Snow White's Apple for the Oak Processionary Caterpillar

Severe itching, rashes, respiratory issues like asthma and eye complaints… All symptoms that affect over 100.000 people in the Netherlands alone as a result of the Oak Processionary Caterpillar (OPC) who returns with vengeance yearly. In this blog we introduce a local issue that we aim at solving during the annual international iGEM competition in synthetic biology.

It is no secret that the OPC has become a major threat over the years, harming fauna around them as well as the flora they reside in. Apart from stripping trees bare to their bark, they form a serious health hazard for humans as well as animals. The allergenic protein *thaumetopoein* on the bristles triggers inflammatory responses of the skin and eyes as well as respiratory problems like asthma attacks. In serious cases, the bristles even have to be removed surgically from the eyes.

In the last 3 decades, climate change seems to be the OPC's best friend. It enabled the invasive species to cross its native borders in Central and Southern Europe, and spread towards all European countries and even parts of the Middle East. It's quite the journey, isn't it?!

Up until now, no insecticide has sealed the deal. Many of the current control methods are very costly, inefficient and non-specific. One of the control measures used at the moment is the biological control; bacterial insecticides (Bt) are sprayed on leaves releasing a toxin, which is a non-specific approach that also causes harm to other caterpillars and butterflies. Another method is the so-called nest removal; where the caterpillars are vacuumed up and burned. This is very dangerous for exterminators themselves and simultaneously also an expensive technique.

With a team of 13 students from the University of Maastricht we aim at tackling the problem by genetically engineering a bacterial pesticide that is specific for the OPC and on top of that, environmentally friendly. This biological pesticide would target specific and essential gene sequences in the caterpillars to reduce their growing population and avoid harming other species. For this we plan to take advantage of the cellular mechanism of RNA interference. It is a form of gene silencing, where the selection and silencing of a critical target gene would lead to the OPC's death.

To realize this project funding must be done because "geen geld, geen Zwitsers". Apart from a crowdfunding campaign that was set up we are proud to have won a $5500 grant from Revive & Restore, an international conservation organization. Nevertheless, collecting money for research in times of pandemic is harder than you might think, but that is for another episode of our blog.

So far yet so close

During the times of a global pandemic, friendships and teamwork are built from behind the computer screens, but with a mutual goal in mind we are set to get the best out of this challenging situation. In this blog it is time for you to get to know the unique iGEM team from Maastricht.

Although the whole iGEM experience is completely new for me and my team mates, we certainly are used to cooperating due to the concept of research based learning (RBL) at our university. Biannually, we are immersed into a 4 week project period in which we conduct research in a team of about 4-12 people. As aspiring scientists partaking in the iGEM competition we are now reaping the benefits of the freedom that our study programme offers us. Something that is also quite intriguing about our university is its global mindset. With more than fifty percent of students coming from abroad, Maastricht University is the most international university in The Netherlands.

With 10 different nationalities, our team has a variety of backgrounds and viewpoints that aid in finding the most efficient solution. This way, our teammate Lars, who is expert in systems biology, takes care of modelling which is of major importance in a period in which wet-lab work is limited. If he requires help with any equations, Juliette will give him a hand with her background in physics. This way you see that the members complement each other in order to get the best result possible.

Apart from having 12 other members who we can rely on at any time, we are lucky birds to have two amazing supervisors who lie one step ahead of us with one year of iGEM experience. This way, we were lucky enough to be able to realize some of the ideas the previous team ended with. For example, we have started a YouTube channel called "Geneducation" (https://www.youtube.com/channel/UCpZQvRTiQNvqz9XcqDGK9pA) where we educate the general public about various topics in genetics and synthetic biology. In addition to that, we upload fun at-home-lab tutorials where you can learn how to make your own yogurt or even grow bacteria yourself. Also the editing of these videos has strengthened our bond:

"We haven't really been able to meet in person but because of the videos and seeing the other person on the screen and editing all their laughs out, it feels like we have met already."
- Juliette Passariello-Jansen.

Next to camera-work and editing, we have worked on a proceedings journal for all iGEM teams. We are so happy that it turns out to be an enormous success with as many as 34 different participating teams, covering more than 40 articles. Via this initiative, even though the Giant Jamboree will not take place in Boston this year, each team still has a nice souvenir to hopefully look back at a wonderful period of the start of their research career. Ideas and experience are not the only asset of our supervisors, but also their endless list of contacts comes in very handy. It enabled us to interview experts in fields like peer review and scientific communication to improve our own strategies.

Together we are a very diverse bunch, with everyone having something to offer to the team. Overall, it has already been a great experience working together and sharing our crazy ideas and many late-night laughs over our Zoom meeting. It seems that soon we can share all of this in real-life, under the pleasure of a drink or two.

A good night's sleep ensured!

Working in the lab starts with a tiring trip to Brightlands Chemelot Campus, which is part of the bigger Chemelot Industrial Park and Campus that also hosts research facilities for companies such as SABIC and DSM. This campus is located about 23 km from our university. First, we take the train, then the bus and then we can finally walk to the laboratories. One big plus is that we are welcomed by unlimited access to hot beverages like coffee and tea that definitely lifts up our spirits during the long days spent in the labs.

Overall, we are very glad to work in such bright and modern laboratories, where we can find all the equipment we need for our research work for this iGEM project. Almost everybody in the team is very familiar with these facilities because most of our practical courses also take place there. As in this project we are genetically modifying bacteria, a big part of our work takes place in a microbiological laboratory with biological safety level I (ML-I). Here, additional safety measurements apply for disposing of waste as well as wearing lab coats designated only to this area among others to prevent the possibility of these organisms getting out in the environment. Of course, in these trying times additional COVID-19 safety measures also apply.

Although for some iGEM teams it was naturally that they had access to the lab, we are very lucky to have started in the beginning of September. Our wet lab work started with making sure that the specific sequences that we found through our research are really present in the OPC. This was done by extracting the DNA from this species and running a PCR (Polymerase Chain Reaction) with primers specific for the target genes designed by us. This reaction amplifies a specific region of the gene, thus making the detection process easier. Afterwards, agarose gels for these samples were run to image the results and to be completely sure that these were the target genes, we sent these amplified regions for sequencing and are still waiting for the results.

At this time, we are focusing on designing and assembling all the parts that are needed for the E. coli to produce the small interfering RNAs (siRNA) specific for the target genes. This includes, constructing an insert with the short hairpin RNA (shRNA) sequences that will produce the specific siRNAs and a terminator for our chosen promoter. Then, this construct needs to be inserted in a plasmid through an assembly. Afterwards, this plasmid will be transformed back into our E. Coli strain.. Later these bacteria will be grown, and we want to test how long they would survive on the oak tree leaves. Our future plans (possibly outside this project) include, hatching the OPC's and performing feeding experiments

Although this year, due to the pandemic, we have not been able to spend as much time in the lab as we would have liked, we still believe that we have made quite the progress towards our goal. As well as, spending time with the other team members in the lab will always be a fond memory. Of course, at the end of the day the long travel time pays off!