



Article Lab-It Is Taking Molecular Genetics to School

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Abstract: The Molecular Genetics Mobile Lab or "Laboratório itinerante de Genética Molecular" (Lab-it) was funded in 2008 by Leonor Cancela to promote the learning of molecular genetics which had been introduced at that time into high school biology programms. The project aimed to introduce hands-on laboratory activities in molecular genetics to complement the theoretical concepts taught in school. These included the development of experimental protocols based on theoretical scenarios focusing on themes of forensics sciences, biomedical applications, diagnostic methods, and ecological research using basic molecular biology techniques, such as DNA extraction, polymerase chain reaction (PCR), electrophoresis, and restriction enzyme application. In these scenarios, the students execute all the procedures with the help of the Lab-it instructor and using the Lab-it equipment, followed by a discussion of the results with all the participants and the school teacher. These approaches help the students to consolidate the concepts of molecular biology and simultaneously promote discussions on new advances in the area and choices for university careers. In addition to practical sessions, Lab-it also promotes seminars on topics of interest to the students and teachers. Since 2008, 18 high schools have participated in the region of Algarve, averaging each year about 400 students participating in practical activities. In 2021, despite the COVID pandemic, 9 schools and 379 students were involved in Lab-it practical sessions and 99% of them considered the activity to contribute to better understanding the molecular biology methods approached in theoretical classes and expressed high interest in those sessions.

Keywords: education; molecular genetics; lab-it; laboratory hands-on sessions

1. Introduction

With the introduction of biochemistry and molecular biology concepts in biology high school teaching programs all over the world [1–4] and particularly in Portugal, teachers have been reporting difficulties in teaching students the concepts associated with the flow of genetic information in cells and the molecular biology methods currently used for biotechnology applications [5–8]. Students have reported difficulties in understanding the correlation of biomolecules like deoxyribonucleic acid (DNA), messenger ribonucleic acid (mRNA), and proteins with cell functions, and basic concepts like genes, proteins, and genetic phenotypes are often not fully understood [6,8]. Biochemistry and molecular biology concepts are somehow abstract because they occur at the molecular level of biological systems. Available information indicates that students find problems to understand those concepts due to the degree of complexity and poor contextualization related to the flow of genetic information due to the increased number of concepts approached by the scientific program of biology classes and the time available to students to learn it [4,6,7]. In addition,



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Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). misconceptions related to the teacher's preparation and lower degree of training updates were identified as contributing factors [1,7,8]. To complement formal learning taking place within the framework of classes and based on the description of theoretical concepts by the teacher, the introduction of practical projects associated with these subjects has proven to help students understand their complexity [8–10].

To optimize the teaching of molecular biology concepts included in the high school biology program in Portugal, several teachers started to contact the universities to help them refresh their know-how in those topics. Attending to this, in 2008, Leonor Cancela, professor of molecular genetics at the University of Algarve, started the project called The Molecular Genetics Mobile Lab or "Laboratório itinerante de Genética Molecular" (Lab-it) to help schools and biology teachers in the Algarve region to explain molecular biology concepts to their students. Attending that the equipment necessary to execute molecular biology experiments was not accessible to schools, the idea was to equip an itinerary laboratory, the Lab-it, with the equipment and reagents necessary to execute molecular genetics experiments in loco, in each participating school. The Lab-it could then visit every high school in Algarve to allow biology students to execute the experiments based on hands-on molecular biology techniques like DNA extraction, PCR, electrophoresis, and enzymatic restriction analysis of DNA fragments. The experimental procedures were contextualized in four theoretical scenarios, namely (i) criminal forensic investigation, (ii) prenatal genetic screenings, (iii) molecular detection and diagnostic of patients infected with bacteria carrying multi-resistance to antibiotics, and (iv) molecular identification of bacteria and fungi species from environmental samples. This study presents the results obtained from high school students that participated in Lab-it sessions in the year 2021. The aim was to understand in which measure the Lab-it activities developed by the students contributed to improve (i) their comprehension of molecular genetics principles, (ii) their level of success in the implementation of hands-on protocols by the students, and (iii) in which measure the Lab-it activities contributed to motivating students to continue their academic training and apply to university, in particular, to follow molecular genetics and biochemistry areas.

2. Materials and Methods

2.1. Educational Contextualization of Students

The Lab-it project develops its activities in the Algarve region, in the south of Portugal. In 2021, the project was implemented in 9 of the 18 high schools contacted. Although more schools were interested in collaborating, the COVID-19 pandemic imposed restrictions on some schools, which prevented them from joining the project. A total of 379 students participated in 33 Lab-it sessions. Portugal's education system is organized into three stages, basic, primary, and secondary education and the students that took part in the practical sessions belonged to the secondary education stage, namely the last two years of high school, 11th and 12th grades. All the students had chosen to study biology when entering the secondary education level.

2.2. Methodology of Practical Session

The Lab-it practical sessions were organized with a maximum of 16 students each, with a duration of 3 h. Each activity starts with a presentation of molecular genetics concepts associated with the flow of genetic information within the cells, discussing DNA hereditary properties, three-dimensional structure, composition, and information on genetic hereditary transmission mechanisms. Additionally, students were also introduced to basic concepts associated with methods like DNA extraction, polymerase chain reaction (PCR), electrophoresis, restriction enzyme analysis, and interpretation of results. Students were provided with examples of how to interpret results from a PCR after electrophoresis, contextualizing the polymorphisms evaluated (homozygotes/heterozygotes or presence-absence of bacteria and fungi by ribosomal gene detection) and molecular context of the target gene amplified for each scenario. After the presentation, students learn the

composition of a PCR reaction and correlate it with DNA replication mechanisms. This is followed by learning how to use micropipettes. After that, each group of students assembled the PCR reaction in a tube (a maximum of four groups with four elements each) and applied it to the thermocycler for amplification. All the practical steps were executed by the students after the initial explanation by the Lab-it organizer. After a small break, the principles of electrophoresis were discussed with the students as well as the equipment to be used. Correlation between the interpretation of the results discussed at the beginning and the fundaments of the technique were approached at this point. The students prepared the samples and applied them to an agarose gel for electrophoresis. During the time that electrophoresis was running, we discussed with the students any doubts regarding the practical sessions, new technical approaches used in research, and options for university courses. After electrophoresis, the agarose gel was observed in an Ultraviolet transilluminator, and the results were discussed. Altogether, this process allows students to contextualize experimental approaches and observe the application of these methods to answer the questions in the scenarios chosen (Table 1).

Scenario	Title	Techniques Used	Objectives
1	Who committed the crime?	Pipetting with micropipettes, Centrifugation, PCR, electrophoresis	Amplification of polymorphisms in two genes (<i>PLAT</i> and <i>AMELX</i>) to contribute to genetic profile of a crime suspect
2	Where are the <i>Pseudomonas</i> ? A case of Multi-resistance infection in Algarve	Pipetting with micropipettes, Centrifugation, PCR, electrophoresis, restriction enzyme cut selection	Amplification of 16S ribosomal subunit fragment by PCR from samples of patients to identify <i>Pseudomonas</i> sp. polymorphism with <i>EcoRI</i> restriction enzyme analysis
3	Prenatal screening analysis for hereditary disease and gender determination	Pipetting with micropipettes, Centrifugation, PCR, electrophoresis	Screening for the presence of Alu fragment insertion in CFTR gene and gender determination through amplification of AMELX fragment
4	Microbial ecology: Looking for bacteria and fungi	Pipetting with micropipettes, Centrifugation, PCR, electrophoresis, restriction enzyme cut selection	Amplification of DNA fragments of 16S and 18S (specific for fungal species) subunits to confirm presence and absence within the ecological niche and <i>EcoRI</i> restriction analysis for fragments amplified

Table 1. Description of Lab-it scenarios applied during practical sessions.

The implementation of molecular biology techniques is included as part of four distinct scenarios (Table 1). These scenarios allow students to assimilate the molecular biology principles within a practical application example.

For human biology scenarios, the students amplify two polymorphisms common in the human genome that should favor the understanding of concepts like DNA heredity of polymorphisms and consequences to the homozygotic and heterozygotic state of the human genome analyzed. The first polymorphism that we have used was a common Alu insertion in an intron of the gene that encodes for plasminogen activator, tissue type (*PLAT*) [11]. This was used in scenarios 1 and 3 (Table 1). In the first scenario, it was used in the context of a polymorphism specific to an individual, which could be involved in the identification of suspects at a crime scene [11]. For the third scenario, this polymorphism was used as a theoretical example for a possible *Alu* insertion into an exon of the gene cystic fibrosis

transmembrane conductance regulator (*CFTR*), as described previously [12], but based on the amplification of *Alu* insertion in gene *PLAT*. Mutations in this gene are responsible for the majority of cystic fibrosis cases in the world [13], and this context allowed students to think about the hereditary recessive polymorphisms that can lead to diseases. Additionally, for scenarios 1 and 3 (Table 1), we have used primers specific to the Amelogenin X-linked gene (*AMELX*), which allowed the identification of chromosomes X and Y [13] and could be used in scenario 1 for gender identification of the alleged suspect and scenario 3 for prenatal gender determination.

For scenarios 2 and 4 (Table 1), the students amplified fragments of 16S ribosomal RNA (*16S*) using primers (8F and 1542R) for molecular detection of bacteria as described by Galkiewicz and Kellogg (2008) [14]. The context of scenario 2 theoretical objective was to detect bacteria in patients suspected of being infected by a multi-resistance strain of *Pseudomonas* sp. The students used samples of DNA isolated from oral epithelium cells, which allowed the detection of bacteria. Additionally, we have analyzed several *16S* sequences of several bacteria strains within the fragment amplified by primers (8F and 1542R) [14] and identified an *EcoRI* restriction enzyme recognition site (5'-GAATTC-3') in most oral epithelium common bacteria strains, producing two fragments (840bp and 660bp). It was also possible to observe that *Lactobacillus* sp. and *Pseudomonas* sp. do not have this *EcoRI* recognition site. This fact allowed the students to do a restriction analysis for possible detection of *Pseudomonas* sp. as a theoretical multi-resistance strain and because *Lactobacillus* sp. are easy to detect in the oral epithelium [12], this assay could be easily used to simulate scenario 2 application (Table 1). This allowed students to understand how to differentiate bacterial strains using a restriction enzyme digestion.

The same principle was used for bacterial detection in scenario 4, but in this context, the main objective was to detect bacteria strains from different ecological niches. Because the *EcoRI* restriction enzyme recognition site is absent in *Lactobacillus* sp. and *Pseudomonas* sp. we can promote the discussion with the students for differentiation of these strains in theoretical niches of scenario 4. Additionally, we used the primers nu-SSU-0817-5' and nu-SSU-1536-3' for 18S ribosomal RNA (*18S*) genes fragment amplification as described by Borneman and Hartin (2000) [15]. The amplification of fungal microbial species within the context of scenario 4 allowed students to think about microbial diversity, prokaryote, and eukaryote, in several niches and the potential of molecular biology techniques for detection and characterization of these ecological niches.

2.3. Data Collection and Statistical Analysis

The inquiry that was prepared for the students was composed of 19 questions. The questions were subdivided into four sections, the first one (two questions) asking about age and high school identification, the second (nine questions) related to the execution and opinion about the activities and methods developed by the students, the third (five questions), related with options of the students concerning possible choices of university courses, and the fourth section (two questions) was about the impact of COVID-19 disease on their motivation to understand nucleic acid detection methods and interest in biological sciences. The inquiry was available on an online platform (https://www.survio.com/pt/, accessed on 31 December 2021). The access to the inquiry was done through a site address or quick response code (QR code) and was answered by the students at the end of each practical session. The inquiry was anonymous. From the 19 questions asked, in this paper, we present the results obtained for 15 questions. The answers presented are essentially related to the practical sessions attended by the students, options for university courses, and the impact of the COVID-19 disease.

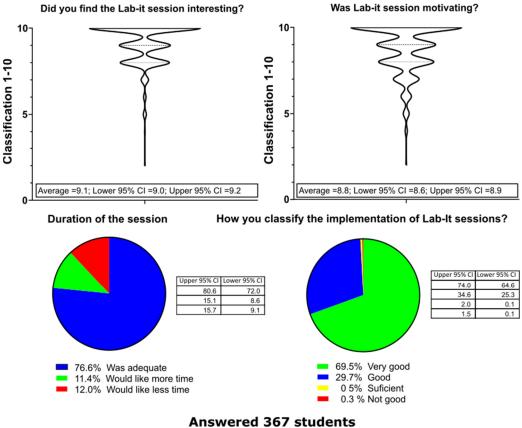
For this study, we applied a univariant analysis for which we have divided the answers into two types, classification from 1–10 and parts of a whole. For the answers requiring a classification ranging from 1 to10, data was presented through a box and violin graphics, allowing distribution of answers analysis, and reported along with the average classification, coefficients of variation (determined by the relative standard deviation) [16], and

respective confidence intervals (CI) at 95%. The parts of a whole analysis were represented by pie charts or bar graphics and results were reported as percentage or number of answers to each category. The confidence interval at 95% was presented along with the graphics presented. The CI at 95% was calculated based on the hybrid Wilson/Brown method [17].

3. Results

In 2021, Lab-it visited a total of nine high schools in the Algarve region, south of Portugal, and a total of 379 students participated in 33 practical sessions. Of the 379 students that took part in those sessions, 367 answered the inquiry. The student's ages ranged from 16 to 20 years, with $42.0\% \ge 18$, 54.8% = 17, and 3.3% = 16 years old, respectively.

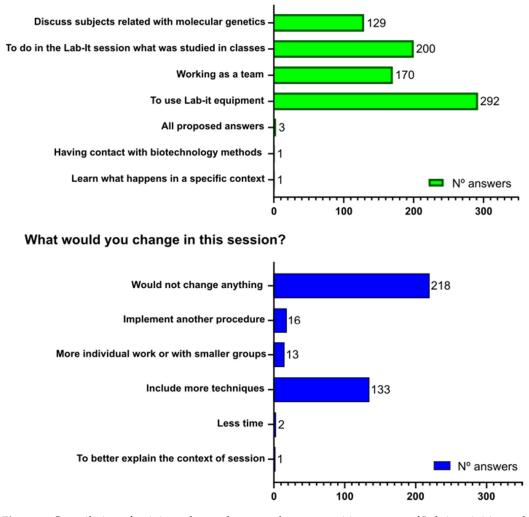
The students were asked to classify from 1–10 how (i) interesting and (ii) motivating were the practical sessions. The students considered the session very interesting with a mean score of X = 9.10 (CV = 12.8%) and motivating X = 8.8 (CV = 16.0%), (Figure 1). When asked about the duration of the practical session (3 h) 76.6% considered that was adequate, 11.4% would like more time and 12.0% preferred less time (Figure 1). Regarding Lab-it practical implementation 69.6% and 29.8% considered "very good" and "good", respectively (Figure 1).



Answered 507 students

Figure 1. Opinion of students regarding the application of practical activities of Lab-it.

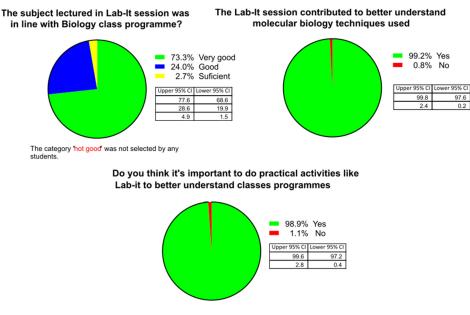
Regarding the most positive aspects of the practical sessions, the preferred option (292 answers) was the opportunity to have a hands-on experience with Lab-it equipment, followed by the application in experiments that related to themes studied in biology classes (200 answers) (Figure 2). Students could answer more than one option or write their option if they wanted to add any comments. In addition, it was asked if students would change anything about the practical session. Results showed that 218 students did not change anything and 133 would like to apply more techniques (Figure 2).



Most positive aspects of Lab-It session?

Figure 2. Compilation of opinions about what were the most positive aspects of Lab-it activities and what would students change in the sessions.

When inquiring the students if what was applied in the scenarios and techniques used in the practical session were in line with the biology discipline program, 73.3% considered "very good" and 24,0% "good" (Figure 3). Only 2.7% answered that it was "sufficient ", and none of them choose "not good (Figure 3). In addition, 99.2% considered that the practical sessions contributed to a better understanding of the techniques and methods of molecular biology applied (Figure 3), and 98.9% of the students believed it would be important to do more practical activities like Lab-it sessions to better understand class subjects given in high schools (Figure 3).



Answered 367 students

Figure 3. Students' opinions about the correlation between the practical application of molecular biology methods and theoretical subject approaches in biology classes.

Another question concerned the student's options of continuing to university and choice of courses. 89.9% of the students indicated they were planning to apply to the university (Figure 4). Of those, 40.6% would apply to biological sciences courses and 31.3% were considering going to a course in this area (Figure 4). About 28.1% did not plan to apply to a course in this area (Figure 4). Regarding the students that answered yes or maybe to the question of continuing into biological sciences, it was asked to classify from 1-to 10 how the Lab-it session contributed to their choice and on average answered X = 7.8 (CV = 24.8%), (Figure 4).

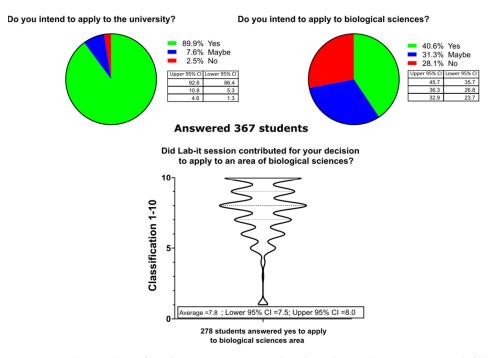


Figure 4. Understanding if students want to proceed with studies in the university and if the Lab-it session could have contributed to students' choice towards going into a biological sciences area.

Attending that the Lab-it practical sessions were done following severe COVID-19 restrictions, it was asked to students to classify from 1–10 if the SARS-CoV-2 pandemic changed the way that they studied, on average answered X = 7.7 (CV = 29.8%), (Figure 5). It was also asked if the COVID-19 pandemic increased their interest in the biomedical sciences area and students classified as X = 6.0 (CV = 47.7%), (Figure 5).

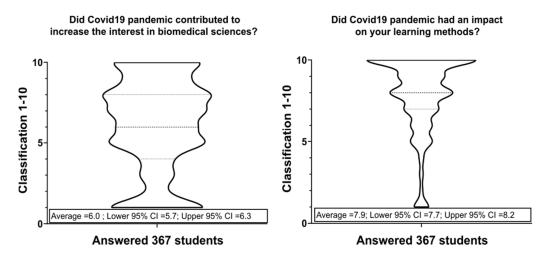


Figure 5. Impact of COVID-19 pandemic on students' interest in biomedical sciences and habits of study.

4. Discussion

The Lab-it project aimed to contribute to improving the teaching of molecular biology and biochemistry concepts to high school students in the region of Algarve, south of Portugal. The objective of the present study was to understand if the technical approaches done in Lab-it sessions within specific scenarios contributed in a significant way to increase the understanding of the molecular concepts discussed in the high school biology program, from the point of view of the students. For that, an inquiry was prepared with questions focused on specific items that could allow us to understand if the Lab-it sessions had contributed to fulfilling the proposed objectives.

The first two questions were done with the objective of understanding if the activity was appealing and could bring attention to molecular genetics concepts. Results suggested an increased interest and motivation to learn concepts developed in the practical activity (Figure 1), contributing to improve motivation and interest, as described before when students attended practical classes [18–20]. In addition, it was asked to students about the application and duration of Lab-it sessions and student opinions were highly positive as represented in Figure 1. The most significant results of this inquiry are related to the fact that students believe that the Lab-it practical session to be an extremely positive contribution to understanding the molecular biology and would like to have similar activities for other subjects (Figure 3), in agreement with what was previously believed about the importance of practical work in molecular genetics and sciences in general [9,10,21]. These results are in agreement with previously published studies which showed that complementing the teaching of theoretical concepts with practical methods can be a significant contribution to students' comprehension of molecular genetics and biochemistry concepts [18,19,22,23]. In line with this, the opinion of students confirmed that practical activities were a positive aspect to understand molecular genetics. When asked about the most positive aspects of the practical session, students indicated to be the possibility of hands-on use of Lab-it equipment and be able to apply concepts thought in biological theoretical classes to answer specific questions (Figure 2). In addition, when asked which aspects they would change in the Lab-it activity, the majority answered that they would like to include the use of more techniques (Figure 2) [18,19] thus confirming their interest in hands-on experiments.

The analysis of the possibility to apply to university courses showed us that the vast majority of students were planning to continue their studies into the university (89.9%) and of this, 40.6% were planning to apply to a course in biological sciences (Figure 4). These results showed that the student population which attends science and technology courses at the upper secondary education level is highly motivated to proceed to study at the university. In addition, students that would like to go to biological sciences course or were thinking about it were asked to classify from 1–10 what was the impact of Lab-it activity in their decision. The students on average classified as 7.8 with a CV of 24.8%, suggesting that most students classified >5 (Figure 4), indicating that Lab-it activities had a positive impact in motivating them to continue their studies in this area.

The Lab-it practical sessions were done during the COVID-19 pandemic after restrictions to presential classes were lifted. Attending to that, we asked students to classify from 1–10 how this context affected studying habits, and on average students classified as X = 7.9(Figure 5), suggesting a significant impact of the SARS-CoV-2 pandemic in schools. These results are in agreement with what was published before [24,25]. Students were also asked to classify if this pandemic context led to an increase in their interest in biomedical sciences and students on average classified as 6.0 with a CV of 47.7%, suggesting that their opinion regarding this question was dispersed and not consensual (Figure 5).

This study describes, from the students' point of view, the impact of Lab-it sessions on comprehension of molecular genetics concepts and clearly shows how these activities can motivate students and contribute to a better understanding of molecular genetics and biochemistry concepts. However, this study had some limitations, in particular by lacking the possibility of a follow-up of the students after participation in Lab-it to evaluate its impact on the continuation of their training. We propose in the near future to follow some of the students that participated in Lab-it sessions and evaluate how their comprehension of basic molecular genetics concepts contributes to their success in the final exams and facilitates their integration into the university courses in this area and compare it with students that did not benefit from similar activities. In addition, it would be important to inquire from the high school teachers that collaborated with Lab-it about the differences found in student's progress and motivation before and after the participation in the Lab-it project. It will be necessary to develop a practical method to quantify this impact. This study would also benefit from a compilation of results of inquiries to the teachers that collaborated with Lab-it. This is planned for future studies.

5. Conclusions

The main objective of this study was to understand, from the student's point of view, the importance of Lab-it practical sessions implemented for their scientific training, and to obtain evidence of the positive points and changes that could be done to improve it. We can conclude with these results that students have a highly positive impression of the execution of practical activities, with a focus on learning to use Lab-it equipment to better understand the technology involved and apply this know-how to solve specific questions through hands-on experiences. They even suggested the inclusion of additional techniques. It was important to understand also if the model of those sessions was contributing to increasing the understanding of molecular genetics and biochemistry concepts approached in biology classes and we can conclude that students considered it highly relevant and important to better understand the subjects taught in their biology program. Results also showed that the students were highly motivated to apply to a university and a considerable percentage found that participating in Lab-it practical sessions help them to decide to choose an area of biological sciences. This study represents an initial step to a follow-up study about the impact of Lab-it activities in high schools and how they could influence students' learning skills and choices for progressing into university studies.

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