

# AN INQUIRY-BASED LEARNING (IBL) APPROACH TO MOLECULAR BIOLOGY FOR BIOTECHNOLOGY UNDERGRADUATE STUDENTS

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**Abstract.** *Recent studies have indicated that conventional modes of instruction, lecture based approaches, are ineffective in harnessing necessary skills for future science professionals. Authentic research experiences lead by undergraduates in science courses are becoming more common and are thought to be more effective in developing essential competencies for Science investigation. Based on these experiences, we propose a modular protocol for practical laboratory sessions based on an inquiry-based protocol.*

*During these sessions, students had the opportunity to plan and to conduct, in groups of two to three, different research projects about a central theme in Molecular Biology. As a culminating assessment, students write their own papers, resembling established publishing criteria for science research international journals and work on typical steps of the scientific writing process.*

*From the satisfaction surveys, we conclude that results were positive in terms of their perceived value of this experience for their education (as measured in questionnaires and written reflections). This activity has helped students experience first hand the strengths, limitations and complex realities of science research, and how communities of scientists come to establish the validity of knowledge. Inquiry-based learning may be a feasible alternative to traditional lab led protocols in Science courses.*

**Palabras clave:** Inquiry-based learning, integrated laboratory classes, molecular genetics, student learning, laboratory procedures

## 1. INTRODUCTION

Most colleges include traditionally segregated Science courses, where a lecture is separated from laboratory classes and where students receive a laboratory guidebook that they must follow without necessarily thinking (Matsou *et al.*, 2011). Standardized laboratory guidebooks are characteristic of introductory biology courses. These protocols are designed to teach students basic procedures, providing them with practical concepts representative of the subject that they learn during the lecture classes. They also give students hands-on laboratory experience. However, these experiences do not emphasize development of experimental skills or knowledge integration. These difficulties highlight the need of reorganize these sessions in order to fill the gap found between laboratory classrooms and authentic

research experiences. Some authorities such as the National Research Council in USA have reported that these traditional experiences do not provide students with a real understanding of science processes and that science education should be transformed in inquiry-based and project-based laboratory courses that could stimulate students to think more like scientist (reviewed in Treacy et al., 2011).

Many inquiry-based courses have been described in the past, where students participate in semester-long guided research projects planned around specific learning objectives (Lopatto *et al.*, 2008). These studies have shown that student retention of key concepts is increased when inquiry or project-based experiences are implemented (Lord and Orkwiszewski, 2006). Protocols must include the development of critical thinking skills and strategies that students need to develop their own experiments. This implementation has been extremely successful in increasing student interest in content areas such as bioinformatics and molecular biology (D'Costa and Sheperd, 2009; Lau and Robinson, 2009).

Considering these facts, we have designed a Molecular Biology modular introductory lab course at the European University of Madrid into a trimester-long, project based laboratory using an inquiry strategy. The main objectives of this work were:

- To introduce students to molecular biology techniques within an inquiry-based framework.
- To promote critical thinking skills through having students develop her or his own protocol instead of using a professor-designed one.
- To provide the students their first opportunity to know how research actually happens in a lab.
- To introduce students to scientific writing and the scientific publication process.

Finally, student perceptions and experience with the activity was collected with two questionnaires and open questions.

## **2. MATERIALS AND METHODS**

### **2.1. Course description**

The activity was developed during the academic year 2012-2013. The objective was to provide our 2<sup>nd</sup> year Biotechnology students with a hands-on example of a molecular genetics project using different techniques under the course “Molecular Genetics”. The group of 10 students enrolled was divided in 4 groups of 2/3 members.

### **2.2. The class research project**

This inquiry-based lab is designed around genetic engineering applications for recombinant protein production. To allow students to work on their own research projects, 4 different projects with 4 different proteins were developed. Students were grouped in pairs or teams of three. The list of research projects are listed in **Table 1**.

Title	Number of students
Cloning of Par6 gene from <i>Drosophila melanogaster</i>	3 students
Cloning of Bazooka/Par3 gene from <i>Drosophila melanogaster</i>	2 students
Cloning of Cdc42 gene from <i>Drosophila melanogaster</i>	2 students
Cloning of aPKC gene from <i>Drosophila melanogaster</i>	3 students

*Table1. List of research project ideas provided to the students*

### 2.3. Organization of Laboratory Activities

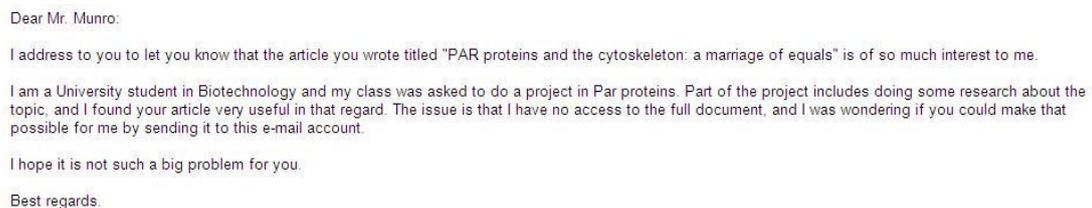
The course was divided in two modules: a Biocomputing module, which includes a bibliography search in biomedical databases and an introduction to bioinformatics, and an experimental module based on classical molecular cloning techniques. The timeline of the modules was established for 10 hours of student work during two weeks.

#### 2.3.1. *Biocomputing sessions:*

The first day students received the topic of their project and started the Biocomputing sessions. This represents a guided approach to inquiry which we feel is more appropriate to our time constraints of a trimester course.

- **Bibliography search:**

Over two computational laboratory sessions, students use tools as bibliography databases: PudMed and Medline as well as specific databases and programs. Student began by reading literature related to their project. They were also asked to collect at least two research articles included in the JCR. If the article was not available in the University repositories to be downloaded, students were encouraged to contact directly the authors (see example of student mail to the author in Figure 1).



Dear Mr. Munro:

I address to you to let you know that the article you wrote titled "PAR proteins and the cytoskeleton: a marriage of equals" is of so much interest to me.

I am a University student in Biotechnology and my class was asked to do a project in Par proteins. Part of the project includes doing some research about the topic, and I found your article very useful in that regard. The issue is that I have no access to the full document, and I was wondering if you could make that possible for me by sending it to this e-mail account.

I hope it is not such a big problem for you.

Best regards.

*Figure 1. Example of email written by a student to one well-known scientist*

- **Bioinformatics, databases search, primer design and virtual PCR:**

DNA sequence of the exons (CDS) assigned genes were extracted from the *Drosophila melanogaster* Database Flybase ([www.flybase.org](http://www.flybase.org)). Using these sequences, students designed PCR primers from CDS using the online program Primer3Plus (<http://frodo.wi.mit.edu>). Primers were synthesized by Sigma-Aldrich, Spain.

Primers and the size of the expected amplified DNA were analyzed using a virtual PCR program that simulates experimental conditions:  
([http://www.ch.embnet.org/software/iPCR\\_form.html](http://www.ch.embnet.org/software/iPCR_form.html)).

### 2.3.2. *Molecular laboratory sessions:*

During the laboratory sessions, the molecular biology techniques used by the students were: PCR amplification, DNA digestion, DNA ligation and Agarose Electrophoresis Gel.

- **cDNAs templates and cloning vector:**

The cDNAs genes for PCR amplification and the expression plasmid pET15-b were kindly provided by Prof. Jose Maria Carazo Laboratory at National Centre of Biotechnology (Madrid, Spain).

- **PCR amplification:**

Par6, Cdc42, aPKC y Baz genes were amplified by PCR. PCR conditions consisted of initial denaturation at 94° for 2 min followed by 30 cycles of 94° for 30 sec, annealing at 65° for 1 min, and extension at 72° for 7 min.

The sequence of PCR primers were designed using Primer3:

Gene	Forward 5'-3'	Reverse 5'-3'
aPKC	ATTCTCTAGATCAGACGCAATCCTCCAGAGAC	CCCATCTAGAATGCAGAAAATGCCCTCGCAAATTC
Par6	GCCGTCTAGACTACAAATGCAGCACTCCATCC	ATTCTCTAGAATGTGGAAGAACAAGATAAACACAACG
Baz	ACACTCTAGATCACACCTTGGAGGCGTGTGGC	AAAGTCTAGAATGAAGGTCACCGTCTCCTTCC
Cdc42	GAAATCTAGATTATAAGAATTTGCACTTCTTTTCTTGTGGGC	TCTGTCTAGAATGCAAACCATCAAGTGCCTGG

*Table2. List of the Primers used in the amplification reaction.*

For the PCR mix students used the PfuI enzyme provided by Biotools and for the reaction preparation the supplier conditions. PCR results were visualized on 1.0% agarose gels as the one showed in the Figure 2.

- **Restriction and ligation reactions:**

Students performed their restriction and ligation reactions following the manufacturer's procedures. Every group individually calculated their reactions conditions according to their specific DNA concentrations. Enzymes were obtained from NEB.

## 2.4. Paper preparation and evaluation

Students received very specific guidelines about writing a scientific paper and a paper template as can be found in the author's guidelines for international scientific journals (example of Guidelines in Figure 3). The objective was to introduce students to a real experience in scientific publication. The paper submission was sent with a cover letter directed to the reviewer, in this case, the professor of the course.

Students had the opportunity to present a preliminary draft of the paper previous to the final one.

### **Written Scientific Paper Guidelines**

Use the following outline to guide your writing.

#### **General considerations:**

This article will be written (using the third person, font 12 and single spacing, as a group of two, following the laboratory arrangement, however, you should hand in a coherent article, which is in print and **formatted appropriately (no pen drives or attachments of emails will be collected)**. **Please allow enough time for printing and putting together the document as late papers will not be accepted.**

#### **A. Title: (0.5 points)**

It should be concise but detailed and specific and include all information about the study, including the design of the project. Remember that it is the abstract and the title that are available in Medline and that it is with the information in the title that makes the article retrieval sensitive and specific. Below the title we should find the name of the authors with the email address of the corresponding author.

*Figure 3. Example of scientific guidelines provided by the teacher*

## **2.5. Student evaluation survey**

Students were asked to complete two surveys: one survey for the project evaluation and some written reflections about the activity and the work done inside the group. The objective was to assess how students feel about the inquiry-based process and to find out what is their experience in writing their papers.

## **3. RESULTS**

### **3.1. Student results:**

#### **3.1.1. Experimental results:**

Because students were performing experiments that did not have set expected results, they were required to think more about how to perform the experiments and why the results were not successful and to think about ways to improve the protocol. This requires pre and post-lab questions so they were requested to do a preliminary report. The work done in the lab was satisfactory although students realize that most of the time it is necessary to do some small modifications in the protocol in order to improve experimental results. A representative image of the work done by students is shown in Figure 4.

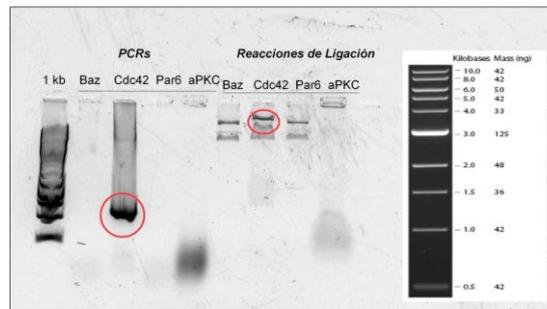


Figure 4. Representative student-produced agarose gel electrophoresis

### 3.1.2. Student ended projects:

After the two weeks experimental and computational work, students were requested to present their research results in a scientific article format. The process to prepare the article was similar to the ones established by a publication protocol for a scientific journal. In Figure 5 we can see some examples of the final papers written by students.

#### CLONING OF PAR-6 GEN IN DROSOPHILA MELANOGASTER BY RECOMBINANT PLASMID USING GENETIC ENGINEERING

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#### Abstract

In this study, different molecular biology procedures were employed to

derived from experiments done in the fruit fly *Drosophila melanogaster*. In *Drosophila*, neuroblasts use distinct mechanisms of asymmetric cell division to generate cellular diversity (Goulas,

#### Cloning of Bazooka/Par 3 cell polarity protein of *Drosophila melanogaster*

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Development: Mar 12, 2013

#### ABSTRACT

The scaffold protein Bazooka (Bar; *Drosophila* Par-3) plays a role in the cell polarity determination of many cells types. Its principal function is as a landmark in the plasma membrane, organizing the apical domain for spots adherens junction (AJ), which are assembly in the "*Drosophila*" embryonic. Bazooka localizes below PAR-6 and aPKC at the apical lateral junction. To obtain its recombinant protein, it is necessary to introduce its coding sequence in an expression vector of "*E. coli*" using a restriction enzyme protocol. For reaching this goal, several techniques had been used to ensure that it has been produced a recombinant vector. Database search, PCR amplification and digestion reactions were

#### Role of Cdc42 on Asymmetrical division of *Drosophila* neuroblasts and cloning protocol

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#### Abstract

#### Obtaining and studying aPKC from *Drosophila melanogaster*.

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Biotechnology students from European University of Madrid.

#### Abstract

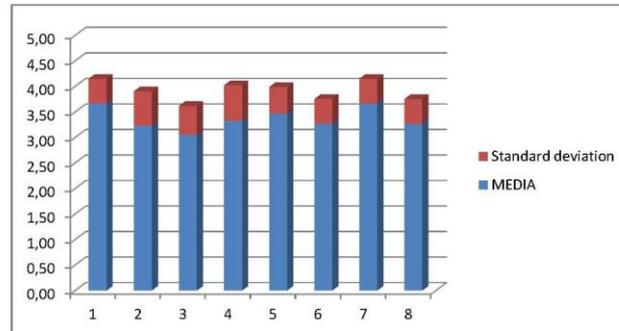
A project has been done to obtain and study aPKC protein from *Drosophila melanogaster* in asymmetric cell division. aPKC is a protein required for spindle planar orientation during asymmetric cell division and to exclude basal proteins from the apical cortex interacting with Par-6 and Par-3 making a complex (The Par-Complex).

For the study it has been done a PCR, DNA digestion and ligation followed by an electrophoresis.

Figure 5. Examples of student ended projects

### 3.1.3. Students' satisfaction:

Student evaluation was satisfactory. In Figure 6, we can see that all items for the project evaluation survey were ranked above in the top part of the graphic. Most students agree that the most interesting points of the experience were the Biocomputing sessions and the possibility to present a draft before the final paper was due.



1. The Bibliography review and the bioinformatic sessions helped me to know the objective of the practical sessions before going to the lab
2. The bibliography sessions helped me to define my project
3. It was easy to localize bibliography sources
4. Writing a scientific paper helped me to know which are the main parts of a scientific publication
5. The elaboration of the discussion allowed me to think about the experimental problems I had in the lab and in the way of improved them
6. The rubric and the paper guidelines were useful
7. I had the opportunity to send a draft before the final paper
8. I would like to do more practical sessions following this format

Figure 6. Student questionnaires and the students replies.

From the written reflections we can say that most of students stated that time management was the less positive aspect of the working group. The most significant positive aspect was the fact to be introduced to the writing scientific communication procedures.

#### 4. DISCUSIÓN

We have presented here our inquiry based experience for a Biotechnology Degree undergraduate class. From experimental results, written papers quality and satisfaction surveys from students we can say that this experience has been quite satisfactory. For this reason, this modular course may serve as a model for other research courses to train undergraduates' students in the life sciences.

We suggest that the steps to adapt this protocol to other introductory undergraduate science projects are: first, identification of a biological or medical problem related to student's research interest or expertise that constitute a key experiment in the discipline; second, to develop a research project that includes these concepts for the research laboratory, and finally, to encourage students to work over the protocol to improve the results within an inquiry-based approach. The objective is to move from traditionally designed laboratory protocols into new IB research modules.

In our experience, class projects also provide opportunities for class discussions that can lead to a deeper understanding of biological concepts. Considering our modest findings it is clear that projects-based learning can be an effective means of preparing students for future research labwork.

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