# Phage Display, Weekly Notes

**Week 1 (25/6-1/7)**
This week we had a meeting with Ylva Ivarsson, who works with phage display at Uppsala University. We asked her for advice on our project and general thoughts about doing phage display on an whole organism. We started to work on a protocol based on whole organism panning and research for ideas on how to not lose the organism when doing necessary washing steps in the protocol. We tried out different kinds of techniques like different sized filters, Büchner funnel , centrifugation, and lower the temperature. But the idea that seemed most realistic was to use eppendorf tubes filters, where we could wash our worms without losing them.

**Week 2 (2/7-8/7)**
We started to design an eppendorf insert in Fusion 360, and printed our prototype on a 3D printer. A lot of prototypes were made and they worked pretty good. Our big problem was to find a good way to add a very small filter to the insert. Therefore, we left our own design for now and start searching for eppendorf inserts to buy with built-in filters.
We ordered a phage display kit, with 12 mer. Meanwhile waiting for it to arrive we researched about ELISA that we will use as a “quality check”. This week we also went through the manual for Phage display carefully.

**Week 3 (9/7-15/7)**
This week our ordered phage display kit arrived. To get more professional help from some in the phage display field we booked a meeting with Gustav Sundell that is a Phd at Uppsala University.
The buffers and medias for experiments were prepared.

**Week 4 (16/7-22/7)**
Ordered eppendorf tube filters arrived. Experiments were performed to make sure that the filter tubes worked as we wanted in our modified phage display protocol.
E.Coli strain, ER2738, that came with the kit is optimized for the phage amplifying step. To not run out of these bacteria, glycerol stocks as well as solid cultures were prepared and stored at -80$^{o}$C and 4$^{o}$C respec.

**Week 5 (23/7-29/7)**
In order to not run out of phages the initial library was amplified.
For optimal phage propagation it is important that healthy bacteria (mid log phase) is infected as it will have the highest chances of producing our phage vectors. Every experiment begins with culturing bacteria to this phase. In order to better approximate bacterias growth rate a growth curve for ER2738 starting from solid culture was studied.
We started a phage display experiment.
Anti-m13 antibodies with conjugated HRP arrived. Their functionality was tested for future use in elisa.

**Week 6 (30/7-5/8)**

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| We started two new parallel phage display experiments. This was done to make sure we can compare the results from the three experiments. |
| **Week 7 (6/8-12/7)**<br>We continued working on the three phage display experiments. |
| **Week 8 (30/7-5/8)**<br>We concluded the three parallel phage display experiments and amplified separate plaques for analysis, followed by phage DNA extraction. We performed a test round of ELISA on our selected phages and used nanodrop and gel electrophoresis to quantify the DNA concentration and purity of the samples. |