Laboratory Evaluation for
Combinatorial Chemistry: Antibiotic Drug Discovery


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Evaluation

Combinatorial Chemistry: Antibiotic Drug Discovery

This is a great experiment at the interface of chemistry and biology (and/or microbiology). When carefully conducted, the results are clear, with two of the eight quadrants clearly showing an inhibition zone.

The experiment requires only 1/2 to 1 hour in the first lab period, with a 24 hour incubation period for growth of the bacteria and 15-30 minutes after the incubation for viewing of results. The procedure effectively illustrates synthesis of hydrazones, applications of combinatorial chemistry, some typical microbiological techniques, and the importance of carefully following instructions. Green chemistry concepts include atom economy and no heating required for the formation of the hydrazones, screening of several compounds for activity in an efficient manner, minimal amounts of waste, and use of water as a solvent for the reagents.

When conducted carefully and with non-toxic strains of *E. coli*, there are no safety issues in addition to the ones mentioned in the laboratory text (wear gloves, goggles, use disinfectant on the bench top, and wash hands prior to leaving the lab).

The experiment works well as written, although some modifications can be made. Because the wells only hold 2-3 drops of liquid, the number of drops of each reagent could be cut in half when preparing the hydrazone mixtures (M1-M8). This would also have the benefit of reducing the amount of waste. One could test the starting hydrazines and aldehydes for antibiotic activity as an example of a control experiment. Based on our experience, use of short Pasteur pipettes rather than longer pipettes for transfer of the hydrazone mixtures to the wells is recommended. Some participants suggested swabbing the agar plate with the bacterial culture rather than pouring the culture on the plates. A good pre-lab lecture may be necessary, especially for the non-biologically oriented students, to ensure that students know what to look for in terms of evaluating the antibiotic action of the hydrazones. In the pre-lab instructions, students should be asked to draw the structures of each hydrazone that will be formed. As part of the lab report, students should be asked to specifically identify the hydrazone(s) that were responsible for the antibiotic action. Additionally, since one of the mixtures is not highly colored, interested students could be encouraged to comment on the reason for the lack of color (one of the hydrazine starting materials is not aromatic, so no extension of conjugation occurs).

This experiment very effectively and interestingly illustrates the principles of both combinatorial and green chemistry. Additionally, it offers the opportunity to collaborate with a microbiologist at
your school in the preparation of the agar plates and the \textit{E. coli} culture, incubation of the bacteria, and autoclaving of the used plates. The instructions are clear and the experimental results are clear. Having to wait to find out the results may be frustrating for some students, and some students may be uncomfortable working with bacterial cultures. On the other hand, the vast majority of students in a typical organic class will really like the idea of synthesizing and testing antibiotics, using "real-world" pharmaceutical techniques. Overall, this experiment is a winner!