October 6, 1952

Dear Norton:

This letter is in reply to yours of the 2d [if we continue to correspond with cross-firing, such asformal introduction may be a good idea].

I am glad to hear that your lab. organization has progressed to the point that you want cultures. They will be sent on (if still viable) very shortly. Actually, not so many cultures have been lost, but only because the beads extracted from cracked tubes that were sopping wet above the cotton still grew out. Your comments on the directions of your work at Rockefeller seem very sound. It is important to remember that a really adequate proof that FA - phage has yet to be made. This may be difficult, as it is like asking whether the particle that transduces might, underwwhatever other circumstances are critical, instead have killed. The UV activation may still be a useful lead.

I'm not sure that I did understand the sentence (p. 2 P.4) that you asterisked: what system are you talking about "heterologous-donor, sensitive-indicator"? In this connection, you may be interested in SW-665 (which I will ship), a Xyl- mutant of SW-541. Bruce found this strain to give an unusually high yield of transmotilisations with PLT22/2, and I think the same may hold for the Xyl+ transduction. It gave a (linear at serial dil.) assay of about 1 Xyl+ per 10^7 plaques.*Control assays with SW-435 and SW-666 (a Gal- from SW-543) were not linear (I don't know why, there should have been enough cells), but roughly 5-10x less. SW-665 also seems to be self-lytic, but the plaques were suppressed by PLT22! Another lead that may be pertinent for you: Bruce and I were impressed by the relatively high efficiency of transmotilisation as compared with antigen transduction. I rather suspect this is, after all, due to inhibition by the serum of the trabsduction itself. The transmotilization of SW-543 by PLT22/2, which gives both i and b was rather drastically inhibited by the addition of either serum to the motility agar. There was a 90% inhibition of Xyl+ output from SW-665 by the addition of .02 ml tymur. serum to the plates 15 minutes <u>after</u> the cells and FA were mixed. I would suspect the somatic rather than the flagellar antibodies, but think this problem belongs to you. Anyhow, I am checking the advisability of growing the cells + FA separately some time before inoculating serum-selection plates.

meas. on LT-2*

Concerning the coli galduction, I may have omitted one point. Esther finds that Lp_1^r Lp_2^s (which adsorbs lambda) can be galduced. The usual result is still immune, but rarely the output is lysogenic! (This does impair the definition of immune, but points up the association of galduction with lambda). In at least one case, the result was unstable jointly for lysogenicity and Gal+, in others the two functions separated. This pretty well shows that the phage is essentially passive, but may or may not proceed into the bacterium along with the Gal+/ Your interference experiment sounds ingenious but difficult. At any rate, as many stages as can be tested must be looked at in studying the association of FA with phage. I don't quite see why you are concerned about a linear FA response at low multiplicities. Either a constant fraction of the temperate particles are

genetically effective, independently of each other, or plaque formation does not necessarily mean the loss of the clone (we have some evidence of this in K-12// with the "contaminated" colonies).

SW-543 is getting more and more complicated. The serum situation has improved, allowing some better experiments. For one thing, the spontaneous h reversions show a second phase (only about 3-5 per small plate) which I have not yet identified. It will be a.little strange if Kauffmann had mislabelled this paraB. I have FA from a good many other serotypes: PLT22 works very well on paraB's, abony, enteritidis, san diego, altendorf, and dublin. I am waiting to have the typing confirmed, but each of these seems to give its own phase as well as b phases in the transmotilization of SW-543. I have been using SW-666 mostly, to check on linked Gal+/antigen trahsductions: so far none. Together with your remarks about SW-572, I think we can forget about the chance of an intrinsic origin for the non-b phases. I have not had any luck so far with FA from typhi, stanley, eastbourne, or heidelberg. Can you give me your setup for typhi? All this seems to point very well to linked transductions of pseudoalleles, but I think the explanation is, in its way, simpler. If, in SW-543+ FA(i) [i.e. typhimurium], the i phase is a two-linked gene transinduction, then it should behave like typhimurium in a genetic test, viz. its FA should in turn transduce both b and i. In the one experiment I have tried so far, this was not the case, and only i phases occurred (tested with the help of b-antiserum selection), whereas the controls worked very well: FA from spont, or transinduced b gave only b, PLT22/2 of course gave both b and i. What this means (and the suspicion of which led to the experiment) is that SW-543 is a very peculiar strain whose genotype can be given as A^b B^-. The b transductions are A^b B^+, the i's are A^i B^-, each a one-gene transduction. This can be interpreted to mean that A^b is inhibited by B-, or needs B+, whereas the other A alleles are not dependent on B+. Presumably other non-dependent A^b alleles will be found, perhaps among the spontaneous b. This hypothesis can be checked further in various ways, e.g., a two-step i transduction, obtain via A^b B+ should be A^i B+, and its FA will transduce both b and i to SW-543. I don't know the details of Alexander's dual transduction, but one should, of course, rely on genetic tests like this, rather than inferences directly from phenotypes, as to the genetic basis.

We are bound to run into points of technique and material that will be reciprocally useful, and I am sure there will be no hesitation about exchanging such information. To help explicit statement, why don't we exchange each other's stock lists from time to time? At least, we can give precise examples. I hope I haven't left anything important out of this letter: it is easy enough to forget the frame of reference.

Your CU's never showed here. Esther will take care of it as best she can when we arrange the renewals, very shortly. CU mailings generally seem to have been in a mess this summer.

Sincerely,

Joshua Lederberg

P.S. Why do you suppose SW-435 is avirulent? How about its prototrophic transin ductions? Or can you transduce virulence from LT-2?

JL