A distance learning enzyme assay and kinetics laboratory in the time of COVID-19

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Abstract
This article describes a straightforward approach to deliver an enzyme assay and kinetics laboratory via online delivery methods in the time of COVID-19.

1 | INTRODUCTION

Following the pandemic crisis with COVID-19 in early 2020, we had to offer all the courses via the on-line mode of delivery. One of the courses that I teach, that is, Biochemistry for third-year students involves a laboratory section. The main objectives of this laboratory are to give basic information on how to purify enzymes, develop enzyme assays, and perform kinetic studies. Before the COVID-19 lockdown students learned (hands-on experience) how to: (a) use the Bradford assay to determine the protein concentration of an unknown solution, (b) prepare buffers, calculate absorbencies, concentrations, using the Lambert–Beer’s law, and (c) make standard curves and determining concentrations from then. Moreover, students purified lactate dehydrogenase (LDH) from bovine muscle heart using the salting-out method.

2 | SET-UP OF AN ONLINE BIOCHEMISTRY LABORATORY

Instead of postponing the course I decided to deliver the laboratory via distance learning while I managed to cover all the learning objectives of the course, that is, to provide sufficient information to students to: (a) determine the specific activity of enzyme fractions and construct a purification table, (b) construct velocity ($V$) versus substrate ($S$) (Michaelis–Menten) and Lineweaver–Burk plots to estimate/determine $K_M$ and $V_{max}$, (c) determine the kinetic parameters $K_M$ and $V_{max}$ using the nonlinear regression method, and (d) perform some basic statistical tests.

It should be pointed out that this online laboratory aimed to improve the skills of students on how to interpret scientist data, transform raw data into results, and present them in the form of tables and/or figures. In detail, I gave a scenario to the students mentioning that we need high amounts of LDH from bovine muscle heart to use them as a control in various experiments, and I also gave them a set of data to calculate specific parameters (discussed further below). In this scenario, LDH activity was determined by measuring the absorbance at 340 nm. Students will submit a lab report that should contain (a) a detailed description of the protocols/methods that should be employed to measure the activity of LDH and to perform the kinetic studies and (b) the appropriate figures and tables. Students should also have to explain and discuss their results.

3 | ONLINE DELIVERY OF THE LABORATORY

I gave online lectures on how to determine enzyme activity and kinetic parameters as well as on how to
use Microsoft (MS) Excel to prepare the abovementioned graphs and to calculate the kinetic parameters, while students were instructed to include data points on the graphs superimposed on the calculated curves. Moreover, I gave an extensive lecture on basic descriptive statistical functions (e.g., mean, variance) and on how to perform simple statistical tests (e.g., Student's *t*-test) to compare different results using MS Excel. I also used various online virtual protocols from the Journal of Visualized Experiments (JoVE) and other resources, for example, Khan Academy (https://www.khanacademy.org/).

Subsequently, a different set of experimental (raw) data was given to each student in an excel file. Students were asked to (a) determine the specific activity of enzyme fractions and construct a purification table for the purification of LDH, and (b) calculate the aforementioned kinetic parameters. To this end, each student was assigned with a different series of data in an Excel file containing raw data to: (a) draw a standard protein curve (Bradford method), (b) calculate the protein concentration and LDH activity in each fraction during the purification of the enzyme, (c) construct a purification table, and (d) draw the Michaelis–Menten and Lineweaver–Burk to determine the kinetic parameters. Furthermore, students had to determine the kinetic parameters from the raw data by nonlinear regression using MS Excel (the appropriate spreadsheet was given to the students).

Subsequently, they had to compare the results obtained by the two methods using the Student's *t*-test to understand that the same data can lead to different numbers depending on the method that is used to analyze the data. Detailed instructions were also given to the students as summarized in Figure 1.

4 | CONCLUSIONS

Even though it is very challenging to deliver laboratory content using online methods, the students find the approach described herein very helpful because they learned how to manipulate experimental data. Students will submit a lab report while further assessment using a virtual lab will take place.

CONFLICT OF INTEREST

The author declared no potential conflict of interest.

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How to cite this article: Papanoptyou C. A distance learning enzyme assay and kinetics laboratory in the time of COVID-19. Biochem Mol Biol Educ. 2020;48:430–432. https://doi.org/10.1002/bmb.21364