

Archiving Phage Lysates FAQs

1. What is the purpose of archiving phage lysates?

One of the tenants that is agreed upon by every SEA-PHAGES member when you join the program is that all phages found in the program will be archived at the University of Pittsburgh. We want to maintain a phage lysate for every phage found.

2. What phage lysates should be archived?

The simple answer is that all phage lysates should be archived. However, some of you find that titers are too low to archive. It is your decision. However, whatever you decide know that an archive sample should be provided for every and any lysate listed on PhagesDB. Phage pages that have no lysate archived will be deleted.

3. When are phage lysates due?

Phage lysates are due at the end of the year in which they are found. Currently the “due date” is December 15 each year. If you miss or plan to miss the due date, contact debbie (djs@pitt.edu) to work out a plan.

4. Who is best qualified to prepare lysates?

Make archiving a student responsibility. Our experience tells us that folks who think they have to do this for the students provide late samples, or no samples at all. It is not acceptable to ‘skip’ archiving your phage lysates.

5. What phage lysate should be archived?

The best lysate to archive is the one that you prepare your DNA from. Have students make the archive samples (both the one that you keep at your institution and the 2 you send to U of Pittsburgh) at the same time. The titer of the lysate should be $> 5 \times 10^9$.

6. There are a few HTLs that we do not have a large enough volume of to make 5 ml of the DMSO:lysate mixture. What should we do?

You have some choices. The best answer is to make more and repeat. The next best answer is to send 1 tube (noting that on the accompanying archive sheet). DO NOT dilute the lysate to meet the volume requirements.

7. How best to label archive tubes?

Label the tubes with the phage name using a permanent MARKER. The tubes go in a tight rack into the freezer, tape or labels can be problematic. Do not send unlabeled tubes.

8. Where are the instructions and archiving video found?

Archive protocol is found [here](#). The link to the video is contained in the protocol. The video currently includes the use of beads. Do not add beads.

9. How do I obtain an archive list?

You can download an archive list from your institution page if you are logged in as a faculty member at seaphages.org. This is a critical component of your archive procedure. Your archived lysates should match the phages entered at PhagesDB.

10. Does it matter what order the tubes are placed in the archive box?

YES! Place the tubes by pairs in the order that they are listed on the archive sheet. When that box is received at Pitt, the archive list and tubes are matched and separated into 2 sets (so that they go into 2 different freezers). Note: Archive lists are generated by calendar year. If you are running the course each semester (Basically twice a year, please contact debbie directly to work out a plan.)

11. When will we receive our archive tubes each year?

We are currently sending a Google Form to capture how many phages you expect to archive. Please complete it promptly! We aim to ship archive tubes the first week of November each year. You may also request them early and we will do the best we can to get them to you.

12. What happens if we miss the December 15 deadline?

If you miss the deadline by a day or two, no worries. Unless that day or two encroaches the holiday recess at the University of Pittsburgh. Use your common sense and send an email with questions or notification that you will ship after the new year. If you are sending them at any other time, please send debbie an email to explain your plan.

13. Who enters the lysate data into phagesDB.org?

You do! Do not just enter the data on the archive sheet and think it is happening at Pitt. Please modify your phage pages accordingly.

14. What happens if a phage page needs to be deleted?

Phage pages can only be deleted at the University of Pittsburgh. Any phages that are to be deleted can be marked on the archive sheet that you send with your archived lysates. Alternatively, you can send an email to debbie and makes those requests.

15. What happens if we are not able to send a lysate?

If no lysate is able to be made/shipped, then the phage page should be deleted. Send debbie an email with a request to delete. The exception is if the phage is sequenced. In that specific case, please notify us that you can't send it and we will mark the archive status appropriately.

16. When we place the tubes in the freezer box, do you want the duplicate copies next to each other or (i.e. 1,1, 2,2, 3,3) or separated into two sets (i.e. 1, 2, 3, 4... and then 1, 2, 3, 4...)?

The first example is best.

17. If we don't have enough lysate to send yet, do you want us send what we have now and the rest after we get more HTL for that phage? Or do you want us to wait until we have all samples?

The best answer is to wait until you have all samples UNLESS that proves to be problematic (as in you can't get a high titer) OR your timeline to do this work is greater than 1-2 weeks. If you are sending an incomplete set of samples, please indicate on the archive list A) what is missing and why, and B) when we can expect it.

18. What temperature should we ship at?

Ship with cold packs at ~-2-8°C. Do not ship with wet ice.