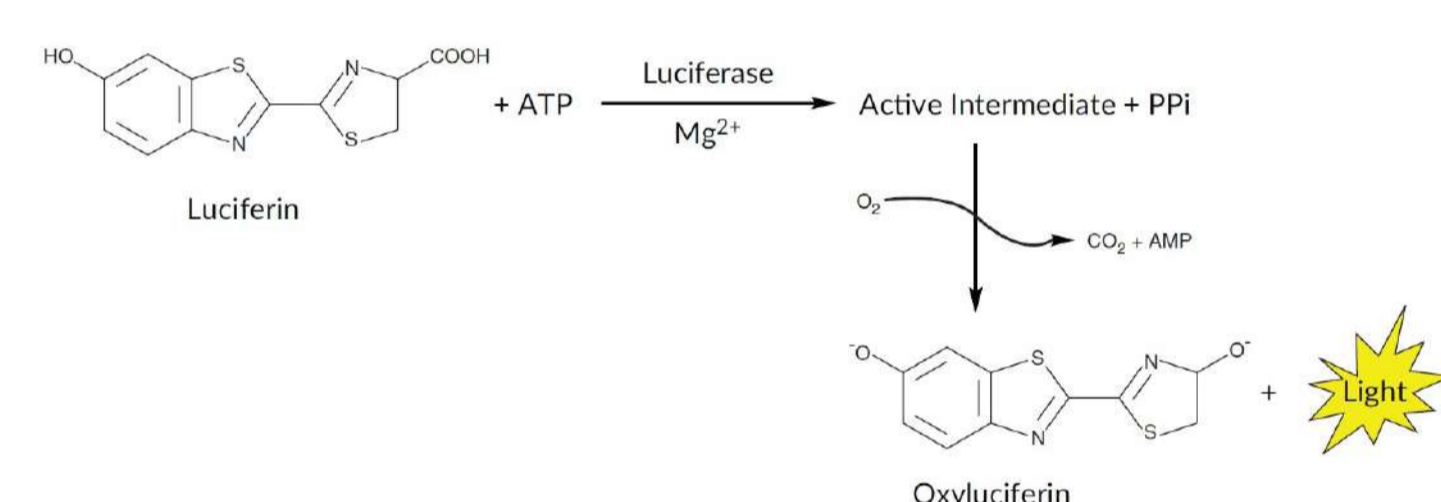


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Introduction

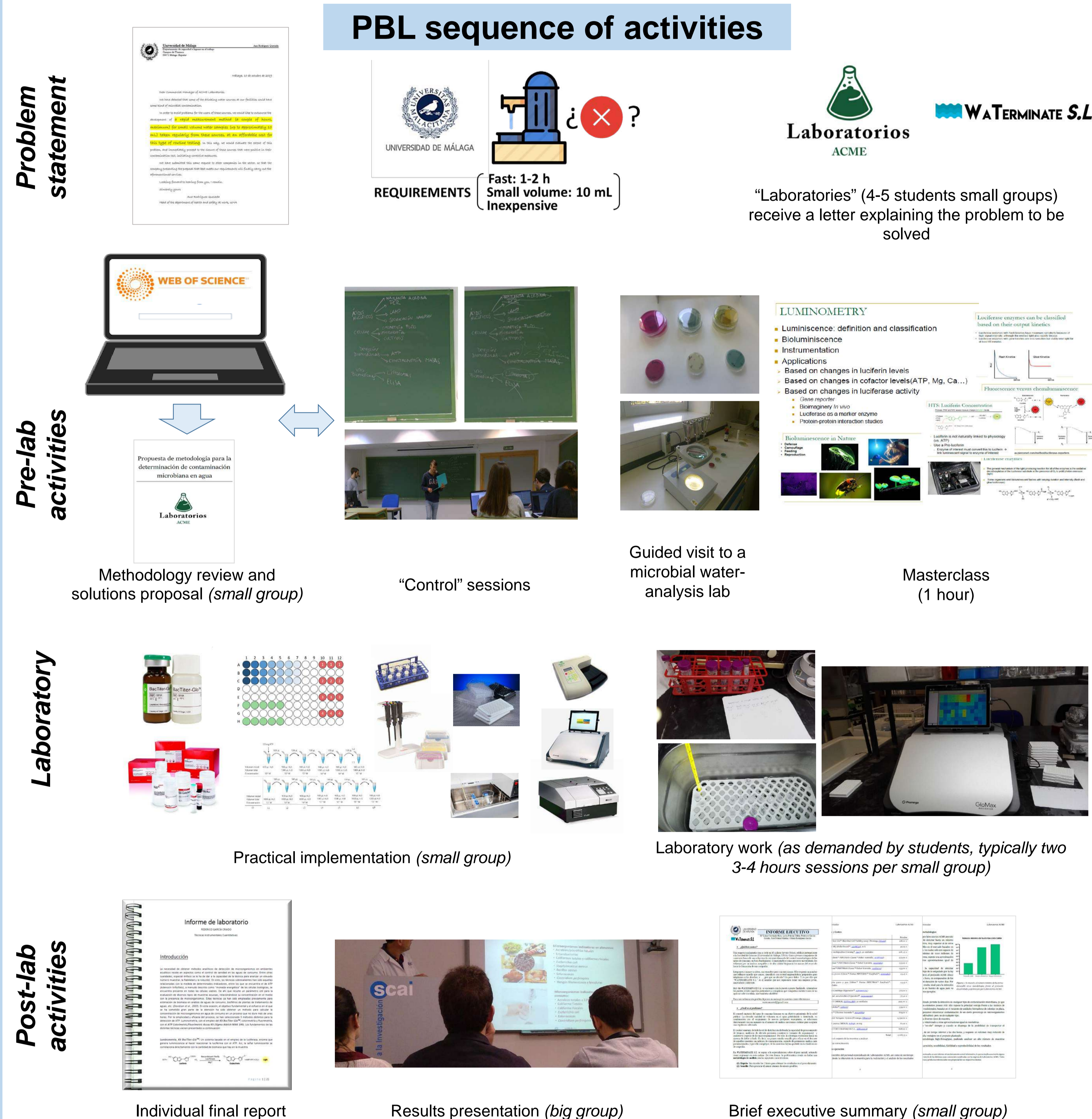
There is a lack in the formation of our chemistry and biochemistry undergraduate students, caused by the shortage of practical laboratory experiences that illustrate the applications of luminometry, a technique that is increasingly used in of experimental and health sciences laboratories. Bioluminescence is a phenomenon that our students easily recognize in some animals, such as fireflies, which emit light for the recognition and attraction of their partner in mating. This light is the result of a chemical reaction catalyzed by the enzyme luciferase (EC 1.13.12.7), which requires the presence of the luciferin substrate and ATP as a cofactor.



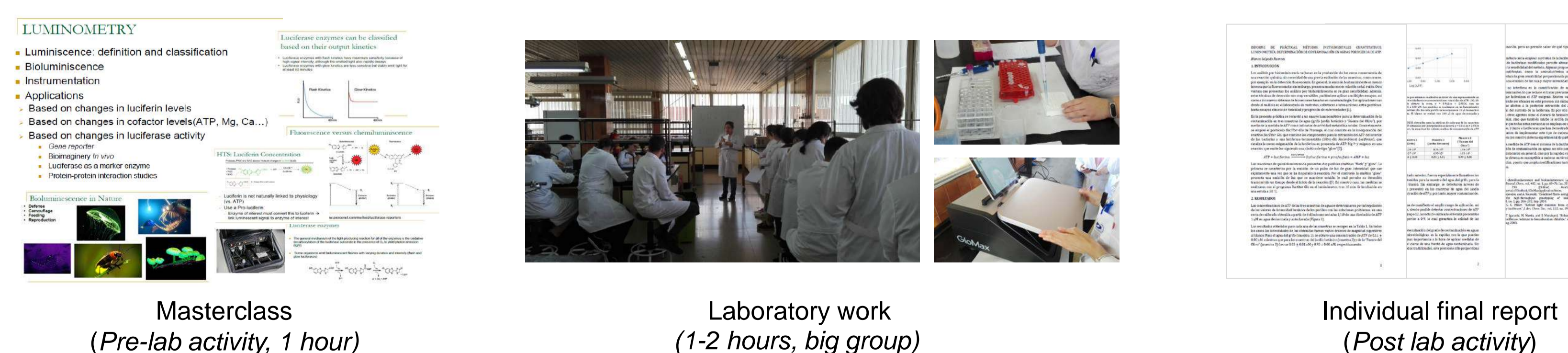
One laboratory experiment, two different patterns

The correlation of ATP concentration and bacteria content is the basis for the development of luciferase-based rapid methods to detect microbial contamination in drinking or stored water, skipping the long delays required by the traditional microbiological methods¹. Throughout the two last academic years, we have implemented at the University of Malaga (Spain) a new laboratory experiment focused to illustrate the use of bioluminescence in analytical chemistry to undergraduate chemistry and biochemistry students. Based on the luminometric measurement of ATP for the detection of bacterial contamination in water, it has been implemented in two different formats, a short protocol and a complete PBL experience. In this PBL students are more actively involved in their learning process, with the teacher playing a mere "facilitator" role in this process.

¹Hammes F, Goldschmidt F, Vital M, Wang Y, Egli T. Measurement and interpretation of microbial adenosine tri-phosphate (ATP) in aquatic environments. *Water Res.* 2010; 44(13):3915-23.



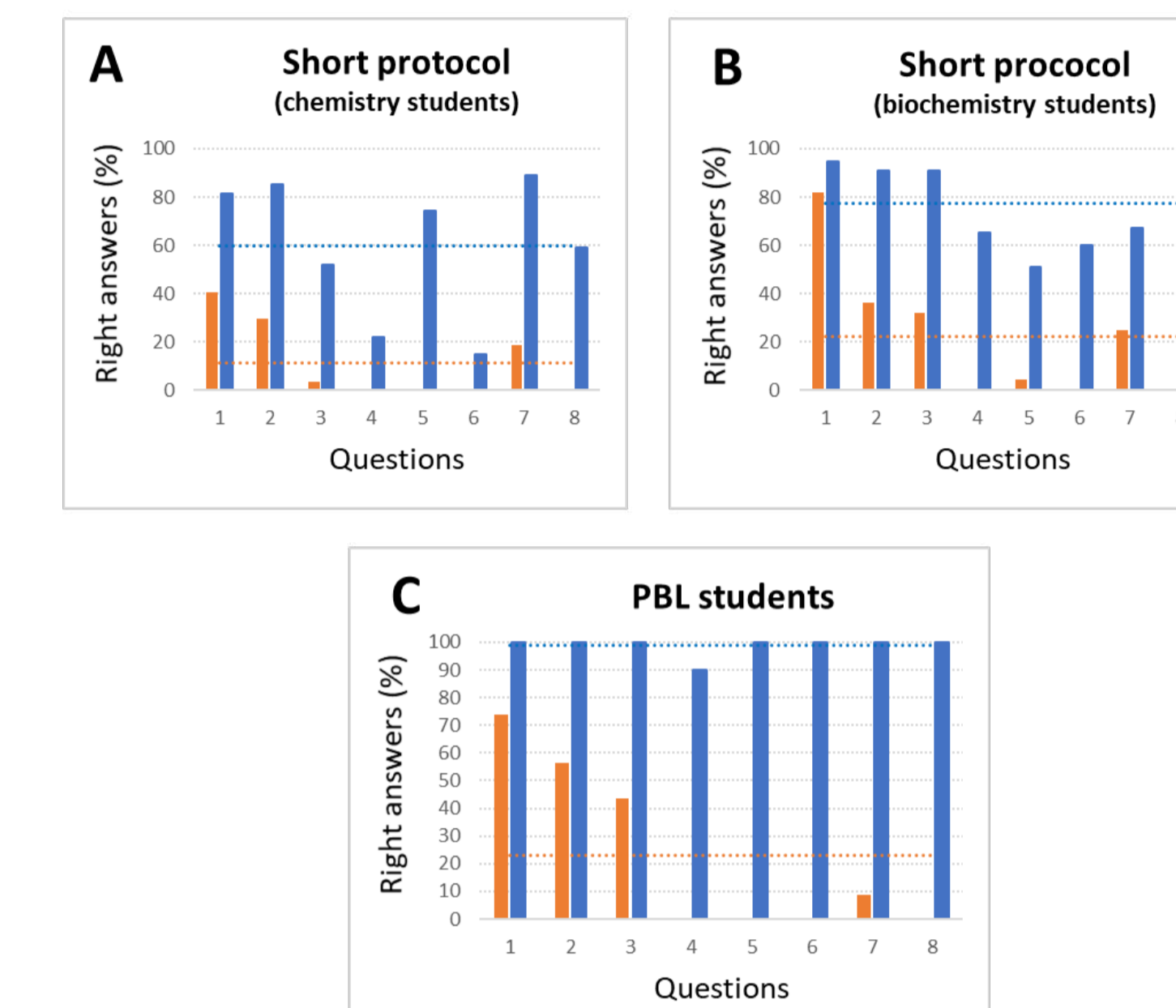
Short laboratory experiment



Students' results: pre post-test comparison

A first estimation of the students' progress was made by means of an assessment test composed by the following eight short open-questions:

1. What do fluorescence and luminescence have in common?
2. How are they different?
3. What do a fluorimeter and a luminometer have in common?
4. How are they different?
5. Could you indicate the substrates, effectors, products of the luciferase-catalysed reaction?
6. How are luciferases classified according to their kinetics?
7. What applications of luminometry do you remember?
8. How would you measure the presence of bacterial contamination in a sample?



Percentages of students who answered correctly the pretest (orange bars) and the posttest (blue bars) questions are shown. Pretest full average is shown as an orange dotted line and posttest full average as a dotted blue line

PBL students' feedback

- The learning methodology (PBL) used in this activity is innovative with respect to that used in other subjects of the degree
- I have been able to learn about the fundamentals and instrumentation of luminometry
- I have known some applications of bioluminescence measurement
- This PBL has allowed me to understand how a laboratory protocol is developed and to establish a work plan, better than with other practices I have carried out previously
- I have been able to determine and compare the detection limits of several analytical techniques
- This PBL has allowed me to approach situations that are similar to those that may arise in my professional future and seek solutions through bibliographical research
- I liked being able to plan the tasks to be carried out in the laboratory, and organize the work to be done
- This PBL has required more work and preparation than other practices carried out throughout my undergraduate studies
- This activity has been worthwhile because I have learned more than with other laboratory practices

Conclusion

The detection of microbial contamination by measuring ATP concentration with luciferase is a useful tool to teach the use of bioluminescence to science undergraduate students. Either as a short protocol carried out in a single laboratory session, or as a long PBL experience, the students' achievement of the learning goals was very satisfactory. Students who followed the PBL approach could, in addition, adopt a series of additional transversal skills that could be useful in their future professional careers. Between these two extreme examples of the protocol, in terms of students' dedication, a whole range of intermediate options could be used by educators to suit their course programming and fulfill the learning objectives to be achieved by their students by means of this laboratory experiment

Acknowledgements: Work supported by University of Malaga (Educational Innovation Project PIE19-086 & PIE19-057) and Spanish government (R&D Project EDU2017-82197-P)