If you approve this draft I'll return a clean copy. Josh.

Tape addressed to Joshua Lederberg of The Rockefeller University in New York City in response to a note from Joshua Lederberg of May 16, 1986 requesting information on the history of the transduction by the bacteriology phage lambda.

Good to hear again from you Joshua. I haven't seen you in quite a few years but I keep noticing your activities. All goes well here and I'll perhaps later on tell you of some of the things that are going on but in response to your general questions I think I'll start at the other end of your list of questions and they specifically relate as to how I got to the University of Wisconsin. I had completed a Masters Degree in microbiology at the University of Kentucky in 1947 and after that time I went to work in the biology division of Oak Ridge National Laboratory with a E.H. Anderson. Anderson should be remembered for two things for sure. One is that he was the person who created the M-9 mimimal medium which many people use in growing E-coli. The other thing is that prior to his going to Oak Ridge which occurred in early 1947 that he had been at Vanderbilt with Max Delbrück and at Vanderbilt he had discovered that certain classes of E-coli B mutants resistant to T1 required tryptophane for growth. They were the result of a deletion of trytobane region and I presume also the receptors for the T1 phage. Anderson came to the biology division and set up a section there and I joined him as one of his technicians and the objective of the research was to see if there were other phage resistant mutants which had nutritional requirements and I worked for him for a couple of years. We irradiated E. coli B with electrons from radiactive phosphorus. We had these petri dish size phosphorus plaques which were about 90% pure phosphorus. They had been steeped in the Oak Ridge pile and were very hot and we used them to irradiate E. coli cells which were then allowed to grow up into colonies and the colonies were picked and tested and I was responsible for testing the resistance of these radiation survivors with bacteriophages of the T series T1 through T7 and I think we must have scored somewhere around 60,000 colonies by hand using this technique and I don't remember now I think he found some additional nutritional requirements but I don't think there was any simple correlation with phage resistance. And it was during that time that I became first aware of your work with Tatum. It was discussed in the laboratory, the Journal of Bacteriology paper in particular. And after a couple of years with Anderson I moved into the biochemistry division with a Nick Carter, Charles Edward Carter, who was a biochemist M.D. from the I think the National Cancer Institute where he had been affiliated with Jesse Greenstein and he was a close friend of Alex Hollander the director of the biology division. And I joined his group to work on the synthesis of nucleic acids and E. coli and also bacillus subtilis. I had done some early work on the comparison between E. coli B and E. coli B/r and had noted some things about differences in their pattern of synthesis of nucleic acids and the fact that the B/R strain that I was working with seemed to arrest the stationary cultures at a different position in its GROWTH cycle such that it contained several nuclei whereas the B strain only contained on the average one nucleus so there was a nucleic acid difference between the two strains. And it was during this interval that you came to the biology division to give a seminar which I believe must have been about the time you were working on the persistent heterozygotes of E. coli because I can recall the slides showing the segregation sectoring of the colonies the one that is in the Proceedings of the National Academy of Science paper that you published around I guess 1947 or 48 and during that visit there you came and visited

all the laboratories and the one in which I was involved and we discussed my work and some other things and at the conclusion of that discussion you said that if I ever decided to go back to school and work on my Ph.D. to let you know. And so a couple of years later, several years later as a matter of fact it was in the spring of 1950 or 51 I think 51. Well Hollander used to come around and bug me every day about going back to school and completing my education and I think it was April of 1951 that he came to the laboratory and for the umpteenth time said "when are you going to school" and I said to him "I'm going to go to school in September". I was tired of his nagging and so I wrote to you and we exchanged letters and you agreed to sponsor me at the University of Wisconsin. So in August of 1951 I packed up my family, my wife Halvise and my daughter Margaret who was then only about two years old. I had bought a 1931 model A Ford and so we packed everything up and got into it and set sail for Madison which took us about two days to get there. I had borrowed a house trailer from my parents who lived in Florida and they weren't using it at that time and so we had it set up at a trailer park on the south side of Madison, Lake Winona I believe it was. The park was called Happy Acres Trailer Park. And so I began working in the laboratory in the fall of 1951 and my page #1 in my notebook the first page, is dated October 5, 1951 and that's when I entered your laboratory. I was there I believe for a couple of weeks before you offered me a job to work in the laboratory and I was given a number of things to work on which I'll speak of in a minute or two. Now one of your questions has to do with my notebooks. Now I have three volumes from that period in Madison from the fall of 51 until I left in June of 1956 and I have three volumes, each about 200 pages. Volume 1, Volume 2 and the third volume is a summary of various things taken from the early volumes. Now you ask about having copies of them. I'd be glad to give you whatever you'd like. If your going to have a permanent repository somewhere I could even contribute them to that if you wish otherwise I'll try and make you copies but it will be a little while before I can copy them because each one of them is about 200 pages.

The first experiments that I did in the laboratory had to do with the fact that you had an atomic energy grant that had to do with the radiation resistance of haploid and diploid strains of E.coli and I'm going to include with this tape actually a copy of a report that you wrote to the AEC that covers the interval from March 1, 1952 to February 28, 1953 and its dated March 1, 1953 and it covers a number of things but it also gives your written summary of the early observations on lambda and lambda transduction. Now as you remember when we joined the laboratory there each one of us in the laboratory was given an assignment. Mine was to study the radiation resistance and the induction of prophages in K12 and I have a number of figures here that indicate the induction of lambda by UV and the differences in radiation resistance or lambda sensitives and lambda lysogenics. There is a table in here somewhere that has a plot of it looks to be about 5 or 6 lambda lysogenics and in the order of about 8 or 9 lambda sensitives and of course as you might expect lambda lysogenics are more sensitive as a rule than the lambda non-lysogenics. And I worked during that fall interval in this area. In addition you remember that there had been a set of E.coli strains isolated which had been tested for mating with K12. These were the strains which were called WG strains and I was assigned a couple of these strains to work with I believe. As I go through my notebook here looking at some of these things - I haven't looked at this book in 10 years I guess at least. I was assigned I think strain WG 14 and WG 16 and in these strains we were to isolate auxotrophic mutants and so there are a number of

references to penicillin selections and whether they are mutants or not and I don't know how far that went. I think somewhere down the line I had a few mutants but they were difficult to select because the penicillin technique didn't work as well for those strains as it did for K12. Now in addition and I don't remember the details on this. It probably had to do with the study of the "Lwoff effect" that is the induction of prophage by UV exposure and in that connection I had gotten from Esther several - she had been studying lysogenicity by several other phages of which I think there's one called sigma and there was one called - there was another phage 882 and I think there was a third one. None of these phages grew very well. I don't find in my notes much of any reference. I don't think they were inducible by UV and we were to turn to other things anyhow later on and I don't recall what became of that. Well the thing that was important because I was radiating lambda lysogenics I had quite a bit of lambda phage on hand and they were fairly good TITERS preparations you know in excess of 2-3 times 10^{10} kind of thing and I find in my notes on page 47 and this dated March 26, 1952 that I did attempt a transduction with lambda to 58-161 on D(O) media for the purpose apparently of replacing the methionine requirement of 58-161. I believe that at that time it was felt that the biotin requirement of 58-161 had been lost and somewhere else in here there is a note to the effect that I had done this experiment assuming that the biotin requirement had been lost. Anyhow I plated it out 58-161 with several amounts of lambda and incubated it for several days. It was discarded about two days later with no colonies appearing on the D(O) plates. That was the first attempt that had nothing to do with the lambda Gal business. And then there was a subsequent attempt in the same vein with W1655 which was a non-lysogenic derivative which came I believe from - W58-761 1655 was a lambda sensitive derivative derived from 58-161 and that was attempted on again at about the same time March and this time I got a few minute colonies on the D(O) zero plates and there is a note on March 31 that I tried the 1655 experiment again, again with no colonies being produced and so that was set aside. Regarding the - and this is followed by quite a number of pages on attempts to isolate mutations - auxotrophic mutations of those other strains and it occurs to me that the business of the lambda transduction with gal came about most successfully or rather a lot of confusion came about during the week that you and Esther were attending some meeting down at I believe at Rutgers and Norton Zinder and I were in the lab alone - I think Dotty Gosting was there on occasion but Norton and I were there essentially by ourselves and in the course of doing some experiments I asked Norton if anybody had looked at lambda for to see whether it could transduce anything and its my recollection that Norton said that he thought that Esther had done something in this area but he didn't know what it was and that in any event it hadn't been productive and so I said well I've got lots of lambda phage should we just try it out on everything available. Remember in those days we had all kinds media selective media and so I believe my recollection is that Norton and I agreed we'd try everything possible available and I gave him lambda preps and we just went up to the shelves and took out all kinds of plates and mixed lambda with the right kind of recipients that would possibly demonstrate transduction and plated them out. You might ask Norton if he has any notes of this but this would have been somewhere around according to my notebook somewhere around April the 20th or 23rd and I see in my notes that I made a serious effort to do a lambda transduction and unfortunately I never wrote down the things that were negative so I don't really know which ones we tried any more except that on page 62 of my notes and its dated April 23rd Wednesday 1952 that I did try a transduction with

W1736. That was a strain that I had been using with regard to those lysogenicity studies with 882 and some of the other prophages that Esther had given me. Anyhow I had plated it out and my notes indicate on that page that I plated it out with on EMB Lac with lambda and I had used increasing amounts of lambda, a couple of plates with no lambda then plate with .05, .1 and .2. Then a standard amount of 1736 cells, then my notes indicate that I looked at the plate on the 24th and there was no papillae on the 24th and on the 25th with two days a papillae began to appear and then on the 28th which would be five days later it indicates that spontaneously there had been produced 32 and 26 papillae whereas with 0.05 mls of lambda prep there were 186 papillae produced with .1 ml of lambda preparation there were 274 and with .2 there were 372 papillae. And then I note next to it that at some later date in red pencil had added some ratio of papillae per lambda plaque on the unit. Now the confusing thing was that these were on EMB Lac plates and subsequent that I purified these papillae by restreaking them on EMB Lac and of course they came out ~~like~~ Lac minus which was very baffling because they had been selected by this EMB medium and how come they were ~~like~~ negative and then I did it again a couple of times. Actually this is followed by quite a bit more information on radiating that HFT strain 267 that I'd been studying getting SURVIVAL AND induction and things on it for the Lwoff effect. So I didn't pay that much attention to the lambda transduction thing with W1736. But over on the 28th I had streaked them out on EMB Lac and they came out ~~like~~ LAC minus and then I replicated them to galactose and it says here that in one case 6 out of 7 were gal+ and 16 out of 17 were Gal+. Now it's my recollection that at this time you had returned and Esther had returned from the Rutgers meeting and that somewhere in the course of this confusion about the selection on EMB Lac and they were ~~at~~ minus that Esther had made LAC a suggestion that I should perhaps look to see how they were with regard to galactose. It's my recollection that Esther had done her Ph.D. thesis on the interaction between Lac and Gal and she'd done a number of things on Gal and it was a good suggestion. And I believe that's when we finally got on to the right track with regard to lambda transduction of the Gal genes and that would have been in late April of 1952 but it was a long time before anything was really published on it. My notes thereafter again go back to referring to radiation of the HFT strains and also working on WC 14 where I'd isolated both the proline and the tryptophane requiring mutant. And this continues on and for some reason which is not clear to me now there are a number of confusing EXPERIMENTS, . I became W1736 involved with a W112 strain which I believe was an unstable Lac- strain and that didn't help things at all. It made things more confusing. But anyhow I still continued with the radiation experiments for many pages here and actually on May 8 I show transduction to W112 Lac- and again I got A RESPONSE a response that's kind of confused and again the papillae that I got were THAT WAS CONFUSED. Lac pink they weren't strong pluses and then some of these appeared within (?) same time same day May 8th a transduction to W1376 again THE PAPILLAE I GOT WERE with lambda and it is at this time that it must be that I discussed this with you because I introduced control here which I associate with some conversation with you that is I used BOILED lambda prep as a control because we were concerned about whether the lambda preparation itself was providing some sort of release of selection and allowed things to grow up sort of a non-specific (?) . On this date I show that I used lambda from K12 which had been boiled and non-boiled and then I also and this preparation gave me papillae. The boiled lambda gave me 17 and 22 papillae per plate whereas the non-heated lambda gave me 324 and 345 papillae per plate which was a significant difference. On Friday the 9th of May I did

SENSE

another experiment with W1736 increasing amounts of lambda preparation and no lambda and I got increasing number of papillae which is almost linear with the amount of phage added. I did this both on Gal and on Lac and again as it shows that the lactose provided some sort of selective thing but the experiment on the EMB Gal was quite clear cut. And I noticed on this date that I had respread some of these papillae and evidently we were still concerned about the possibility of a selection of the lambda prep by itself and not the transduction because my notes say that this finding suggest that papillae on EMB Lac control are different than papillae on EMB Gal that the Gal transduction effect does not now appear as a release of negative inhibition against Gal+ clones. And I note that a few days later on the 13th of May 1952 that I tried a transduction with lambda and boiled lambda with regard to the SERINE - glycine markers the W1678 and this is on D(o) medium and the results indicate that as of six days later I got one colony and three colonies on these plates and that there was not an indication of transduction.

Now this thing continues and I'm still radiating H267 for survival for lambda induction. I notice on May 15th that I try to do a lambda transduction of the leucine through the leucine requirement of W1736 which again came out negative. I used heated lambda at this time as a control. But I also noted at this time that in some of the streaks of some of the Gal papillae it says on page 75 that papillae that I picked from the W1736 transduction on Gal they all grew, they had no other nutritional requirements that they all appeared Lac- and that there ~~receptoring~~ and again at this time on the Sunday the 17th I picked from the papillae from the ~~W1376~~ W1736 transduction and this was done on Gal and they were picked to lactose ~~in the~~ AND SHOWN TO BE LAC ~~light~~ negative or very slightly light positive and they were then replicated to Gal and of the ones that I tested 34 out of 35 were Gal+. But again I'm still working with a phage called 882 on this page and there is still something going on with this W112 strain which seemed to be contaminated with some kind of phage. And for some reason which is not clear in the next few days on like May 20th I'm doing experiments with a strain called 1998 which apparently is a lambda lysogenic derivative of this strain WG28 and again it's a case of looking for the induction of lambda as a consequence of UV radiation looking for induction in one of these other coli strains. And on the 27th of May I did for reasons which are not clear I irradiated a lambda preparation and used it as a source of transducing phage for Gal 2W1736 and I showed that with increasing dose of UV going from no radiation to 240 seconds that the number of papillae increased from somewhere around 200 to something like 3200 with that increasing dose of UV such that irradiating the lambda made it a better transducent vehicle than non-irradiated lambda. And I note also that I tried some transductions in another strain W1662. By the 28th of May we had not decided to go full speed on lambda although I did do an experiment with W811 which is Gal 4-lysogenic and I did get evidence for transduction there again using irradiated phage and boiled lambda as a control and I think that - but I'm still continuing to do the Lwoff type experiment. We hadn't decided yet. And on the 29th I tried to do a transduction with lambda with irradiated lambda with regard to METHIONINE MARKERS of 58-161. And yes at that time also we decided to do some reconstruction experiments by putting Gal+ strains in the presence of a large background of Gal- and this was done with W1736 again and again I see that it shows evidence for the increasing numbers of papillae as a function of lambda concentration. And it was this stage that we decided that we had to see if it had something to do with lambda specificity and

so I obtained from somewhere strain W1439 which is a Mal- -derivative I believe of W1436 and it would be lacking lambda receptors and would not absorb lambda and I did a transduction with it as the recipient using heated lambda and even irradiated lambda and my notes for June 1 show that these exposures of W1439 to lambda and irradiated lambda did not produce any Gal+ papillae suggesting that lambda was the vector involved in the formation of the Gal+. It is here my notes start mentioning Gal "transduction" in a couple of places. And on subsequent pages the experiment was run on xylose, EMB xylose medium with 1821. It's my recollection that the xylose medium had some selective potential for Gal+. This is indicated by the fact that there was a large number of papillae TO BE produced on xylose which subsequently must have tested AND SUBSEQUENTLY SHOWN Gal+. And it must have been about this time around the 1st of June in 1952 that we decided not to continue the studies of the UV induction of lambda and of the radiation resistant differences between lysogenics and non-lysogenics because I see my notes and I begin to devote most of my time to Gal transduction with various strains and included checking the Gal+ to see if they had been changed for any other markers. We then moved to some other strains W902 which was a Gal 2- strain and I see that it was done on the 10th of June and again for reasons I don't understand that W112 strain is involved, I DID A CROSS with W1655 for some reason doesn't tell my why. But I now begin to see that I worked with W1736, W1811 with Gal transductions and I begin to see in my notes that I HAVE ACCUMULATED PHAGE preparations and on June 16th of 1952 I received W750 which is a Gal 1-strain so I've now looked at Gal 2, Gal 4 and now beginning to look at Gal 1 for transduction. It says I did this experiment back a few days earlier on the 10th of June I did with Gal 1 and showed that the control Gal 1 strain is a very stable gave very few spontaneous reversions of Gal and I note that with no phage I got zero papillae and with .10 ml of lambda prep I got 409 which is a very clear cut difference. I also see now that perhaps some of the W112 crosses had to do with the presence LP2R because it looks like there was some crosses made here to see if LP2R was involved in whether the transduction would go successfully or not. But on page 94 which is June 23, 1952 I used as a control I did a transduction to Gal 1 on EMB Gal using Gal 1 cells and using heated lambda and using lambda derived from Gal 1 and the results indicate that heated lambda gave only 2 papillae, day 12 lambda gave 405 and lambda derived from Gal 1 itself gave only 2 as expected. I see a few days later that we hadn't given up on what kind of genes lambda could move around. I see the evidence for an experiment to see if it could transfer streptomycin resistance which again was unsuccessful. Now for the hetergenotic state of the transductions of lambda I find on page 96 on Tuesday, July 1, 1952 that I had picked the transductions from the Gal 1 transductions mentioned on page 93 and I had streaked them out and they had been streaked out, four streakings it says, and that they were still segregating and stocks were made and these were now given the label of their origin that is to say 750T902 was a transduction clone derived by phage from 902 being applied to 750. Be it this time that we really became aware of the fact that they were heterogenotic because I see further down the page that I had taken a papillae from a transductions from wildtype to Gal 1 and from wildtype to Gal 4 and these papillae had been streaked out in the case of Gal 1 five times and their still mixed after five individual colony selections and this then now done with a W1736 which I believe is Gal 4 strain also and they were picked and streaked out. Now it says three times, three times and five times and their still segregated.

Now in early July I received from Esther W892 I believe it was a Gal 3 culture and I spent I think another month or so on this culture not realizing that it was mixed. It was a mixed culture which confused things. But I note also that I continued to look at the segregation aspect of the heterogenomes and seemed like I've now ceased to do the other kinds of experiments on Lwoff effect and on the radiation things. And this continues until the next number of pages covering up until the end of 1952 had to do with looking at the stability and that sort of thing and whether they are lambda lysogenic. These were still from the LFT preparations and it ISN'T UNTIL over around February early 1953. I had not bothered to irradiate any heterogenOTES to see what kind of phage they had given. It's my recollection that on one occasion I had done this and had got such a high titered phage preparation that I thought there was something wrong. Remember in those days, this is before the Millipore era, and we used to sterilize our phage preparations by adding a few drops of chloroform and shaking them up. It's my recollection that I had done this and used this a particular preparation for transduction and it came up with the amount of phage plated overwhelmingly plus and so I felt that the thing had not been sterilized sufficiently or hadn't been sterilized. With the chloroform I remember putting additional chloroform in and shaking it all up again and repeating the thing and finding again that it was still giving such a high yield of transductions that I thought it was contaminated and so this continued and I see that on March 10, 1953 that I have notes stating transduction with high activity phage and I got tremendous number of papillae. This was the phage derived from the - it doesn't give the exact source here but I believe these were experiments that were taking the segregants from say a HETEROGENOTE made between Gal 2 and Gal 4 and attempting to classify them as to what they were 4 or 2 and using I guess LP T lambda but I'd taken a - my notes are not clear here but it's my recollection that I did not use Gal+ heterogenotes as a source of phage and that I had taken some of the segregants for a heterogenote between 2 and 4 and attempting to classify these had made preps from them such that the high frequency transducing lambda actually was first picked up with regard to segregant from a heterogenote which happened to be actually itself was a homogenote. And it gave me very high titered phage preparations. I note on March 30, 1953 that I tested a phage prep W892 high powered lambda it says and asking the question is this high powered ~~DNA~~ susceptible? And so I treated it with DNase for 10 minutes at 37 degrees centigrade and it indicates that this lambda prep diluted 1 to 100gave ~~no~~ gave more than 10^4 papillae per plate which indicates that it probably had a titer of about somewhere 10^6-10^7 transducENT 10^6 particles per millileter which is about the way they ran at that time. That the transducing particles SO-CALLED lambda DG - that was a term coined by John ~~Gigall~~ and Werner ARBER Kallenberger in Switzerland around 1956 a couple of years after this time ~~but we needed~~ some of the lambda particles WERE defective is actually contained in the first paper that we wrote where there is a table which shows that some of the transductANTS were defective. Yes in the first paper that we wrote on page 152 Table #8 examination of the colonies after exposure to HFT lysate. There is a column headed LPR which meant that they were defective phage particles. Actually John ~~Rigall~~ apologized to me for not having noted that. They made such a big thing of the defectiveness of lambda that he had overlooked that we had said in fact that some of the lambda transducing particles were defective and that he had overlooked that particular Table. There's some other things that relate to this that I can tell you separately with regard to Allan Campbell's participation in this - End of Side A.

WEIGLE

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Side B

CAMPBELL HAD A PAPER → WEIGLE WEIGLE

publish which was incorrect and as a consequence WEIGLE discussions that he'd had with Bigel I think this was at Cal Tech and ~~Vidal~~ had communicated - this would have been sometime in 1956 which is two or three years after the material we're talking about in my notebook that he must have communicated to Campbell that ARBER , he and Kallenberger had shown that lambda particles that were doing the transductions were probably defective and that Campbell changed his whole manuscript to fit this concept and John ~~Bigel~~ wasn't very much pleased with this. Okay to return to my notebook and the interval now is late April, early of May 1953. I began a serious classification of the Gal- which were segregated from a heterogenote and I see notes on the back that I made a heterogenote between Gal 1 and Gal 2 and that I had collected a number of segregants from this heterogenote and I had classified them by testing them against low titered lambda preparations from Gal 1 and Gal 2 to classify what they were and that indicates that there were 18 segregants which were Gal 1 and there were 5 segregants which were Gal 2. The source of the phage had been Gal 2. And then there are indications that I had taken some of these segregants and also tested them by crossing. There were available strains that I could do bacterial crosses with. And I'm still stumbling around with this HFT phage which I didn't understand and did some experiments with it to relate the transduction to lambda I find an experiment on page 199 which is May 16, 1953. This is an experiment I don't remember at all. We obtained somewhere I guess maybe Cal Tech some anti-lambda serum and I tested the inactivation with this anti-lambda serum. I tested the inactivation of placque-forming ability and transducing ability and I find on page 199 that after treatment with anti-lambda serum that I got placque survival of 43.3% and on the same preparation I got 39.8% survival of the transducing activity a suggestion that in fact the transducing agent is lambda. It was also about this time - I see the first experiment - that I began irradiating lambda preps to show - this would be the 18th of May 1953 - that irradiation of lambda greatly stimulated the transducing activity - I have a graph that's on page 200A which shows that I irradiated a lambda prep and plated it on several hosts and got increases in transducing titer of the order of like 50 or 60 fold in this graph. And this is at a time when the placque forming ability of the lysate had dropped two decades. And that page 200B shows a similar prep that was asssayed on Gal 4 and it was assayed on recipients which were lambda sensitive, lambda lysogenic and lambda carrying a defective prophage.

I see by page 202 it appears I now have available HFT lambda preparations that could be used for typing Gal 1, Gal 2 and Gal 4 I see that there is a mention of an HFT Gal 4 on this page. This is dated June 6, 1953 and this is followed in the next two pages by an attempt to transduce not from - to + but from + to - with these new HFT preparations. That's on June 16, 1953. I'm now nearing the end of my first volume which ends on page 218A and these pages between say 180 and 218A have to do with classifying the segregants from heterogenotes, irradiating lambda to show that it increases the efficiency of transduction in a number of experiments. However as yet I have found no indications that they were tested for whether they were defective LYSOGENIC or not. There are all sorts of experiments in here where I used a lambda, Gal 4 culture which was lambda defective as a recipient in transductions and it shows that most of the transductives came out defective because the recipient was defective but

are occasional cases where the transductant came out lysogenic. On page 216 there's a case one of the transductants came out lysogenic, 28 were still defective. This comes to the end of my first volume so I'll have to stop here and get the second one - bring it with me to the library.

There's been an interruption of about a day in this tape. I'm now resuming on June 11, 1986 and I'm starting in Volume 2 of my notes which begin on page 219. The starting date is August 26, 1953.

In the pages that follow here there are a number of experiments which were done for the purpose of tidying the lambda transduction up that is to say they are experiments that deal with varying amount of cells per constant amount of phage, and varying amount of phage vs a constant amount of cells in order to get linear relationships between the number of transductions ~~in~~ and the dilution of the phage preparation. And this is then followed in a bit when I first began to try to grow lambda lytically and on page 227 there is a reference to lambda ~~made~~ grown lytically on W518 and evidence that this lambda did not transduce Gal genes when it was applied to W2175 on EMB Gal. The titer was very good titered phage 3.3×10^9 ~~3:3 X 5~~

Then there's another experiment shortly thereafter page 228 was lambda grown on W1485 which also was not active when plated against three separate strains W750, W518 and W2175. I then began to study something of the charactistics of the segregants from heterogenotes and was surprised to find that on October 6, 1953 that I was beginning to find some of the Gal minus' that were segregated from transductions were actually lambda sensitive which was kind of puzzling because we had not ~~talked~~ much about that. Thereafter I did a number of these same sort of things and remained a bit puzzled by the fact that the segregants were sensitive. Thats in some cases not every case but in some cases they were sensitive. Then on page 241 this is November 8, 1953 I did an experiment with HFT lambda such as to get low multiplicities of infection and I note that my notes say that I obtained some transductions under these conditions and that of 24 that I tested that one of them of lambda lysogenic and the other 23 were "LPR" which could mean that they were lambda defective and this is then followed on note from November 17, 1953 which says in a repeat experiment one unstable plus was obtained. An analysis of this plus, 4 examples of it showed that it was lambda resistant but it was not lysogenic which I take to be the best definition of a lambda defective.

On November 16, 1953, page 244, I note the isolation of W2341 which was the result of an attempt to put Gal 2- into a Gal+ HFR and it turned out that W2341 and the notes indicate that it was not sensitive to lambda and it was not lysogenic when it was tested against W518 and it was sensitive to lambda vir. And so that this was another definition of a transduction made by a defective lambda. Going back a few pages I notice that on October 26, 1953 the isolation of W2350 which is the first case of a double Gal- produced as a recombinant from a heterogenote between Gal 1 and Gal 2. And thereafter there are many pages that deal with quantitation getting linear graphs as a response. There are also quite a bit of space to go to collecting Gal minus' from heterogenotes and classifying this to which allele they were. And then there is a series beginning in December of 1953 where I began to make transfers of Gal alleles to other strains by means of HFT lambda and then I also began to isolate some new ~~lambda strains~~. gal alleles.
I believe I referred earlier to Table 8 in our first paper on page 152

and I find by looking at my notes that that was an experiment which occurs on page 254, an experiment which was begun on January 11, 1954 and my notes indicate at that time I had done a transduction with HFT Gal 2 onto — on I don't see what the recipient was here but this was the origin of W2866. Perhaps the stock book would say what that was but anyhow this is a case where I got three defective transductants vs 23 lambda lysogenic transductions. Also this page shows that I did another experiment with lytic lambda ~~litaglanda~~ and this is labelled retest #3 where I applied ~~litaglanda~~ to W750, W2281, W2373, W518 and W811 and there were a very small number of papillae produced in each case and these were streaked out. The notes say that the papillae were either all stable or slow pluses against indicating ~~litaglanda~~ does not transduce. In February 13, 1954 I did a double transduction here. I took the Gal 1, Gal 2- produced mentioned earlier and I applied to it HFT phage from Gal 2 and HFT phage from Gal 4 and obtained heterogenotes apparently the result of double transduction. I guess that was just sort of a fun experiment. No I'm sorry W2350 is 4-8-, Gal 4- Gal 8- according to the notes written on the margin here. Again in February 1954 I did an experiment where I got 3% of the cells that transduced to Gal+. This was some experiment apparently done in connection with Esther because it says that Esther's EM analysis in her book on page 667. Then in March of 1954, the 25th, there is mention of an experiment with Tom Nelson with regard to the transmission of Gal the fragment in crossing. I don't know exactly what the results of this experiment was. During this time I had a great deal of difficulty maintaining HFT stocks particularly the homogenotic Gal minus' and there is considerable time spent in reisolating strains that were lost. And then I see in April 1954 I was working on making triple Gal minus' 1, 2 and 4. Also in April of that year I noticed that I did some experiments with HFT lambda preps applied to the Mal- lambda 2 resistant Gal- W902 for example and obtained from this apparently lambda — penetrated the cell in the absence of lambda receptors and I got some transductants small in number. We had been concerned because of the failure to get good wildtype transductants from applications of HFT 1 onto Gal 4 or HFT Gal 4 onto Gal 1 And then the question was asked and this was on April 14, 1954, is the complex the heterogenote between 4 and 1 phenotypically plus? And there is a note that the pluses appear to be secondary and segregating minus'. And then I did an experiment at that time with HFT 1 applied to W518 and got 2 transductants which papillated and gave off pluses. The heterogenotic cells were phenotypically negative at the early stages and in the course of growth apparently recombined to give Gal pluses. Again over in June 1954 I see reference to additional experiments with ~~litaglanda~~. Again as if we were not completely convinced that ~~litaglanda~~ did not transduce Gal genes.

In July of 1954 I was back working on the quantitative aspects of HFT phage where I was finding out if HFT Gal 2 was linear on Gal 1 and Gal 4. There is a graph showing this response. Then I see also again on September 9, 1954 a further examination of W2341 to find out what — Hfr its status was with regard to lambda. It was isolated as a ~~HFT~~ Gal 2- which turned out to be lambda resistant and Tom Nelson isolated some lambda sensitive segregants from ~~them~~. And there is a question here at the head of the page that says, is this an indication of its LPR or LPS? And so it was streaked out and tested and the LPR's (those would be lambda defectives) Gal+ reversions were selected, segregating minus' on streaking, ~~and the~~ 6 lambda sensitive, that were picked ~~up and they~~ were ~~scored to be stabilized~~ stable ~~streaking and then the~~ ~~is~~ therefore 2341 is shown to be 2-LPS

Gal+ That that were lpr were

over 2-LPR. Coming from the following reaction it is a transduction of the 2-LPR phage into a gal+ lambda sensitive recipient which then yielded this heterogenote which is now recombined to become Gal 2- but is ~~heterozygous for~~ carriers of defective lambda phage. There is an interesting experiment on September 20, 1954 on page 288 which is raising a question about W2344 which is a T1 resistant derivative of 2341 which is the Gal 2- which is carrying a defective prophage (homogenote) And there is a question of testing colonies of 2344 and of the 17 of these colonies were tested and they were all lambda sensitive and it says suggestion that only LPS segregants of 2341 can be made BIR. Then there is a note that says since ~~reporting~~ to Cavalli suggestion crossing over between fragment and chromosome Gal- results in a diploid for BIR and Lac? I now see that on October 1954 on page 292 that I was examining the segregants from a heterogenote which is drawn as being R over S with regard to lambda and then there is a classification of some 15 or so segregants from this both with regard to their Gal type the Gal-allele and also with regard to whether they were lambda lysogenic or lambda sensitive ~~or lambda defective~~. ~~You disperse~~ with this there were a number of experiments that had to do transductions to W1924 which was a Gal 4- strain that carried a defective prophage. I never really looked at these experiments. I'm not sure what they show.

Over on page 295 November 1954 in another experiment I tried to make transductions with virulent mutant of lambda and I don't know what the outcome is. I have to study this a while before I can figure it out. In January of 1955 I notice that I'm studying the position effect between Gal 1 and Gal 6 and that there were heterogenotes that were phenotypically ~~particularly~~ minus which papillated and then made on petri dish analysis ~~of~~ the segregants that they produced. Then I see the notice with double Gal- of this experiment as well. There follow many pages where there are making various kinds of stocks with the various Gal minus' and HFR and then over in June of 1955 I began making measurements in a Luria-Delbruck kind of experiments on the cross-over rates. There produced between certain non-complementing Gal minus'. And I see that in January 25, 1956 page 390 that I have a section heading Preparation and Stocks for ~~Calcar~~ Kalckar NIH for Study of the Biochemistry of the Galactose Fermentation. And here I made a number of transductions into a common background which I believe is 2637 which as I recall was a strain that you gave me for some experiments that you were doing with the micromanipulator and this was a number of transductions and then there is a classification of some of the progeny and it says then go see page 396 and this is where they were first categorized as a group and they were first given the numbers W3091 through 3097 and W3100 to 3110 and that is February 17, 1956. However it didn't stop there. There was some confusion and we started again on March 8th making preparation of the prototrophic F stocks with Gal markers for the Kalckov stocks and again there's a table that sets up the strains and there are actually filter paper ~~tape~~ pick ups of the some of the transduction plates that classified ~~this~~ these stocks. Then in April I began doing some experiments that had to do with the yields of ~~translucent~~ transducing particles from induced, UV induced cultures, and I see from information on one step growth curves which I believe indicated that UV only got a very small particles for induced lambda. It has to be in the absence of ~~halperphage~~ I presume. And I go over and I find that my last entry which is page 421 is dated May 30, 1956 and it was at about that time that I left Madison. I arrived here in Denver at Webb-Waring Lung Institute June 20, 1956.

That is an overview of my notes verbally. I'm prepared to send you whatever you wish from them but perhaps after you've heard what I've recorded for you, you may want to make a little more selection. I have no objections to copying everything for you.

Aside from that here in Denver everything goes pretty well still. I'm still working in the lab. I have a grant in radiation biology on the oxygen effect which is very promising at the moment and it runs until November 30th of next year and hopefully with a good performance it will be renewed. Otherwise here Halvise is involved herself in cytogenetics. She works for Childrens Hospital doing cytogenetics of leukemia. She works for the Eleanor Roosevelt Institute, that's Ted Puck's empire. She doesn't work for him personally but she works for that group where she does cytogenetics on hybrids between human cells and hamster cells. Otherwise Margaret - we went to Margaret's commencement the first part of May. She finally got her Ph.D. from the University of North Carolina in comparative literature and now is looking for a job. She has a great many talents but the good jobs aren't very frequent so I don't know what she is going to do. Larry after finishing school here went to Yale and got a Bachelors Degree in molecular biochemistry and biophysics and he has returned here. He was unable to get jobs with some of the companies around here that are in molecular biology but he has finally obtained a job here as a technician in the gastroenterology division where he is doing quite well although we are encouraging him to go back to school which I hope he will do at some stage. I have nothing more to add. I hope to hear in some kind of a response to you. I'm sorry if I have to do this on a tape. It means you have to take a hour to hear the whole thing but you can always play it in the bathroom if nothing else.

So good luck and thanks. Larry

Glad to hear about Margaret + Larry.

Jack

ewq