chatter plant

SAFETY

PLANT SYNTHETIC BIOLOGY

Universidad Politécnica de Valencia
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iGEM UPV VALENCIA

UNIVERSITAT POLITÈCNICA DE VALÈNCIA

1. INTRODUCTION

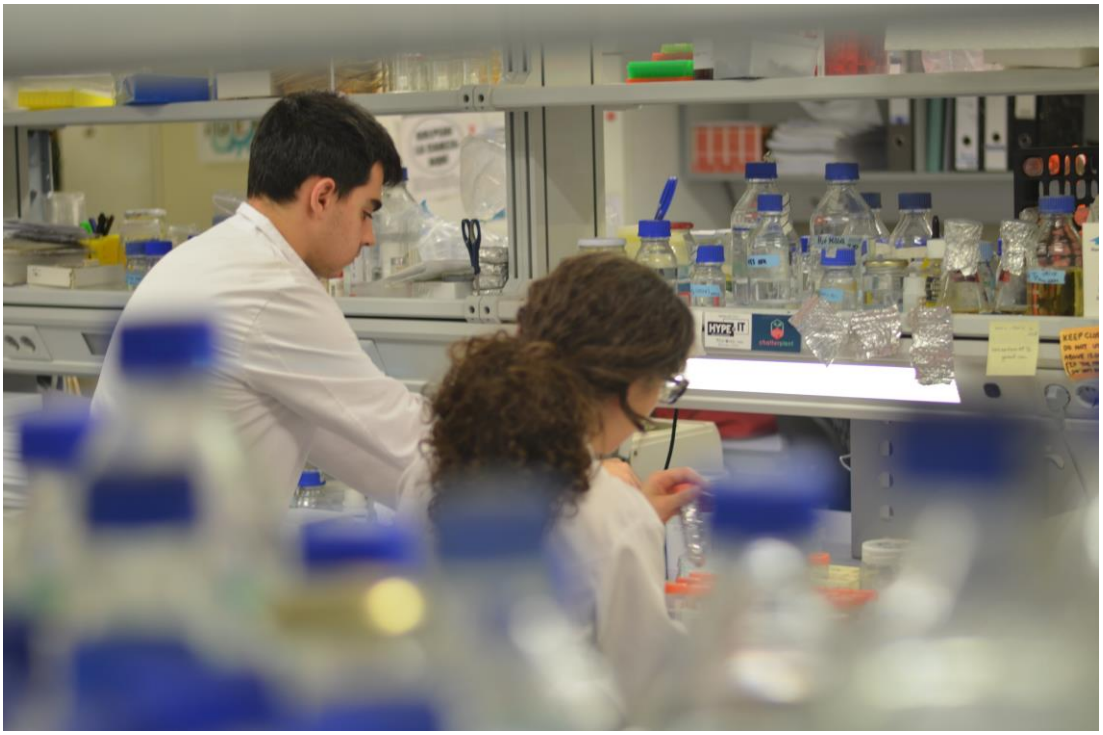
Synthetic biology is a consequence of both innovative advances in biological technologies and engineering disciplines. SynBio progress makes a whole set of potential applications become possible. Nonetheless, biological engineering requires **precautionary levels** so safety and ethical aspects of Synthetic Biology must be ensured.

Firstly, SynBio deals with biosafety to prevent accidental exposure to pathogens and unintentional liberation of harmful biological material. Secondly, security prevents any misuse through that can be a threat for society. In this area, awareness needs to be further enhanced through better communication and cooperation. Finally, Synthetic Biology also deals with the ethical questions related to the design and creation of new forms of life. In this context, SynBio security has been further considered in each discipline of our project to exhibit a **safe and responsible work** in iGEM.

2. SAFETY IN PROCEDURES

Before starting with the **laboratory work**, our instructor, Dr. Diego Orzáez informed us about how to work in the lab and what is usually considered as a good practice.

Personal safety precautions are mandatory in wet lab work. Wearing lab coast, gloves and disposing each biological waste in its corresponding container are the basis of our daily work.



Furthermore, bacterial cultures are highly sensible to contaminations so working on sterile conditions is required. A special area for running electrophoresis gels is designated in order to handle hazardous chemicals like Ethidium Bromide. Other dangerous chemicals, such as beta-mercaptoethanol or dimethyl sulfoxide were handled under the fume extraction system.

3. LAB SAFETY

We have been using genetically **modified organisms** in our project so an exclusive focus should be set on laboratory's work safety. Different bacterial strains were used depending on their characteristics and the purpose of their culture. Escherichia coli DH5 alpha and Top 10 were used in cloning process. Moreover, Agrobacterium tumefaciens GV3101 strain was utilized for Nicotiana benthamiana transient expression. In both cases, GoldenBraid plasmids were used in the assembly process which are **not harmful** for humans nor other living organisms.

The transient transgene expression in Nicotiana benthamiana was carried out in an equipped laboratory. It has to be clear that transient expression is not the same as the stable one. The plants that we have used do not have flowers so there is no problem of spreading anything harmful to the environment through pollen.

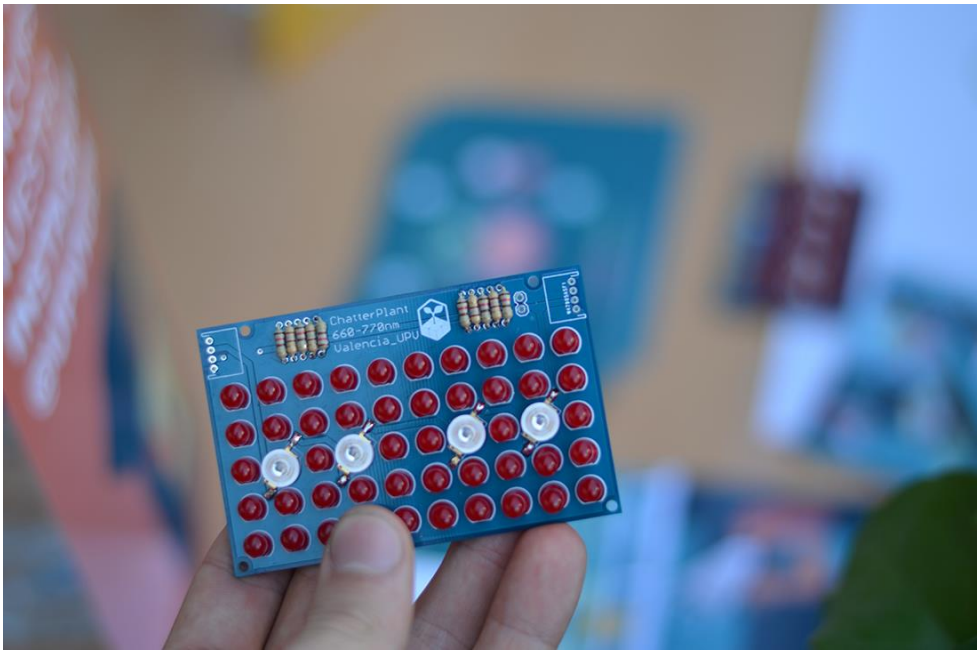
After finishing the analysis and web-lab procedures of the plant, we discard it as a biological waste following autoclave protocol in order to maintain security conditions.

As a proof of concept, we have been working with plant viral vectors. They are not viral particles (observe difference between particle and vector) so they cannot spread from plant to plant. When we introduce viral vectors in Agrobacterium tumefaciens, it still doesn't replicate inside the bacteria. When we finally infect the plant with Agrobacterium (with the viral vector inside), the viral vector replicates itself and the viral particles are synthetized. At that point, it is possible the transmission from plant to plant. To avoid that, the infected plants are stored in a separate room in our equipped greenhouse. As these plants don't have flowers, we can't obtain seeds from them and **no transmission** through the environment is possible. Finally, we always autoclave these plants and viral particles are destroyed due to high temperatures.

Finally, it is important to highlight that the virus used do not express the coat protein, so they are **not able to spread** from plant to plant. This is relevant for avoiding contamination of the environment with the modified plant.

4. HARDWARE SAFETY

The **Chatterbox** is a high humidity ambient with liquid water flowing, this represents a challenge in terms of security. Our goal was to keep the system as **watertight** as possible, to achieve this we used different approaches. The led illumination was chosen with an IP65 protection level, meaning that it can withstand direct water flow. In the case of the electrical connections heat shrinking tubes were used. The most sensitive equipment was the optogenetic lamps which were protected with epoxy resin and a box.



5. SOFTWARE SAFETY

As a multidisciplinary team, we have been working in different disciplines and we are proud to present an **open-access online platform** where laboratories will be able to publish individual results supported by a modelling software tool integrated. Our aim is to unify Plant SynBio research work in iGEM community and scientist in general. Naturally, we cannot control the proper use of published experiments but our documentation enables readers to better understand how our software works and how can they **use it properly**. Furthermore, we offer the possibility to contact us personally, so that we try to reduce any form of improper use of PlantLabCo.