Steglich in 1978 has been cited over 900 times since then. A more comprehensive review of the utility of the reaction and the potential benefit of acetonitrile as a solvent would strengthen the manuscript. The following papers could be referenced:  
  
Nat. Prod. Rep., 2015,32, 605-632 "Ester coupling reactions - an enduring challenge in the chemical synthesis of bioactive natural products"  
  
Org. Biomol. Chem., 2015,13, 2393- 2398 "Peptide synthesis beyond DMF: THF and ACN as excellent and friendlier alternatives"  
  
*Response:* We agree that these additions will strengthen the manuscript. In the introduction, we have added additional statements regarding the Steglich esterification in general and the use of acetonitrile for this reaction, along with corresponding references. (Lines 45-52 and 72-76, References 4-6, 13-21)

Minor Concerns:  
The major utility of the Steglich esterification over other methods is the ability to generate esters from carboxylic acids in the presence of acid labile moieties. For example, tertiary alcohols not accessible by Fischer esterification can be made by the Steglich using CH2Cl2; however, this remains a limitation in this modification (as the authors point out). In addition, the acid work up minimizes the potential for a broader utility and scope. This submission adds a few representative acids to generate a small library of esters that may be of interest to some above and beyond to the work already reported in the Biorg Med Chem paper.

*Response:* We agree that there are limitations to the method in regard to tertiary alcohols (described previously in the text) and potentially to other acid-labile groups. We have added a statement of the potential limitation regarding acid-labile groups for the acid wash steps in the introduction. (Lines 92-94). While this could limit the utility of the wash sequence in lieu of column chromatography for some compounds, it does not limit the use of acetonitrile as a greener reaction solvent.

**Reviewer #3:**  
Minor Concerns: Will this protocol succeed for dicarboxylic acid esterification. The author can try and report reactions of succinic acid and 1.3 -acetone dicarboxylic acid

*Response:* We are interested in this application and plan to investigate the possibility in the future. We feel, however, that it is outside the scope of the current protocol.