The following is a true copy of my notes, Volume III, labelled "Summaries" which were recorded during the interval 1951-1956 while I was in the Lederberg laboratory in the Department of Genetics at the University of Wisconsin in Madison.

The notes were entered in intervals with some pagination and represent various types of summaries, speculatiuons, etc. I have numbered them sequentially in the upper right hand corner from page 1 to page 253 for convenience.

There are a number of irradiation experiments (UV) plotted which have never been published. I remember communicating some of them to E. Kellenberger who may have giventhem to Werner Arber since I believe there are some similar experiments in Arber's doctoral dissertation. There are also some drafts of my own disseration.

Items of possible interest are the handwritten notes of J. Lederberg (4/10/54) labelled "Remaining Questions" on pages 90-92; the typing bill for my disseration, page 155; a status report of the Lederberg lab for 1953, pages 161-195; some notes of JL on putting the stock book on keysort cards, page 197; a matrix by JL for transduction mapping, page 201; and an index to Volume II of my notes, pages 202-206.

This Volume is a hodge-podge and doesn't represent any temporal order - I believe page 253 is really ahead of page 1 which was a preliminary report leading to my dissertation.

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Summaries

Research

M. L. Morse Genetics

Materials and Methods

The principal cultures used are listed in table 1. In summary they represent three distinct mutations which lead to the loss of ability to ferment galactose (Lederberg, E., 1950). The Gal_1- and Gal_4- stocks are the result of a single mutation to (-) in each case, while the Gal_2- stocks represent two independent mutations to (-) whose identity is based upon the observation that no(+) recombinants have been observed in more than 11,000 prototrophic recombinants from crosses between them and upon the synonymous behavior of the stocks in transduction experiments. These three loci are closely linked to one another as indicated by the data in table 2, but the order of the loci is not specified.

In addition, each of these loci is known (Lederberg, E. and Lederberg, J., 1953) to be closely linked to the Lp or latent phage locus of E. coli K-12. Three alleles are known to exist at the Lp locus; (1) Lp+, overtly lysogenic and showing evidence of free phage in cross brushes with Lp^s forms, and resistant to lysis by free lambda phage, (2) Lp^r, not overtly lysogenic and showing the presence of free phage in cross brushes with Lp^s forms, but resistant to lysis by free lambda phage, (3) Lp^s, not lysogenic, and being lysed or lysogenized by free phage.

More extensive delineation of the interrelationship of these loci has not been feasible until recently and it is hoped that with the aid of a new method of distinguishing the minus recombinants that mapping of this region may be accomplished.

Another locus which has a direct bearing upon the problem is the locus controlling resistance to lambda-2, the lytic mutant of lambda. This locus, Lp_2,

has an epistatic effect upon events controlled by Lp (Lederberg, E. and Lederberg, J., 1953). Thus a change from Lp_2^s (lambda-2 sensitive) to Lp_2^r, (lambda-2 resistant) results in a loss by the cell of the ability to adsorb lambda as well as lambda-2. Sensitivity to lambda by a cell therefore can be masked by the presence of the r allele of Lp_2. The Lp_2 locus is not closely ~~is not closely~~ linked to either Lp or to any of the galactose loci in question.

Methods of cultivation and media used were as detailed in Lederberg, J. (1950). Liquid cultuivations were in penassay broth medium, with or without added aeration, solid media used were of EMB base, either with or without added sugar, or for crosses, a synthetic form of EMB, EMS was used.

High titxered lysates of cultures were prepared after the method of Weigle and Delbrück (195) by inducing lysis of penassay grown cells by means of irradiation with small doses of ultraviolet. The UV was adinistered to saline x suspehsions of the cells and the cells subsequently diluted with 2X penassay broth and incubated with aeration until maximal clearing was obtained.

Lytically grown lambda was obtained by infecting the sensitive cells by exposures to lambda prepared by the Lwoff technique, discarding the supernatants after the adsorptions and resuspending the sedimented cells in nutrient saline broth. The NSB suspensions were then incubated with aeration until maximal clearing was obtained.

Table I

List of pertinent cultures

Culture	Genotype
K-12	$F^+ Lp_1^+ Lp_2^s$
W518	$F^+ M^- Lac_1^- Gal_4^- Lp_1^{+s} Lp_2^s$
W750	$F^+ M^- Lac_1^- Gal_1^- Lp_1^+ Lp_2^s$
W811	$F^+ M^- Lac_1^- Gal_4^- Lp_1^+ Lp_2^s$
W902	$F^- T^- L^- Th^- Gal_2^- Lp_1^+ Lp_2^r$
W1210	$F^+ M^- Lac_1^- Gal_2^- Lp_1^+ Lp_2^s$
W1436	$F^+ T^- L^- Th^- Lac_1^- Gal_4^- Lp_1^s Lp_2^s$
W1924	$F^+ M^- Lac_1^- Gal_4^- Lp_1^r Lp_2^s$
W2175	$F^+ gal_2^- Lp_1^+ Lp_2^s$
W2281	$F^+ M^- Lac_1^- Gal_2^- Lp_1^s Lp_2^s$
W2342	$F^+ Lac_1^- Gal_2^- Lp_1^+ Lp_2^s$
W2373	$F^+ Hist^- Leuc^- Gal_1^- Lp_1^s Lp_2^s$

Table 2

Recombination between the Galactose Negatives

Cross	Minimum Number of Prototrophic Recombinants	Percent (+) Recombinants
F^+ Gal_1- X F^- Gal_2-	(1) 1500	0.13
	(+) x (-) (2) 6517	0.06
	(-) x (-) (3) 3603	0.027
	11620	0.06
F^+ Gal_4- X F^- Gal_1-	4588 $\frac{Gal^-}{Gp^+} = \frac{5}{1289}$	0.13
F^+ Gal_4- X F^- Gal_2-	2654 $Gp^+ = \frac{1}{1365}$	0.23

Results

When varied amounts of high titered lysates of wild type cultures are mixed with Gal_1-, Gal_2- ~~and~~ or Gal_4- cells and plated on EMB galactose medium, results such as those in figure 1 are obtained. Since each of these mutations to gal- is capable of reverse mutation the data shown in figure 1 have been corrected for the number of reversions by subtracting this number as determined from control platings with no added lysate. Figure 1 shows that with increasing amounts of added lysate there is a linear increase in the number of galactose fermenting papillae per plate. In addition, figure 1 indicates that lambda sensitive cultures appear to be more capable of showing the effect of added lysate than lysogenic cultures or cultures carrying a non-plaque-forming type of lambda.

When lysates of gal- cultures are mixed with the various gal- cells and plated upon galactose medium results similar to those shown in table 3 are obtained. Each of the lysates of the gal- is capable of evoking galactose fermenting papillae uppn plates spread with the other gal- cell types but not with plates spread with cells of its own type. The ability to induce gal+ clones in other gal- but not with cells of type corresponds to the differentiation of these gal- mutations by recombinational analysis. Evidence complementing this is shown in table 4 which shows that the ability to evoke papillae with cells of type is restored by reverse mutation. Presumably phenotypic reversions can be of two types, reverse mutation at the mutated locus, and mutation at a second locus whose action mimics the action of the first gene. Reversions of this second class should not be able to evoke papillae from cells of type. Such reversions as the latter have not as yet been investigated.

Table 3
Interaction of Gal_1-, Gal_2- and Gal_4-

		Lysates				
		None	Gal_1-	Gal_2-	Gal_4-	Wild Type
	Titer($\times 10^{10}$)	-	2.4	4.9	1.7	1.4
Cells						
Gal_1- Lp^+	(1)	2*	-	176*	43	-
	(2)	2	2	-	-	405
Gal_2- Lp^+	(1)	14	52	11	43	-
	(2)	20	-	10	-	356
Gal_4- Lp^+	(1)	89	-	202	-	-
	(2)	50	85	-	-	417
	(3)	47	-	-	50	394

* Number of papillae per plate, 0.1 ml lysate plated. Between 10^8 and 10^9 cells plated

Table 4
Restoration by Reverse Mutation of the Ability to Transduce Previously Nontransducible Loci

Locus (all Lp^+)	Reversion	None	Addition Reversion Lysate
Gal_1-	Gal_1^+ #1	0	648*
Gal_2-	Gal_2^+ # 1	10	96
	Gal_2^+ # 2	6	552
Gal_4-	Gal_4^+ # 5	39	204
	Gal_4^+ # 8	25	291

* Number of papillae per plate, 0.1 ml lysate plated Between 10^8 and 10^9 cells plated.

Examination of the other characteristics of the cells transduced to gal (+) by lysate exposure has uniformly shown no changes in any of them with the exception of the induction of lysogenicity in the lambda sensitive forms. Direct attempts to transduce other factors have been uniformly negative. A summary of the avilable data is given in table 5. In connection with the negative results in attempts to transduce xylose and lactose loci it should be noted that both xylose and lactose containing media have some selective value for galactose fermenting clones.

Transduction in K-12 thus far has been found to be limited to several galactose loci closely linked to the latent phage locus, Lp. These loci include Gal_1, Gal_2, Gal_3, Gal_4, Gal_6, and possibly several more that have not as yet been classified. The experiments reported here will concern only Gal_1, Gal_2 and Gal_4 although some observations on Gal_3 and Gal_6 have been made. Not all loci controlling galactose fermentation are transducible. One occurring in W2312 will be mentioned later, and another induced by copper treatment by Helen Bayers has been found.

The transductions described above have been effected by means of lysates perpared by the Lwoff technique of inducing lysis with a small dose of ultraviolet. Lysates prepared by lytic growth of the phage on a sensitive culture apparently have no transducing activity and have lost the transducing activity included in the inoculum. The inability of this type of lysate to transduce is demonstrated by the results given in table 6.

The necessity for lambda adsorption for transduction is illustrated by the results given in table 7. When the various gal- are found coupled with the r allele of Lp_2, a combination which is incapable of adsorbing either lambda or lambda-2, transductions are not observed. The presence of this allele of Lp_2 does

not interfer in the capacity of a culture to give rise to transducing lysates and the transducibility of a transducible gal- locus found coupled with Lp_2^r is demonstrable when a suitable cross is made and a gal- Lp_2^s recombinant obtained.

Recovery of the transducing activity of a lysate by the method of mixing lysate and cells on plates appears to be good in the case of lysogenic cultures, the variation being less than two-fold over a thousand-fold change in the number of cells plated (figure 2). This is not the case when the added cells are lambda sensitive, the variation being in this case two or three-fold greater over a similar range of cell concentrations. It should be noted again that the lambda sensitive cultures give approximately ten-fold or more transductions at any cell density, and that the relationship of the activities on the two types of cells is not known. The ratio of number of transductions to phage content of the lysates approximates 10^{-7} for lysogenic assay cells, about 10^{-6} for sensitive cells.

Alternatively to mixing cells and lysate on plates the transducing activity of the lysates may be adsorbed upon cells and the cells then plated out on agar. Table 8 gives some indication of the adsorption of the transducing activity and also some indication of the adsorption of the phage under the same conditions.

Table 7

Effect of the Lp_2 Locus on Transducibility

Galactose Locus	No Addition	Wild Type Lysate
Gal_1- Lp_1^+ Lp_{2r}^s	1*	426*
Gal_1- Lp_1 Lp_2	1	2
Gal_2- Lp_1^+ Lp_{2r}^s	20	356
Gal_2- Lp_1 Lp_2	14	14
Gal_4- Lp_1^+ Lp_{2r}^s	89	296
Gal_4- Lp_1 Lp_2	50	57

* Numbers of papillae per plate, 0.1 ml lysate plated
Between 10^8 an 10^9 cells plated.

Table 5

Other Loci tested but not found Transducible

Locus	Number of Experiments	Cultures Involved
Lac_1	4	W112
(ser or glyc)	1	W1678
Leuc	3	W1736, W1436
Methionine	4	58-161, W811, W1821, W518
Xylose	3	W1821
S	1	W518
Pro1	7	W1692, W1920, W2062
Mal_x	1	W2071

Table R6

Action of Lytically Grown lambda in Transduction

Experiment	Culture	No Addition	Lytic Lambda Lysate (2.4 x 10^{10} lambda/ ml)
228	W750	3*	2*
	W 518	9	8
	W2175	7	8
239	W750	2	0
	W518	13	8
	W2175	6	2
254	W750	-	3**
	W518	-	6**
	W2281	-	9**
	W2373	-	6**
	W811	-	39**

* Numbers of papillae per plate, 0.1 ml lysate plated. 10^8-10^9 cells plated.

** these papillae picked and streaked out, all found stable.

Lytically grown λ as follows. W811 galK⁻ λ adsorbed on W1485 galK⁺ in two exposures. Centrifuged and resuspended in NSB. Aerated 4.5 hours with aeration. Control tube consisted of both exposed cells.

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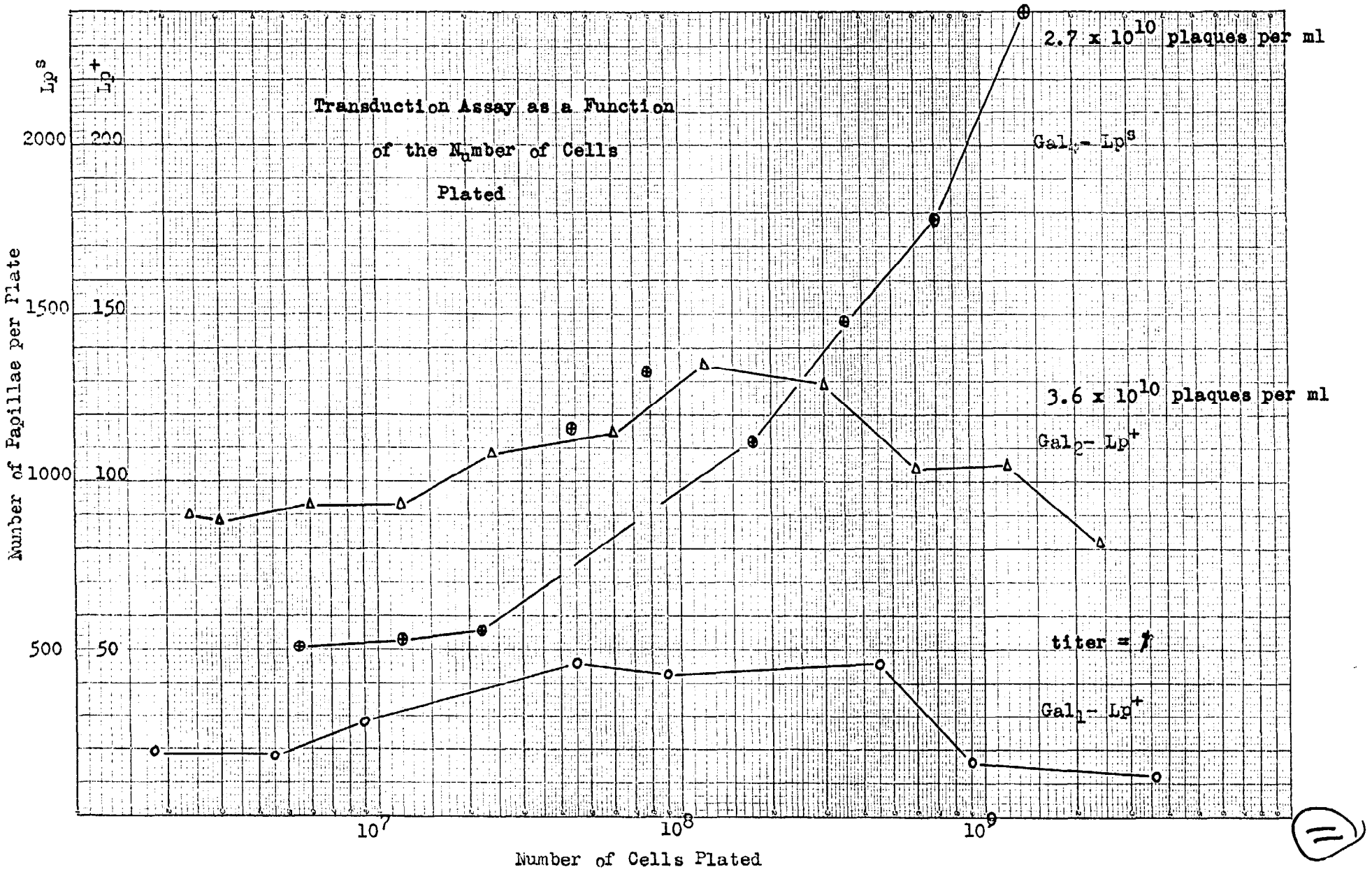

Table 8
Adsorption of the Transducing Activity from Lysates

Adsorbing Cells		Phage Titer X 10^9	Cell Titer X 10^9	Percent Adsorbed 1st Ads. Phage	1st Ads. Trans.	2nd Ads.* Phage	2nd Ads.* Trans.	3rd Ads. Phage	3rd Ads. Trans.
Gal_4- Lp^s	(1)	2.5	0.71	60	79	50	41	16	46
	(2)	3.9	0.55	52	33	-	-	-	-
Gal_4- Lp^+	(1)	14	c. 10	-	79				
	(2)	18	16	-	72	-	56	-	0.0
	(3)	14	c. 10	-	97	-	-	-	-
Gal_2- Lp^+		18	6	-	35	-	33	-	0.0
Gal_1- Lp^+		18	6.5	-	45	-	100	-	0.0

* The supernatant from the first adsorption was decanted and an equivalent volume of fresh lysate added. Similarly for the third adsorption. Assays were made of the amount of material remaining in the supernatants. Titers given are for the phage-lysate adsorbing mixture. In the experiments involving Gal_4- Lp^s assay of the sediment was made in some instances. Total recovery in these cases was more than 100% - presumably due to the fact that total activity was underestimated by the use of too few assay cells.

Some of the papillae evoked by lysate exposure have a property which distinguishes tham at once from spontaneous reversions. That is, they are unstable for galactose fermentation and segregate (-) cells over many single colony transfers. The ~~xxxxx~~ frequency of unstable transductions and the nature of the segregants will be taken up in a later section, it is necessary to mention them now in order to consider the realtionship between the transducing agent and the phage lambda. It is also necessary at this time to mention some special cultures encountered during the analysis of the segregants mentioned above. These special cultures are notable for the fact that they give rise to lysates by the Lwoff technique in which the ratio of transduction activity to lambda plaque forming activity is much closer to unity than is found in the usual cultures of K-12. These cultures will also be considered in a later section and it will suffice to say here that exposure of a population of gal- cells to one of these lysates can result in the transduction of several percent of the cells to gal+.

The data in table 9 indicates that when lamda sensitive cells are transduced the resultant cells and their gal- segregants have for the most part become lysogenized. When Lp_1^r forms are transduced they also may become lysogenized, but much less frequently than sensitives. However, these results may be misleading since the platings involve large quantities of phage and it cannot be certain that lysogenization was not prior or subsequent to transduction. When the transductions are made with the special lysates mentioned above, results such as those shown in table 10 are obtained. Under conditions where one percent of the cells ~~xxx~~ have been transduced to gal+ the transductions have become lysogenized, or Lp_1^r, while the gal- cells in ~~this~~ the same environment have remained lambda sensitive.

Table 9
Correlation of Lysogenization with Transduction

Locus transduced and Lp_1 genotype	Lysate source	Transductions Number	Transductions Percent Lp_1^+	Segregants Number	Segregants Percent Lp_1^+
Gal_1^- Lp_1^s	wild	23	87	1	100
	gal_2^-	24	75	7	100
	gal_2^-	12	58	0	-
	gal_4^-	22	77	9	100
Gal_2^- Lp_1^s	wild	13	85	13	85
	gal_1^-	20	95	20	95
	gal_4^-	23	100	23	100
	wild	18	100	-	-
	wild	-	-	28	50
	gal_1^-	-	-	44	86
	gal_4^-	-	-	40	83
Gal_4^- Lp_1^s	wild	-	-	18	100
	gal_2^-	-	-	19	100
	gal_x^-	-	-	45	100
Gal_4^- Lp_1^r	wild	-	-	29	3.1
	gal_2^-	-	-	18	5.5
Totals		154	86	267	89

Table 10

Correlation of Transduction with Lysogenicity Using Lysates Giving a High Frequency of Transduction

Cells Exposed to	Post Exposure Cell Titer	Number of Colonies Observed			
		Gal-	Gal+	Gal- partially lysed	Total
Broth	4.1×10^9	3280	0	0	3280
HFT lysate*	3.5×10^9	2801	31(1.1%)	54	2886

* titer = 1.2×10^9 λ plaques per ml.

Table 10a

Examination of Colonies after HFT Lysate Exposure

Colony Type	Number of Colonies Examined	Numbers of Colonies of Each Class		
		Lp^s	Lp^+	Lp^r
Gal-	31	31	0	0
Gal+	26	0	23	3

The occurrence of stable transductions among the various combinations of transductions possible is indicated by the data shown in table 11. With but six exceptions the difference between expected and observed fequency of stable gal (+) on the transduction plates is sufficiently great not to require statistical treatment. In setting out this data it has been assumed that the only source of stable (+) on the plates is from spontaneous reversions and that the use of a no lysate addition plate as an indicator of the number of spontaneous reversions is adequate in this sense. It is notable that transductions involving gal_1 and gal_4 are nearly all stable and it will be remembered that lysates of these cultures have less papillae promoting activity upon one another than upon other cultures. These two loci are readily distinguishable by crossing test and by use of the HFT lysates mentioned above. In the other combinations of transductions possible stable transductions occur, varying in frequency from less than one percent to more than 50 percent.

The segregants from the unstable transductions can be classified for locus by three separate methods: (1) by the lysate by which they are <u>not</u> transduced (transduction test), (2) by that locus which cannot be transduced to (+) via a lysate (lysate test), (3) by allelism test in crossing (crossing test). In classifying the segregants it will be convenient to refer to the origin of the locus by specific terms. By homotypic will be designated the locus of the cell transduced to (+), by heterotypic will be designated the (-) locus (if any) of the transducing lysate, and by homo-heterotypic will be designated cultures with the loci of both transduced cell and transducing lysate.

Since the order of segregation from a transduced cell can not be specified without micromanipulative means the analysis of segregants from a single transduction in its absence is without great significance. However, the data in table 12 indicate that a single transduction can give rise to all three types of segregants, homotypic, heterotypic, and homo-heterotypic,

Table 11

The Occurrence of Stable Transductions

Cell Genotype	Numbers of Stable Transductions — Source of Lysate: Wild Type Exp't	Wild Type Obs.	Gal_1- Exp't	Gal_1- Obs.	Gal_2- Exp't	Gal_2- Obs.	Gal_2- Exp't	Gal_2- Obs.	Gal_4- Exp't	Gal_4- Obs.	Stable	total	% Stable
Gal_1- Lp_1^s (2373)	1/33	14	-	-	1/11	11	0/56	20	1/30	29	74 (61)	130	53.8 ~~57~~
(750) Lp_1^+	1/46	2	-	-	-	-	1/92	0	-	-	2 (0)	138	~~1.5~~ 0
(2343) Lp_1^+	1/143	42	-	-	1/84	4	-	-	12/27	27	73 (59)	254	23.3 ~~28.7~~
Gal_2- (2281) Lp_1^s	0/46	15	0/214	27	-	-	-	-	0/98	4	44	358	7.8 ~~12.8~~
(2175) Lp_1^+	17/248	21	14/83	61	-	-	-	-	14/79	52	135 (90)	410	22.0 ~~32.4~~
(1210) Lp_1^+	4/23	6	2/65	0	-	-	-	-	5/56	0	6 (-5)	144	0 ~~4.1~~
Gal_4- Lp_1^s	19/835	383	29/72	72	11/472	20	4/128	21	-	-	496 (433)	1507	28.7 ~~32.9~~
Lp_1^+	41/573	133	51/96	96	-	-	-	-	-	-	229 (137)	669	20.5 ~~35.8~~
Lp_1^r	31/320	127	-	-	31/238	50	-	-	-	-	177 (115)	558	20.6 ~~31.9~~

Exp't = number of stable expected = $\frac{\text{no. papillae control}}{\text{no. papillae lysate plate}}$

Obs. = number of stable observed = $\frac{\text{No. stable observed}}{\text{no. pap. in sample}} \times \frac{\text{no. papillae Transd.}}{\text{no. pap. in sample}}$

Note: A number of different lysates were employed. In the case of Gal_2- lysates, the first column represents lysates of W902, the second column, W1210. In the case of the Gal_1- Lp_1^+ cells, the first is W750, the second W2343, a prototroph derived from W750

Table 12
Segregants from a Single Transduction, tested by Transduction Test.
The sequential order of the segregants is unknown

Cell Genotype	Lysate Source	Classification of Segregants		
		Homotypic	Heterotypic	Homo-heterotypic
Gal_4-	Gal_2-	17	2	1

but it can not be stated that the segregations occurred in any sequence or if sequential. The analysis of single segregants from a large number of transductions was undertaken to clarify this process. In the initial experiments the transduction tests were performed by mixing a portion of lysate from a culture of specific locus and the culture to be tested upon EMB galactose medium, but after the discovery of the HFT lysates test for allele was by cross brush with lysates of this property upon the same medium.

The results ~~of a large number~~ of tests of segregants by transduction test is given in table 13 and a summary of the cultures in this table which were also tested by lysate test is given in table 14. The agreement between the two tests was complete, that is, a culture classified by the first method as gal_4- was also classified ~~by~~ as this locus by the second test and the. A summary of the segregants which were tested by all three methods of determination is given in table 15. Agreement between the crossing test and the other two tests was also complete. Some indication of the distribution of the segregant types, as judged by transduction test, can be obtained from the distribution data given in table 16.

With regard to the crossing data given in table 15 it will be noted that no crossing data for gal_1- segregants ~~is~~ are reported or crosses of heterotypic segregants from gal_1 transductions by gal_1- testers. ~~This is~~ because a suitable stock is not yet available. W2373, a $hist^-$ $leuc^-$ gal_1- made by transducing W1765 to gal_1- has not been found sufficiently fertile in crosses with $Meth^-$ stocks to warrant its use. A new $T^-L^-B_1^-$ (also Het) gal_1- also made by transduction to (-) may prove suitable. It should be noted that the number of prototrophic recombinants given in table 15 is probably low by as much as 25 percent since in many instances only the ~~smallest~~ plates with the smallest number of prototrophic recombinants were counted in ~~many~~ experiments involving many replicate plates.

Table 13

Analysis of Segregants by Transduction Assay. Summary.

Nature of Original Transduction	Type of Segregant: Homotypic*	Heterotypic**	Homo-heterotypic***	Total
Wild type on Gal-	169	0	0	169
Gal- on Gal-	240(85.4)	37(13.2)	4(1.4)	281
	~~409(91.0)~~ 407	37~~(8.2)~~	4~~(0.88)~~	450

* having the Gal- locus of the transinduced cell
** having the Gal- locus of the transducing lysate
*** having the Gal- loci of both transinduced cell and transducing lysate.

Table 14

Analysis of Segregants by Lysate Test. Summary. Agreement between Lysate Tests and Transduction Tests was Complete

Nature of the Original Transduction	Homotypic	Heterotypic	Homo-heterotypic	Total
Wild type on Gal-	21	0	0	21
Gal- on Gal-	39	~~16~~ 19	0	58
	60	~~16~~ 19	0	79

Table 15

Summary of the Analysis of Segregants by Transduction test, Lysate test and by Crossing test.

Original Transduction	Number of Segregants	Transduction test	Lysate test	Classification by Crossing test: Homotypic No. (+)	Homotypic Tot. Prot.	Heterotypic No. (+)	Heterotypic Tot. Prot.
Gal_2^- --x Gal_4-Lp^s	5 (1)	Gal_4^-	Gal_4^-	0	2786	3	3183
	(2)	"	"	0	2675	2	3471
	(3)	"	"	0	3485	23	5342
	(4)	"	"	0	5952	1	1665
	(5)	"	"	0	5000	1	891
	2 (1)	Gal_2^-	Gal_2^-	7	3102	0	1988
	(2)	"	"	10	4364	0	1187
Gal_2^- --x Gal_4-Lp^+	4 (1)	Gal_4^-	Gal_4^-	0	16104	3	1389
	(2)	"	"	0	5730	1	164
	(3)	"	"	0	3358	0	202
	(4)	"	"	0	12848	1	171
HFT	3 (1)	Gal_2^-	Gal_2^-	1	11200	0	827
	(2)	"	"	6	10608	0	718
	(3)	"	"	3	5000	0	409
Wild --x Gal_2-Lp^s	4 (1)	Gal_2^-	Gal_2^-	0	7805		
	(2)	"	"	0	4992		
	(3)	"	"	0	106		
	(4)	"	"	0	4552		
Wild --x Gal_2-Lp^+	4 (1)	Gal_2^-	Gal_2^-	0	4070		
	(2)	"	"	0	5384		
	(3)	"	"	0	2072		
	(4)	"	"	0	6988		
Wild --x Gal_4-Lp^s	4 (1)	Gal_4^-	Gal_4^-	0	896		
	(2)	"	"	0	918		
	(3)	"	"	0	1134		
	(4)	"	"	0	863		

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Table ~~II~~

Distribution of the Segregant Types by Transduction Assay

Transinduced cell		Source of Lysate: Wild type	Gal_1-	Gal_2-(W902)	Gal_2-(W1210)	Gal_4-
Gal_1-	Lp_1^+(W2343)	18 Gal_1-	-	18 Gal_1-, 5 Gal_2-	-	no seg. found
	Lp_1^+(W750)	16 Gal_1-	-	18 Gal_1-, 1 Gal_2-	18 Gal_1-, 3 Gal_2-	no seg.
	Lp_1^s	9 Gal_1-	-	1 Gal_1-	6 Gal_1-, 1 Gal_2-	1 Gal_1-
Gal_2-	Lp_1^+(W2175)	20 Gal_2-	14 Gal_2- 3 Gal_1- 2 Gal_1-Gal_2-	-	-	8 Gal_2- 7 Gal_4-
	Lp_1^+(W1210)	15 Gal_2-	19 Gal_2- 2 Gal_1-	-	-	
	Lp_1^s	16 Gal_2-	20 Gal_2-	-	-	21 Gal_2- 1 Gal_4- 1 Gal_2-Gal_4-
Gal_4-	Lp_1^+	20 Gal_4-	nsf	16 Gal_4- 3 Gal_2-	-	-
	Lp_1^s	13 Gal_4-	nsf	18 Gal_4- 3 Gal_2-	17 Gal_4- 2 Gal_2-	-
	Lp_1^r	29 Gal_4-	nsf	15 Gal_4- 3 Gal_2-	-	-

nsf = no segregants found

Cultures giving lysates with the HFT property have been prepared for each of the gal- loci which have been given consideration to date. These cultures have the common property that each is derived from the transduction of a gal- culture by a lysate of gal_2-. It is not known whether the transductions themselves of this type are capable of giving rise to HFT lysates or not, but the HFT stocks thus far obtained have been segregants from such transductions. Whether the transductions of gal_2- by other cultures gives rise to HFT segregants is not known, but one instance in which the transduction of gal_2-by gal_1- resulted in an unstable (+) which had HFT property has been encountered. Some idea of the frequency with which the HFT occur can be obtained from the following. In the case of transductions of gal_1- by gal_2-, out of 28 gal_1- segregants examined 4 had this property and of the heterotypic gal_2-, one out of five examined was HFT. In the case of transductions of gal_4- by gal_2-, of 31 gal_4- segregants tested one was HFT, while of the three heterotypic gal_2- tested one was HFT. In the above tests segregants which had been purified through several single colony isolations were used. Since the HFT cultures segregate NFT lines it is possible thatthe above estimations are low.

Attempts to obtain (+) cultures with HFT property by reversion of (-) have been unsuccessful in the limited attempts made thus far. This too may be in part due to the fact that the HFT cultures segregate NFT lines and since it was not known at the time of examination that this was the case the NFT reversions obtained could well have been from NFT components of the culture. The conversions of a HFT culture to NFT is fairly rapid and the HFT cultures are easily lost. On one occasion it was noted that a culture which had been on stock slant only a few weeks had changed such that of ten colonies tested 4 were found to be NFT. The NFT cultures which are derived from HFT lines have not been investigated except in the instances. In addition to NFT property (or possibly no activity at all) the segregants were in the one case of a gal- type which was not transducible except by a lysate of an unstable gal(+).

In one case (and the negative results in the other cases can possibly be explained

by the contamination of the HFT cultures with NFT cells) gal (+) reversions of an HFT culture were found to be unstable for gal(+) and segregated (-) which were of the same (-) type as the reverted locus. The examination of more HFT cultures to to determine the relationship between duplication of certain loci and HFT property is in progress.

The lysates of the HFT stocks which have been prepared thus far have not had high phage titers although they have been prepared in a manner which gives high titered lysates in NFT stocks. Whether this indicates a lower yield per bacterium of plaque forming particles or different conditions for induction is not known at the present. Preliminary experiments to determine the yield of HFT particles per bacterium are regarded with reservation since the purity of the culture with regard to NFT cells was not known.

The HFT lysates have been used principally for allelism tests. Transductions can also be made via these lysates and the resultants studied. This has not been carried very far. The data in table 10 indicate that transductions by HFT lysates are not appreciably different from those of NFT lysates as regards occurrence of stable tramsductions and distribution of segregants.

The HFT lysates can be used for transduction from gal(+) to gal(-) and have proved of value in creating new stocks. Table 10 lists some of the information available on the stocks transduced to (-). Since the completion of the table gal_1- and gal_2- Lp^s $T^-L^-B_1^-$ Het_Λ stocks have been prepared. The (-) stocks prepared thus far have been made starting with Lp^s cultures. The resultant cultures may be Lp^s, Lp^+ or Lp^r. In general the procedure has been to mix HFT lysate and cells on EMB(0) and incubate for 12-18 hours and then to streak out the growth and search for gal (-). On other occasions examination of single colonies from cell populations exposed to HFT lysate has been used.

Table 1b
Transduction by HFT Lysates. Distribution of the Segregants by Transduction Assay

Transinduced Cell Genotype	HFT Lysate Gal_1-	Gal_2-	Gal_4-
Gal_1- Lp_1^+	-	10* Gal_1- 2 Gal_2- 1 Gal_1- Gal_2-	9 Gal_1-
Gal_2- Lp_1^+	6 Gal_1- 3 Gal_2- 1 Gal_2- Gal_1-	-	8 Gal_2- 4 Gal_4-
Gal_4- Lp_1^+	not done	15 Gal_4-	-

* Out of a total of 18 transductions (or transductions and spontaneous papillae) analyzed. The difference between the number of segregants reported and 18 represents the number of stable papillae observed.

Table 17
Transductions to Inability to Ferment Galactose

Culture Transduced	Lp_1 Genotype	Galactose Locus Transduced	Resultant Lp_1 Genotype	Comment
W1485	Lp^s	Gal_2-	Lp^+ or r	8 distinct (-) obtained from single colonies
			?	2 distinct (-) obtained
		Gal_1-	+ and r	2 distinct (-) obtained
W1673	Lp^s	Gal_2-	+ or r	-
W1765	Lp^s	Gal_1-	s	-
W2252	Lp^s	Gal_1-	r ?	2 distinct (-) obtained
		Gal_2-	r	2 distinct (-) obtained