

3/6/53 Friday

Exp J Examination of prototypus from cross of 578 x 892
 Prototypus Strained 811A-5 8421-2 LP Rx medical cultures

1. } gult	1 st smear	2 nd	LP Rx	Stability
2. }				
3. }	52	18	S. smear	4-
4. }	20	153	S. smear	?
5. }	24* 5	42*	Solid smear	4-
6. }	72	347	S. smear	?
7. }	44	227	S. smear	?
8. }	1	6	S. smear	4-
9. }	40	21	S. smear	4-
10. }	41	36	S. smear	4-
11. }	37	11	S. smear	4-
12. }	23	21	29	LP ^R
13. }	1	8	S. smear	4-
14. }	39	10	S. smear	4-
15. }	2	61	S. smear	?
16. }	77*	211	S. smear	?
17. }	33	60	S. smear	4-
18. }	23	208	S. smear	?
19. }	2	11	S. smear	4-
20. }	7	7	S. smear	4-
21. }	4	9	14	LP ^R
22. }	1	17	S. smear	4-
23. }	30*	35*	46*	LP ^R
24. }	87	12	S. smear	4-
25. }	30*	18	S. smear	4-

Could be from smears

* small in bkgrd.

14 4- 6/14 S
 6 ?
 3 LP^R

medical cultures

3/6/53

Transducers

811	EM13 fuel	N. pop 3 days
1.	no add	43
2.	B8	122
3.	D4E750	42
4.	A3	47

2050	1. no add	16 (+ some small)
	2. B8	1/4 = 110
	3. D4E750	1/2 = 210
	4. A3	1/4 = 177

750	1. no add	2
	2. B8	78
	3. D4E750	2
	4. A3	18

2175	1. no add	24
	2. B8	18
	3. D4E750	12
	4. A3	64

Tuesday 3/10/53

STF Transduction with high activity ~~of~~ phage.

0.1 ml of saline resuspended STF cells (overnight culture)
 0.9 ml of DI lysate - 1 - one at 37 1.0 min - dil

10², 2x10¹ ^{actual} → 10 plates.

	unstable (+)	(-)	total
1.	72	1708	1780
2.	83	1/4 = 493 (172)	1855
3.	77	about the same	
4.	83	" " " "	
5.	89	" " " "	
6.	95	" " " "	

24 of these (+) streaked out - all mixed - many mosaic col.

$$1708 \overline{) 1780} \\ \underline{682} \\ 1098 \\ \underline{682} \\ 416$$

c 4% of cells transduced

remainders about the same

6 plates of STF untreated dil to contain c. 150 colony / plate showed no phage colonies

1 plate of undiluted STF also showed no (+)

Extended Examination of prototypic form STF 892 mixed culture. Step J.

J 4. no add

2.	750A-2	22
3.	902A-2	1/8 = 66 - 528
4.	892A-2	Solid smear
5.	811A-5	1/8 = 70 = 560

} says it's probably got - ?

J 6

2.	750A-2	c. 50
3.	902A-2	1/8 = 73 = 584
4.	892A-2	1/8 = 87 = 696
5.	811A-5	Solid smear
		1/8 = 68 = 534

} says it's a new locus

J 7

2.	750A-2	21
3.	902A-2	1/8 = 57 = 456
4.	892A-2	1/4 = 107 = 856
5.	811A-5	Solid smear
		1/8 = 72 = 576

} says it's a new locus

3/10/53

1655 (-) gal house dust = 892
0.1 ml 1655 saline suspended (aerated ant) + 0.9 ml 892-2
- incub at 37C 10 min dil 10¹, 10² → 0.1 ml / 10 plates EMB gal

	cf. no. colonies	no. mosaic col.	
1.	155	5	3.2% mosaic
2.	294	8	2.7%
3.	not about the same		

2 plate made to indicate the amt. of phage added to the gal plates. indicate about 10⁷ 10⁸ / ml

Lysate sterilities

- 892-2 bottle 1 - sterile 0.5 ml
- D1-1 0.2 ml sterile
- D4-2 bottle 1 sterile 0.5 ml
- D1-2 ~~bottle 1~~ not sterile 0.5 ml. - Re chloroform

1655 ^{about 27} mosaic colonies picked and streaked out - no sign of colonies
evidently E1 ductum not easily accomplished

3/17/53 Tuesday.

Transductions

To identify proto types from 518 X 892

750

EMBP	Rep 2 days
1. no add	4
2. J4	6
3. J7	3

2175

1. no add	8
2. J4	26
3. J7	30

2050

1. no add	$\frac{1}{4} = 56 = 224$
2. J4	$\frac{1}{4} = 58 = 232$
3. J7	$\frac{1}{4} = 41 = 164$

4. 892A-2 (1-100)	> 2000
5. D2 (prominent 4- for 518 X 892)	$\frac{1}{4} = 83 \quad 332$

518

1. no add	33
2. J4	18
3. J7	23
4. D2	15

J4 } appear to transduce
J7 } gene - only what
this means I don't know
J4, J7

probably says D2 is gal⁻ - other ductin
of 2050 is low (hi ok prod?) also this
ductin is lysozyme for "normal" - not
hi set 1

1485 - Attempt to transduce \bar{c} hi set 1 of 892 - Prep 2

0.2 ml of mixed
culture c. 10^9
+ 0.1 ml 892A-2.

5 Control plates
EMB fob c. 400 colonies/plate - some intermediate gal⁺
& possible mosaic colonies - streaked out.
on streaking showed
+ and abt forms.

5 EMB plates
EMB fob c. 80 colonies/plate - represents about 80% killing?
c. 24 mosaic colonies - streaked out
 $\frac{0.04}{400/25} = c. 4\%$ of the 24 colonies - 1 segregating (+) on 1st
streak.

to avoid
selection agents
(-)
5 EMB plates
EMB(-)

c. 80 colonies/plate -
31 colonies showing evidence of lys

3/21/53 Sat.

involved - 892 a mud

Exp K 811 & 892 (-1 segregant analysis)

On EM13 gal

Seq #	Flu add	1-100 det. 892 A-2	811 A-5	Probable locus
1.	31	c. 2000	33	4
2.	21	c. 3000	54	⊙ 1/8 mixed 2 1/8 mixed 3
3.	71	c. 2000	68	4
4.	37	c. 2000	33 63	⊙ 1/8 mixed 2 1/8 mixed 3
5.	26	c. 2000	28	4
6.	26	c. 2000	29	4
7.	17	c. 1000	59	⊙ 4/8 mixed 2 1/8 mixed 3
8.	37	c. 2000	32	4
9.	36	c. 1000	73	⊙
10.	27	c. 1500	31	4
11.	32	c. 1500	48	4
12.	28	c. 1500	33	4
13.	24	c. 1500	22	4
14.	24	c. 500	54	⊙ 4/8 mixed 2 1/8 mixed 3
15.	12	c. 1000	45	⊙ 5/8 mixed 2 5/8 mixed 3
16.	33	c. 1500	19	4
17.	47	c. 2000	43	4
18.	28	c. 2000	21	4

switched

892

1 on the stability of spontaneous pop - after 2 all appear pure +

Exp D

instability of D1 spontaneous pop. confirmed (again) 3/8
 stability of D4 " " " " 8/8

2050 - from EM1 - transferred by 892 A - 2 to keep ^{frequency} titer

892 - from EM1 - ~~the~~ loc - colony distribution about the same from as over 892

- on a chr loc - small col form + spontaneous colony form

Sun
Mon: 3/22/53 - 3/23/53

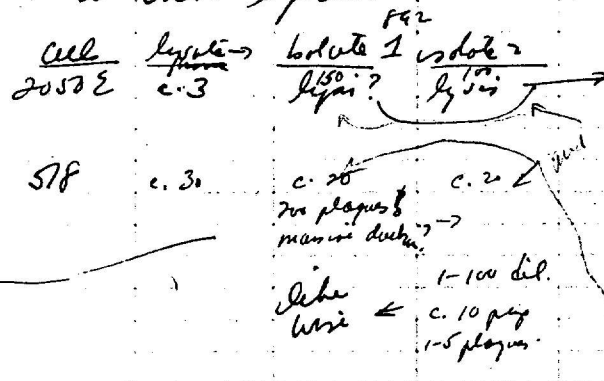
518 Perfect stage / Transduction as a function of multiplicity

2050 Euc -

EM13
892 - streaked out lue - appears homogeneous lue. main - 2 colony types - small round, large spreading. well isolated of each yellow, by outis made
892 - void / 892 - isolates } don't know which is which! det'm. from culture

2050 Euc is those lysates:
518

Abundant
not abundant



c. 200 plaque plus from lysate - transduction deficient to one - massive? streak and see extent of jul + near
no transduction at jul (-) in 2 different areas

Results suggest cool 1 + 2 at jul 4 - ?

518 + 2050 - 3 (purified) stocks made of separate transductions
not a high incidence of transduction 2050 - x 518

3/24/53
E4 + K stored in EM13 gel plate

518 - one plate - transduced E 892 hi purer & to see if transduced one MT replica (slightly) to D(0) - no growth D(0) - in prototypic

3/16/53
Thursday

892 - all confused - probably complicated by a switch in media.
Apparently 892 mixed with gal⁺, gal⁻, possibly (-) of
a couple of types.

578 Examination of D4 to see hi activity of d present.
1. to add = 19
2. 04 d = 7109

Summary 518K-12 See pages 170a

page 1. Examination of a single gal⁻ segregant from ~~2~~ separate transductions
of W518 gal⁻ by lysates of K-12

170a	A.	<u>Receptor test</u> No. of trans. trans. by gal ⁻ lysates	22	0	by <u>No. trans. by gal⁻ lysates</u>	22	<u>Probable locus by receptor test</u>	22 gal ⁻
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- 1. (13)
- 2. 41
- 3. 42
- 4. 43

2. Examination of a number of gal⁻ segregants from a single transduction

145 A. Receptor test

No. of seg.	No. trans. by gal ⁻ lysates	No. trans. by gal ⁻ lysates	Probable locus by receptor test
15	0	15	15 gal ⁻

163 B. Conjugation test (X1426)

Seq. no.	No. prototrophs	No. (+)	Probable locus
A3	975	0	gal ⁻
A11	1048	0	gal ⁻
A13	c. 1250	0	gal ⁻

C. Donor Test

Lysate of seg.	gal ⁻	Transducer gal ⁻	gal ⁻	gal ⁻	Probable locus
A1	+	+	+	no	gal ⁻
A3	+	+	+	no	gal ⁻

165, 1766

Summary of 518E902

page

7. Examination of ~~a series~~ single gal-segregants from different transduction.

2. Examination of a series of gal-segregants from a single transduction

A. No of Seg.	No transd. by gal ₂ by gal ₃ but not by gal ₁	No transd. by gal ₁ - gal ₂ but not gal ₃	Possible locus
146	29	17	2 gal ₂ - , 17 gal ₃ -

B. Crossing test

1. Seg.	gal ₂ -	by receptor test (x 902)	No prototrophs	No. (+)	Possible locus
175	B7		c. 2200	0	gal ₂ -
	B8		c. 1600	0	gal ₃ -

C. Down Test

Hyate of Seg	gal ₁ -	Transduces			Possible locus
		gal ₂ -	gal ₃ -	gal ₁ -	
176B	B7				
	B8	+	no	+	gal ₃ -

76
16
446
766
1216

Sunday 3/29/53

811 Analysis of 892 mixture

892-1 } = 2 (-) got colonies from stocks of 892 - lysate made
892-2 } of them - both are loc - TLR-

811		Age 2 days
1. no add	28	
2. 892-1	39	
3. 892-2	33	

} these lysates appear to have at least 2 phages in them - 1 resembling A, other giving plaques on A+ cultures!

2175 for the purpose of separating, reagent of 578+902

1. no add	- 6	
2. K-12 30-1	176	- pick 24 for analysis

750 For the purpose of picking transductions for analysis - of segregants.

Monday 3/30/53

1. no add	2	
2. K-12 30-1	1/8 = 18	342
3. 902 A-3	1/4 = 29	116

518 For the same purpose as 750 above

1. no add	28	
2. 750-2	79	
3. 902 A-3	1/16 = 76 = 1216	
4. K-12 30-1	1/8 = 65 = 280	

5. plate of mixture of 1485 E.O. } 57 ← indicates that if A from 'natural' lysate is not very active - assuming high titres & few plaques resembling A-2

578 a second attempt to test a lysate of soil from mixture 892 7 day incub

1. no add	0	20
2. lysate of 892 soil	> 104	high activity
along with broth culture of this appeared		

Monday, March 30, 1953

1673X D1 to see if ^(H) from this cross have hi-act lambda-purity of the cross.
 D1- ^{pure -}
 1672- +, with slow gel present - a plus picked for purif.
 And a new

4/8/53 Thursday

- 1485tD1 - the negative transduction - Examination of (-) segregants to determine their nature.

Exp L.

		no sup 2 days	Locus suggested
all these found Apr 5 and prototypic	L1	no add 9021-3 811A-6	4 19 37 2-
	L2	no add 9021-3 811A-6	6 5 21 2-
	L3	no add 9021-3 811A-6	10 9 22 2-
	L4	no add 9021-3 811A-6	5 5 21 2-
	L5	no add 9021-3 811A-6	5 8 13 2
	L6	no add 9021-3 811A-6	3 7 21 2-
	L7	no add 9021-3 811A-6	3 8 14 2-
	L8	no add 9021-3 811A-6	2 3 22 2-

Reason for low transduction not known.
 Data appear ok. but better recheck.

892 hi powered A - to see how DNA are susceptible?

578	no add	16	
5	892) untreated (-100)	>104	no
	892) DNAase treated at 37c 10min 1-100 x 6/5	>104	

4/2/53

518 gal⁺ - is there a diff. between 518 gal⁺ and 811 gal⁺ because 811 was made 4⁺ + 10⁺? Data on 811 reverses as gal⁺ does not clear because strains not transducing ph⁺ do not give (+) in crosses with Y-10. Examine 518 gal⁺ to find strains giving (-) with Y-10, then make lysogenic and examine transducing phage.

Check in the crossing cultures. Y-10 all (+)
518 gal⁺-1 " "
518 gal⁺-2 " "

518 gal⁺-1 x Y-10 - all + protoph. found
nos/plate = 65, 48, 75, 114, 67, =

518 gal⁺-2 x Y-10 - all + protoph. found
nos/1/4 plate = 75, 128, 95, 87, 108

842 mixed stock - plated against A² - large proportion of cells 4⁺ 2^R
Selection for 4⁺ portion of stock.

2175 CK-12
24 (-) say obtained - carried thru ③ purifications

518C700-1 } made from ② purification
518C700-2 }
750C902-1 } made from ② purification
750C902-2 }
750C12-12-2 } made from ② purif.
750C11-11-1 }

April 5, 1953

892 (mix) preliminary examination of occurrence of transducing phage in bursts from single cells of irradiated 892

1. growing culture of 892 diluted to contain c. 500 cells/0.1 ml
2. irradiated 15 seconds.
3. plated before and after with 750, 1485, and by itself to

- 7. 892 cells / 0.1 ml before irrad. = 585 (gal + = 101) 5
- 2. 892 cells - plaques / 0.1 ml on 1485 before = 1
- 3. 892 cells - transd. / 0.1 ml before on 750 = 78 (gal + from 892, 78)
- 4. 892 cells / 0.1 ml after irrad. = 16.8 (gal + = 26)
- 5. 892 cells / 0.1 ml plaques on 1485 after irrad. = ~~247~~ 415 (gal + = 415)
- 6. in 892 cells 9' on 750 cells = 35 (gal + from 892 = 31) / no effective particles

750 no add = 2

58 gal + #3 X Y-10 controls o.k.

7. 1/4 = 60	110	no. (-)	
2. 1/4 = 33	192	"	
2. 1/4 = 46	184	"	
4. 1/4 = 40	160	"	
5. 1/4 = 51	204	"	
	930		0(-) / 930 (+)

58 gal + #4 X Y-10 controls o.k.

7. 1/4 = 53	212	more	
2. 1/4 = 57	228	"	
2. 1/4 = 50	200	"	
4. 1/4 = 58	232	"	
5. 1/4 = 55	220	"	
	1114		0(-) / 1114 (+)

1485 EDI - unstable gal + → lyses his ant. |
 gave rise to stable + → low out |

902⁵⁴ X 2175 (gal₂ - x gal₂ -) on low sm - about 200 per plate.

2251

Tuesday 4/7/53

- 1821 - w: hi poured 892)

On EMB/Pl

- 1. no abd - c10
- 2. 892/ 1700 - c. 1000?

} appears that hi act 1 has less act. on 1821 - situation similar to "nause" 1.

On EMB xgf

- 1. no abd - many small 3-4 large
- 2. 892/ - 1100 - " " c10 " }

Repeat xgf using undiluted 1892.

1485T D1 - Re-examination of the possibility of (-) ductin.

1. Lysoate D1d - 2 pur.
2. Cross pushed on 1485 on EMB/10.
3. After 1 day lysed area picked by HAH - streaked out several times on EMB/10.
4. a single (-) colony observed. several intermediate gdt + col. a few aggregating (w/det?) colonies. many gdt + colonies.
5. some of above colonies picked and streaked.
6. Results:
 - a. 2 pure (-) col.
 - b. several mixed?
 - c. mostly gdt + col.

Summary Stock culture in Plates in Refrigerator

-	1924E K-12	Unlabeled +	11 in all
-	811 gal + R.		10 in all
-	578 gal + R.		8 in all
Exp	A	578E K-12	all reg single transductant
"	B	578E K-12	all reg from single transductant
"	C	578E 892 (mix)	
"	D	578E 892 (mix)	
"	E	2050E 811	
-			
-			
-			
Exp	J		
"	K		
"	L		
"	M	750E K-12	(-) reg ¹⁷ 18 in all
"	N	750E 902	(-) reg 23 16 in all
"	O	2175E K-12	(-) reg 21 in all
"	P	578E 750	(-) reg 3 in all
"	Q	578E K-12	(-) reg 13 in all
"	R	578E 902	(-) reg 23 in all
"	S	811E 902	(-) reg 19 in all
"	T	2175E 750	(-) reg 23
"	U	2175E 811	(-) reg 16 in all
"	V	811E K-12	(-) reg 20 in all
"	W	1924E 902	(-) reg 18 " "
"	X	1924E K-12	(-) reg 24 " "

replaced
using stock
just -
switched to
E-750 & V
E-902 + 2257

4/9/53

1485T01 - Occurrence of hi-act 1 in bursts of single cells mod

Pre
viad.

1. 1485T01 cell count pre viad. = 493 / 0.1 ml

2. Spurt pop. 750 1

3. No. pop in mix of 1 + 2. EMBgal
Spiked 494

4. Plaque by 1485T01 / 1485 Found 494
0

Post
mod.

5. No. 1485T01 cells surv. 124 ³

6. No. " cells giving plaque / 1485 140

7. No. " cells giving pop. 750 123

" Sp. pop 750 1

Total 124

8. No. pop mod. 1485T01 in 750 222

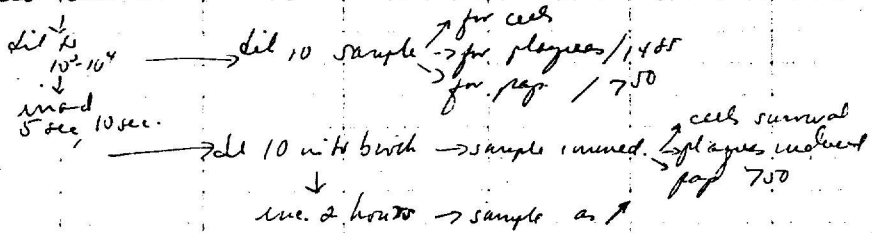
No. of lysing 1485T01 cells
giving pop 750 202 - 124 = 98

Fraction of lysing 1485T01 cells
giving rise to 1 hi-act.
λ particle 98/140 = 70%

4/10/53 Friday

750 and D1 Occurrence hi-act-1 in single bursts of D1

C. 10^3 - 10^4 cells of D1 - sampled before inoc. then inoc. \pm u.v. sampled immediately and after a hour inc in broth inoculated cont. D1



Reprod

	10.1 ml
1. Total no. cells =	310
2. plaques /1485 =	1
3. pop. /750 (3)	6

750 appop = 1.

Post Inoc.

5 second exposure

4. Surviving cells	159	40% plaques 15% discrepancy cells
5. plaques inoculated /1485	40	
6. pop. /750 (3)	6	
7. plaques after 2 hrs /1485	3344	
8. pop /750 after 2 hrs. - (2)	>47 (many small) upper limit = c. 600.	

Plaque assays
against 1485
when streaked

10 sec. exposure

9. Surviving cells	84
10. plaques inoculated /1485	40
11. pop. /750 (3)	15
12. plaques after 2 hrs /1485	2720
13. pop. /750 after 2 hrs	>53 (many small) upper limit = c. 200

out-cults of
plaque show
evidence of
(-) ductin

572 mix gult + (also unstable)
lyse has hi act /578

1485 TDI - (-) neg

lyse probably has hi act /578 but appears quite like also.

578 gult R. #5 and #6 tested in crosses X Y-10 for
minic revision.

578 gult #5 gult parts = 420, 650, 522, 416, 266 (all obtained by counting either $\frac{1}{4}$ or $\frac{1}{8}$ of plate)
< 0/23071

578 gult #6 gult parts = 440, 440, 432, 440, 340, 672 (also obtained by counting $\frac{1}{4}$, $\frac{1}{8}$ of plate)
< 0/2764

3/10/53
Sat.

Various transductions to secure (-) seg for classification

		seg 2 days
2175	1. no add	13
	2. 811A-4	39
	3. 811A-7	50

} (-) seg isolation made from both - discarded

~~~~~

|     |            |    |
|-----|------------|----|
| 750 | 1. no add. | 2  |
|     | 2. 811A-4  | 10 |
|     | 3. 811A-5  | 14 |
|     | 4. 811A-6  | 33 |
|     | 5. 811A-7  | 25 |

} discard - transduce gal<sup>-</sup> prot. mixed and use 750<sup>+</sup> as crossing tool

~~~~~

518	1. no add.	29
	2. 750A-2	72

~~~~~

1821 On EMB xyl -

|                                      |                       |        |
|--------------------------------------|-----------------------|--------|
| 1821                                 | 1. no add             | 3      |
| gal <sup>-</sup> ductin <sup>o</sup> | 2. 8921-2 (hi powder) | c. 150 |

in ✓ these are gal<sup>+</sup> xyl - Apparently gal<sup>-</sup> can select for gal<sup>-</sup> ductin -

~~~~~

3/16/53 Thursday

750^{sr} = ²³¹⁹ - test for validity

	1. no add	21
	2. 750A-1	25
	3. 902A-3	14707 428

4/16/53

750 pr = gal₁, prototroph - Isolation of (-) segregants

1. no add	1.
2. 750A-1	0
3. K-12A 30-1	143
4. 902A-3	84
5. 811-6	#

(check on authenticity)

after 2 single colony isolations 2/4 are crossed - probably indicates that stable transductions result 811 -x 750

gal₂⁻ 1655 x 2238 (= loc₂⁻ gal₂⁻) to isolate tp₂^s from (tp^s also if possible)

Control	1. 1655	pure +
plating	2. 2238	pure -
	3. (4)	total pure
	30	220
avg	4. 33	211
freq	5. 44	433
	107	864

$$864 \overline{) 1107.0} \\ \underline{864} \\ 2430 \\ \underline{2430} \\ 0$$

f = 12.4%

- Pickings for tp^s, tp₂^s or both
- 5/34 tp₂^s all tp^s
 - 30/30 tp^s or tp₂^s / 1

750 pr (gal₁⁻ prototroph) - Inoculation to obtain tp^s form

5 pink colonies on EMB loc noted. Pick and determine

none

1821 purifier - try consistency of single colony isolate - still more of this

1. no add	10
2. K-12A (30-1)	84

148STD1 - 2 additional segregants tested

Seq #9	1. no add	10
	2. 750A-1	415 = 60
	3. 902A-3	10
	4. 811A-7	29

} apparently gal₂⁻

Seq #10	1. no add	8
	2. 750A-1	1/16 = 54 = 864
	3. 902A-3	24
	4. 811A-7	1/3 = 55 = 1750

} clearly possibly gal₂⁻

Saturday 4/14/53

on the job, secondary structure?

2175 - (attempt to observe "satellite" phenomena in 2175, failed - check to see if other transduction

- 1. no add 17
- 2. K-14 3-7 248

Elimination of some lysals

578

- 1. no add 11
- 2. 872-4 (aunt) 18
- 3. 892 M (aunt) 11
- 4. 81/1-8 11
- 5. 902-1-4 1/4 = 110 472
- 6. 015 1/16 = 52 896
- 7. 844 1/16 = 44 704

unknown (see 271.0) →

W234~~2~~3

192 A

4/25/53 Saturday

(tested by spotting 0.05 ml of prep in plate - 1/2 plate rows also control) ^{Supports} _{cell lines of single}

1/2 control

Expt. M 750 prototrophic gal, - transduced by K-12 30-1 - Examination of Segregants

Segregant	No. of Spots (4 plates)	No. pop. 750-2 spot	No. pop. 902-4 spot	Possible focus	To be taken by X	Satellite of gal. d
1.	7	0	6	gal,-		0
2.	2	0	70	"	✓	0
3.	4	0	9	"		0
4.	0	0	77	"		0
5.	0	0	11	"		0
6.	7	0	7	"		0
7.	0	0	9	"		0
8.	0	0	6	"		0
9.	0	0	8	"		0
10.	0	1	10	"		0
11.	0	0	13	"		0
12.	2	7	9	"		0
13.	0	0	13	"		0
14.	1	0	8	"	✓	0
15.	0	0	10	"	✓	0
16.	0	0	15	"		+
17.	0	0	13	"	✓	+
						2/17

2/24/54

Stocks of this experiment discarded as not working of saving

Examination of lysates M2, M14, M15, M17

	750	578	2175
1. no outd	1	70	14
2. M2	3 ^{ok}	29	50
3. M14	5 ^{ok}	62	70
4. M15	4 ^{ok}	15	33
5. M17	5 ^{ok}	28	74

Test crosses of M2, M14, M15, M17 x 750⁺ - Control ^{ETB gal} 750 M14 M15 M17 ok, M2 no growth

Strain	Control	2nd cross	Total	3rd cross
M2 O(R) ch	128, 89, 115	0 ⁺ /332	2, 2, 1, 3 = 11	0 ⁺ /343
M14 O(?) ch	121, 126, 136	0 ⁺ /383	0, 3, 2, 4 = 9	0 ⁺ /392
M15 d(?) ch	210, 130, 171	0 ⁺ /511	9, 6, 2, 1 = 18	0 ⁺ /529
M17 O(?) ch	87, 181	0 ⁺ /268	0, 4, 8, 9 = 21	0 ⁺ /289

ch = check

SUMMARY

- 17 segregants
- by trans. test 17/17 gal,-
- by donor test 4/4 gal,-
- by cross test 4/4 gal,- with following his. prototrophs (332, 383, 511, 268)

Some 4 with missing

3
4/25/53

4/25/53 Sat.

750 petriograph to 902

Experiment N

750-902

(-) by Examination

Tested as 750EK+L(-) by p. 192A

Sequestant	No. spots Pg. (% plate)	No. pop 750A-2 Spot	No. pop 902A-4 Spot	Probable locus	Solubility Ø
1.	0	0	20	gal,-	0
2.	1	0	24	1-	0
3.	4	c. 50	0	2-	0
4.	3	0	32	1-	0
5.	3	48	0	2-	0
6.	0	0	14	1-	++
7.	0	0	19	2-	0
8.	0	0	25	2-	±
9.	5	45	7	2-	0
10.	0 (2)	many 1 (2) small	14	1-	±
11.	1	0	26	1-	0
12.	4	0	26	1-	0
13.	0 X	0 X	13 X	X	0
14.	7 X	0 X	16 X	X	0
15.	9	0	0	2-	0
16.	4 (2)	53 (98)	4 (37)	2-	±
17.	0	7	22	1-	0
18.	3	1	13	1-	+
19.	0	0	33	1-	±
20.	3	0	25	2-	0
21.	0	0	12	1-	0
22.	0	1	14	1-	0
23.	0	0	23	1-	0

Test curves
= 750BY 44
control

OK
SEE
BELOW

OK
SEE
BELOW

OK
SEE
BELOW

OK
SEE
BELOW

5 gal,-
18 gal,-

21
4±
170

(-) = 4/26/53

See next page
CROSSER

Gal,- by X 902 on EMG gal SM

1. N3 (no + prot. observed) (-) = 679, 582, 531, 443 = 2135

2. N5 (") (-) = 369, 280, 274, 359 = 1283

3. N9 (") (-) = 363, 477, 419, 540 = 1799

4. N15 (") (-) = 455, 529, 479, 466 = 1728

5. N16 (") (-) = 638, 530, 535, 455 = 2358

N16 to 25 (5/4/53) returned no (+) = (61500) 6 = 0.9000

Activity of spores of these Sequestants

750	518	2175
04/2135	78	52
05/1283	79	60
06/1799	41	46
07/1928	44	43
08/2358	44	43

named as appear. named as appear.
with 750BY 44, 518, 2175, 37 pop.
control area control area
1/2 plate = 18 c. 16 control area = 1

5/4 N10 streaked out picked re gal duce d)

1/750 = 0 1/11 = 4 1/10 = 16 - control area 1

1/750A = 0 1/11A = 3 1/902 = 0 - control area = 0

1/750B = 0 1/11B = 0 1/902A = 0 - double (-) ? or hp? 3/5

5/19/53 N16 dots
translucent III

N16 " " " " " "

5/8 curves

1. N1	37, 26, 34, 23 = 0/114
2. N4	21, 28, 11, 28 = 0/88
3. N7	39, 66, 38, 35 = 0/178
4. N10	50, 33, 41, 52 = 0/176
5. N11	32, 20, 13, 40 = 0/105
6. N16	37, 33, 25, 44 = 0/139

state of curve test

8, 3, 6, 1 = 19 0+/132

7, 4, 1, 3 = 15 0+/103

5, 3, 7, 8 = 23 0+/201

Activity of gal,- lysate above

Spotted/2475	Spotted/911	Control
N1	0	Control = 0
N4	0	Control = 1
N7	0	Control = 2
N11	0	Control = 2
N16	0	Control = 2

Does this indicate? = blend

control a hole light

5/19/53

1/1000/2175 1/2 plate = 736

1/1000/518 1/2 plate = 580

1/1000/902 1/2 plate = 27

1/1000/902 1/2 plate = 19

1/1000/902 1/2 plate = 27

1/1000/902 1/2 plate = 19

197c

Expt. N continued

5/26/53 N16 - single colony isolation - ~~of plate~~ referred to as N16 original or prev
 lysate (pure, original) ^{labelled}
 original ~~1/150~~ 0(3) ~~control~~ ~~1/2175~~ 0
 1/2175 0 2
 1/238 c. 10 c. 10 ← not likely to be sufficient 222x 4x
 1/811 c. 7 c. 7

N16 original	transd	control
1/750	0	0
1/900	0	1
1/2238	0	8
1/811	0	4
1/1211	0	4
1/892	Solid smear	4

← this lysate also fails to transd 1485 → (-)
 1/14/56
 incubation
 N16 with 222 is 1-2?

N16 "plate" - because o.c. of N16 plate suspension as lysate gave no transd. and was not transd. by anything but 892 golt - (This repeated) - lysate
 N16 "plate" 2 } plate inoculation was made.
 1/811 solid smear - streak of this showed +, gave faint growth D(1)
 1/2175 control = c. 5 hi act. reconfirmed.

Additional crosses gal₂ - x 2257
 N7 1 plate = 1432 x 4 = c. 4200 (-) no (+)
 N4 1 plate = 844 x 4 = c. 3376 (-) no (+)
 N1 1 plate = 2676 x 4 = c. 10,800 (-) no (+)
 N11 1 plate = 1640 x 4 = c. 6560 (-) no (+)

Additional crosses gal₂ - x 2251
 N3 1 plate = 2660 x 4 = > 10640 total > 12,775
 N5 1 plate = 974 x 4 = > 3896 total 5359
 N7 1 plate = 1484 x 4 = > 5936 total > 7735
 N15 1 plate = 968 x 4 = > 3872 total > 5900

Galt nervous material of
 N1 - 1 mm HFT
 2
 N7 - 1 mm HFT
 2
 3

5/16/53 N16 lysate of 30', 40', 60' exposure don't show a great deal of diff. All HFT

4/20/53 Tuesday -

Lysate checks: On single plates - 0.05 ml lysate added (spots)

518	7. no add sector	2	- from lysate - activity stopped intermediate - may contain new phage active in top
	2. 22381 - 1 spot	4	
	3. 2175 1-4 spot	109	
	4. 750 pr "	3	
	5. 750 sr "	3	
750.	7. no add sector	0	
	2. 750 pr spot	1	
	3. 750 sr "	0	
	4. 2175-4 "	148	
	5. 2238 "	2	
2175	7. in add sector	4	
	2. 2238 spot	7	
	3. 2175 "	0	
	4. 750 pr	10	
	5. 750 sr	16	

Gal 4 - gal4-prts X 750sr = W2319 on EMS gal SM both Ft in cross?

7. Control sup.	- no growth	SM gal	
	- pure (-) on EMS gal		
X plates	(+)	(-)	Total
1.	5	0	5
2.	12	0	12
3.	21	0	21
4.	16	0	16
			<u>54</u>

0(+)/54

750pr X 750sr = W2319 (both probably Ft in cross)
EMS gal SM

7. Control - no growth of both EMS gal SM
- both (-) on EMS gal

2. X plates	(+)	(-)	Total
	0	42	42
	0	35	35
	0	30	30
	1?	32	<u>33</u>
			140

0/140?

H.M. ✓

5/2/53 Saturday

750E 811 - (1 day 1 Examination

- 1. no add - 7
 - 2. 750A-2 4
 - 3. 811A-8 10
- } no decision possible -

750pr - For the purpose of making production of 750 by ^{gal-} to observe segregation (-)

- 1. no add 3
 - 2. 811A-8 12
- pick and look for unstable

811 - For the purpose of making production gal- by gal- to obs. segregation (-)

- 1. no add 51
 - 2. 750A-2 96
- pick and look for unstable no unstable observed

5/4/53 Monday

750pr - For the purpose of making production of 750 by gal-

Spread plates

- 1. no add 3
 - 2. 811A-8 3
 - 3. 811A-8 2
 - 4. 811A-8 1
- } again! in mass

5/4 -> 5/7 conference about gal EMB - due to new tech?

2175 ductins that partly nullified by part of medium

17/22 gal very came out + on med. re-trying of these showed all to be gal-

5/7/53

gals -

Expt. 0 2175" tK-12 (F) regurgants.

Sequegant	no. pup. 902A1 spul	no. pup. 811-8 spul	Primate locus
01	0	11	2-
02	0	4	2-
03	contaminated		
" 4	10	1	13
" 5	6	1	17
" 6	1	0	12
" 7	11	0	13
" 8	8	1	13
" 9	← wt spread →		
" 10	2	1	12
" 11	1	0	13
" 12	0	0	6
" 13	0	0	15
" 14	0	0	4
" 15	0	0	9
" 16	0	0	10
" 17	1	0	9
" 18	0	0	10
" 19	0	0	11
" 20	0	1	7
" 21	0	0	13
" 22	0	1	10

20/20 2-

Lat Curves - c) Seq X	(2151) 90254 in	EM 5 gal SM - control	total
03	162, 154, 89, 95, 130 = 630	601, 608, 564, 796, 864	4070
05	139, 167, 162, 161, 728 = 757	1248, 1064, 964, 1092, 1016	5384
011	66, 78, 67, 47, 77 = 335	237, 228, 400, 416, 456, 5	2072
014	278, 201, 242, 318, 264, 723 = 1323	1089, 1328, 1322, 444, 1296	6988
			22,677

Deposits	/ 811	2175/
03	c. 15	0
05	c. 50	0
011	c. 50	0
014	c. 30	0
no add	2	0

locus

011 X 225 - 2nd cur
569, 501, 733 = 0/2163

2/24/54

Stocks of this experiment discarded as not worth saving

5/1/53 Sat.

Various 'ductins' for the purpose of obtaining seq. for classification

2175

- 1. mixed 16, 15
 - 2. 81A-8 32
 - 3. 750A-2 50
- 3/24 unstable after 2 streaking
 — 4/21 unstable after 2 "

Hiact. 1 - Anti serum test.

Hiact. 1 = O1 - dil 1-100 of O1-1 c. 10^{10} / ml

1-100 = 10^8 ml pre-heat → phage assay - dil $10^1, 10^2, 10^3 \xrightarrow{0.1 \text{ ml}} (10^6 \text{ dil}) = 35,24 = 2.95 \times 10^7$
 'ductin' assay - $0.1 \text{ ml} \rightarrow \frac{1}{4} = 212, 2176$

+ $\left. \begin{array}{l} 0.5 \text{ ml} \\ 0.5 \text{ ml} \end{array} \right\}$ 1-100 Anti Ph
 incubate 37C for 10 minutes

post-heat → phage assay - dil $1/10, 1/5, 10^2 \xrightarrow{0.1 \text{ ml}} (10^6 \text{ dil}) = 206,254 = 2.30 \times 10^7$
 'ductin' assay $0.1 \text{ ml} \rightarrow \frac{1}{4} = 213, 852$

$2176 \cdot 10 = 10 \cdot 10^2$
 2.2×10^7

to equate to pre heat
 mult. $\times 2 = 1704$

Franklin plaque summery = $\frac{2.30 \times 10^7}{2.95 \times 10^7} = 0.78$

Franklin plaque summery = $\frac{1704}{2176} = 0.78$

$\frac{2.3 \times 10^7}{2.95 \times 10^7} = \frac{1}{1.3 \times 10^2}$

2/24/54 Stocks of
 Trans Experiment
 198 discards

(in poor shape)
 at

Source: duck: # 191

5/14/53

Expt. R- 58T 902 (-) seg. tested by spotting = 702-4, 841-8

4-X

Seq	Cont. Avg	902 spot	811 spot	Phys. locs
R 7.	0	0	13	2-
8.	1	7	1	4-
9.	0	10	0	4-
10.	0	1	9	2-
11.	0	14	0	4-
12.	0	4	2	4-?
13.	contaminated (-)			
14.	1	10	1	4-
15.	1	11	1	4-
16.	contaminated with (+)			
17.	0	13	0	4-
18.	0	17	1	4-
19.	2	11	0	4-
20.	0	14	0	4-
21.	1	20	1	4-
22.	2	15	2	4-
23.	1	0	14	2-
24.	1	8	2	4-
25.	0	8	1	4-
26.	2	15	7	4-
27.	0	12	1	4-
28.	0	6	0	4-
29.	0	18	0	4-

not gal₂ - see below

Hydrols of Gal₂ - R1, R4, R17

Seq	SN	%
R1	c. 70/2	0%
R4	c. 60/2	0%
R17	c. 80/2	0%

3. 2-
 18 4-

Hydrol tests (looking for low act. d gal₂ variety) Crosses Gal₂ - X 2257

Seq	16/1	18/1	Cont. Avg
R2	16/1	18/1	4-
R3	33/8	26/42	4-
R5	83/8	34/42	4-
R8	67/8	34/42	4-
R9	16/1	0/4	4-
R11	8/1	0/4	4-
R12	17/1	2/4	4-
R13	4/1	1/4	4-
R14	14/1	2/4	4-
R16	11/1	5/8	4-
R18	5/1	2/1	4-
R19	2/1	1/1	4-
R20	2/0	3/4	4-
R21	7/0	1/4	4-
R22	7/0	2/4	4-
R23	5/0	0/4	4-

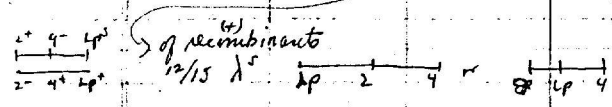
in those
 class irregular
 + lysate pop/cont. pop
 others spot test
 no pop. spot/cont. pop
 R7 was AT
 against 2170, 780

1. crosses R1, R2, R4, R17, 2257 in EMB gal - pure. only 2257 active in this locality
 R1 = 104, 99, 117, 39, 110, 126 = 0.595%
 7/1/53 1st 368, 488, 348, 247 } 1988
 2nd 152, 136, 318, 444 also correct 1436 }
 ignore R6 = 147, 175, 225, 127, 108, 109 = 17/91 NOT gal₂
 with heads R17 = 83, 41, 76, 78, 113, 85 }
 7/6/53 2nd 221, 107, 231, 152 } 1187 4/7 X 5

2. Gal₂ - X 1426
 7/6/53 R1 1/116, 2/873, 2/857, 2/226 = 1/3102 = 0.22%
 R17 - 6/1063, 892, 554, 3/511, 7/344 = 10/4364 = 0.228%

3. Gal₄ - X 1436 - cont. d.o.k.
 R2 - 4 plates are counted = 696 total c. 0+/2786
 R3 - 5 " " " = 538 " " 0+/2675
 R5 - " " " = 697 " " 0+/3485
 R8 - " " " = 368 " " 0+/1840
 7/6/53 " " " = 1028 " " 0+/4112 } 0+/5952

4. Gal₄ - X 902 - cont. d.o.k.
 R2 885, 583, 1474, 1410, 1465 = 3+/3183 = 0.094%
 R3 800, 679, 14563, 14625, 804 = 3+/3471 = 0.057%
 R5 4849, 21513, 47361, 14417, 705 = 17+/1722 = 0.35%
 R8, 788, 269, 369, 585, 14254 = 1+/1665 = 0.06%
 20+/121,611 = 0.21%



5/16/53 Saturday.

Effect of anti A serum on hiast A -

anti serum = anti P1
hiast A = D1 dil 1-100
Experimental

plaque assays against 1485
ductus assays against 750

antiserum dil 1-10

0.2 ml
0.2 ml
incubate
10 min
at 37C

1-100 dil D1-1

plaque assay
dil 1-100, 1-100, 1-10,

0.1 ml → 75 plaque
63

ave 69

titer 1-100 dil = 6.9×10^7 / ml

PRE-ASSAYS

gaeductus titer

→ 0.1 ml = 709 ductus
60F

ave 6.58×10^5 ductus/ml

$\frac{6.58 \text{ ductus/ml}}{6.9 \text{ plaque/ml}} = 1 \text{ ductus} / 100 \text{ plaque}$

1-100

ductus assay

123
139

ave 131

1-10, 1-5, 1-100

ductus assay

c 1000

0.1 ml 313 plaque
285

ave 299 plaque

sensitivity

plaque titer = 2.99×10^7

surviving ductus

titer = $131 \times 10^5 \times 2 = 2.62 \times 10^5$

Plaque survival = $\frac{299}{690} = 0.433$

ductus survival = $\frac{2.62}{6.58} = 0.398$

750 x 902 W. across unit of recombination between parental suspensions in an EM13 gel

Cross plate

1.	296 (-) 300	14
2.	c. 300	0
3.	c. 300	2(4)
4.	"	0
5.	"	0
	c. 1500	2

$\frac{2}{1500} = c. 0.13\%$

↳ a minimum no. since there were many small c. appearing colonies not counted

Recombination between the various gals -

Ref. above	% Recomb	no. plaques
1. gal ₁ x gal ₂	0.13	c 1500
gal ₁ x gal ₃	0.13	c. 1600
2. gal ₄ x gal ₁	0.38	1289
518 x "	0.07	1365
811 x "		
3. gal ₁ x gal ₂	0.13	54
1936 x 750	0.13	4588
1. gal ₁ x gal ₂		

0.3-0.6

5/18/53 Monday.

811 is lysates of 2277, 2278 - cultures of 14th genome for 892 X

1. m oad	42	
2. 2277	52	plaque c 10 ³
3. 2278	6	many plaques c 10 ⁴

5/25/53 Monday.

750X 2238 to obtain double minus purity. Controls ok.

	(+)	total
7.	24	c. 400
2.	0	c. 400
3.	0	c. 400
4.	0	c. 400
	2	1600

$$800 \frac{0.00125}{11}$$

$$= 0.13\%$$

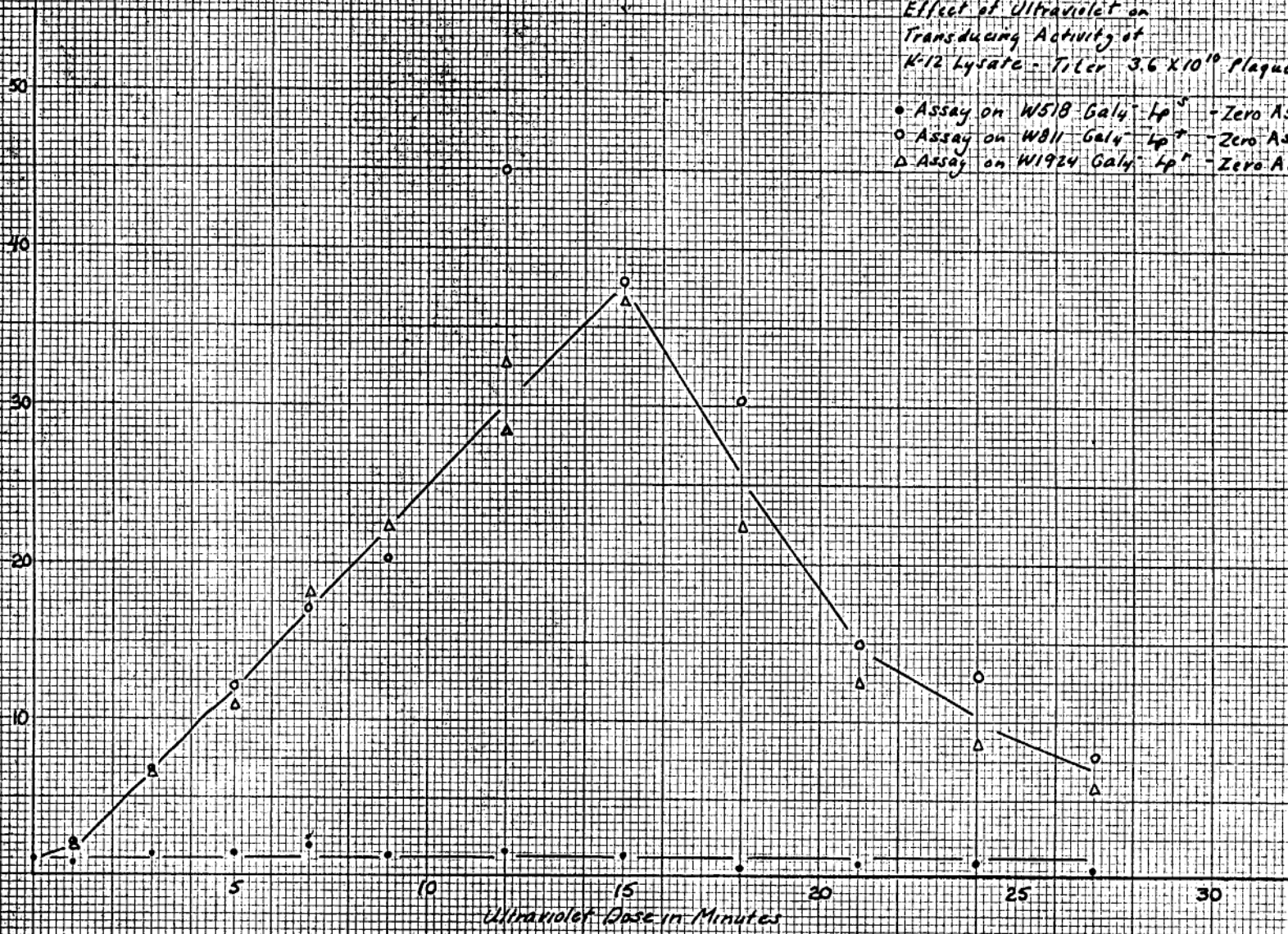


Relative Number of Transductions Per Ml.

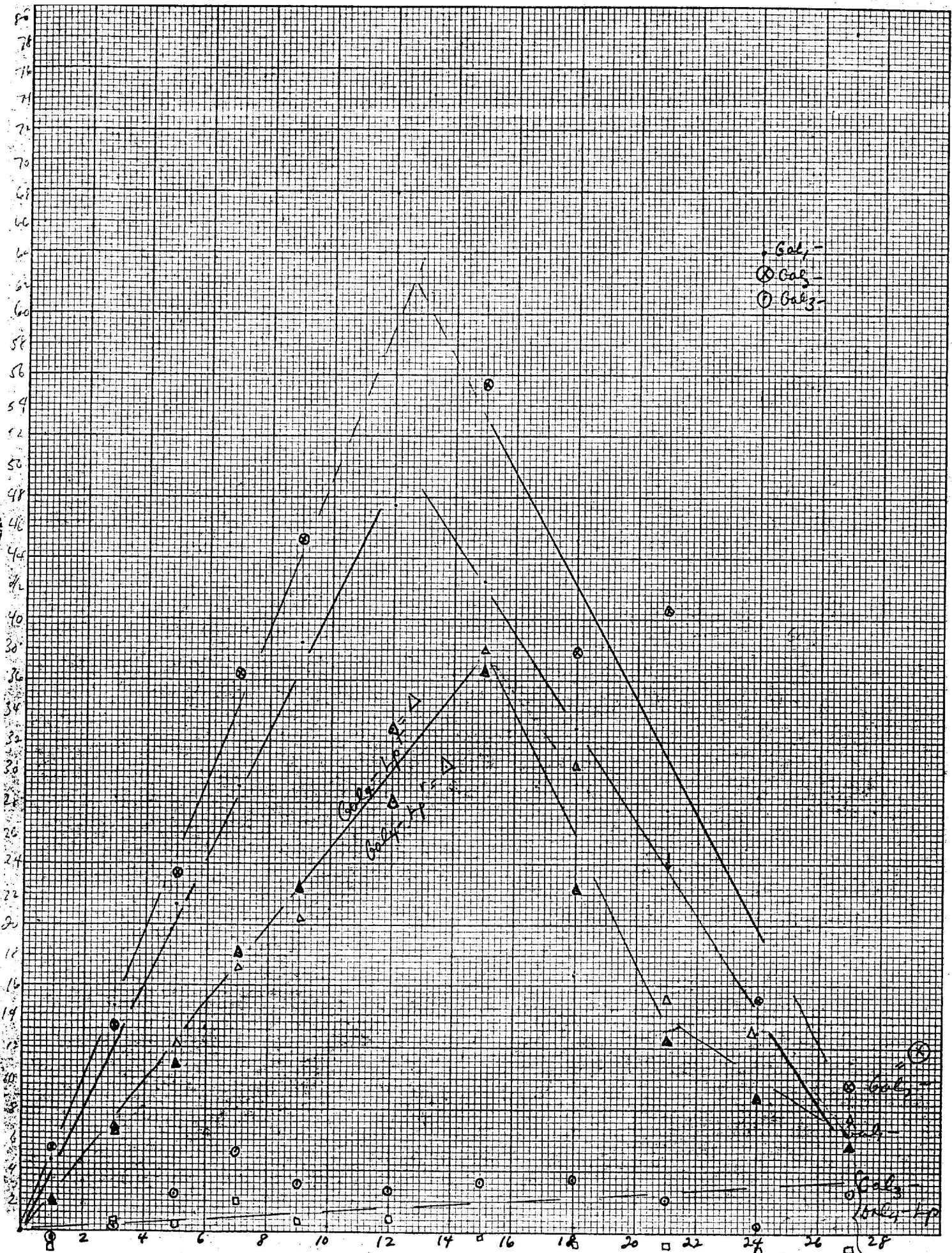
λ phage = 7.4×10^9
 Bacterium = 5×10^8

Effect of Ultraviolet on
 Transducing Activity of
 K-12 Lysate: Titer 3.6×10^{10} plaques per Ml.

- Assay on W518 Gal⁻ λ p^S - Zero Assay 8.2×10^3
- Assay on W811 Gal⁺ λ p^T - Zero Assay 1.9×10^3
- △ Assay on W1924 Gal⁺ λ p^T - Zero Assay 1.8×10^3



Ultraviolet Dose in Minutes



lysozyme

no's of papillae / ml lysate

Irradiation of K-12 λ - Effect on galactosidase

Irradiation	W2750			W2175			(1-10 die) W2297		
	+λ	-bkgrd	ratio	+λ	-bkgrd	ratio	+λ	-bkgrd	ratio
control - b/k	3 (4)	-	-	9 (14)	-	-	6 (12)	-	-
control + 0 dose	89	86	1.0	83	74	1.0	43	370	1.0
1 min	410	407	4.7	401	392	5.3	39	330	0.89
3 "	1257	1254	14.5	1006	997	13.4	49	420	1.3
5 "	2034	2031	21.2	1641	1632	23.4	82	760	2.3
7 "	2512	2509	29.1	2706	2697	36.4	185	1790	5.4
9 "	3306	3303	38.4	3356	3347	45.2	109	1030	3.1
12 "	3420 (413)	4100	47.6	4051 (60)	6011	81.4	97	910	2.7
15 "	368	3640	42.3	424	4100	55.5	119	1070	3.3
18 "	277	2730	32.9	294	2800	37.9	131	1190	3.6
21 "	243	2390	27.8	313	2990	40.5	82	700	2.1
24 "	116	1120	13.0	115	1100	14.9	58	460	1.4
27 "	55	510	6.0	84	700	9.7	102	900	2.7

W518 Lp⁺

W811 Lp⁺

W1924 Lp⁺

Irradiation	W518 Lp ⁺			W811 Lp ⁺			W1924 Lp ⁺		
	+λ	-bkgrd	ratio	+λ	-bkgrd	ratio	+λ	-bkgrd	ratio
control b/k	19 (41)	-	-	32 (36)	-	-	44 (49)	-	-
control + 0 dose	835	816	1.0	226	194	1.0	223	177	1.0
1 min	686	667	0.82	443	411	2.1	392	358	2.0
3 min	1118	1099	1.35	1369	1337	6.9	1264	1220	6.7
5 "	1182	1163	1.4	2409	2377	12.2	1996	1952	10.9
7 "	1680	1661	2.04	3384	3352	17.2	3296	3252	18.1
9 "	1108	1108	1.36	3977	3945	20.3	4056	4012	22.4
12 "	1060	1320	1.62	4036 (878)	8784	45.0	5144 (49)	5700	28.4
15 "	115	1150 (776)	0.1	776	7400	38.0	330 (660)	6551	36.6
18 "	48 (plaque)	480 (478)	0.1	626	5900	30.4	201 (402)	3971	22.2
21 "	74 (plaque)	740 (328)	0.2	328	2920	15.0	114 (228)	2231	12.5
24 "	79	790 (458)	0.17	290	2540	13.8	79 (158)	1531	8.6
27 "	40	400 (135)	0.1	186	1500	7.7	54 (108)	1031	5.8

$$\frac{132 \times 10^4 \text{ d/nd}}{1.8 \times 10^3 \lambda/\mu\text{l}} = \frac{1.3 \times 10^4}{1.8 \times 10^3} = \frac{13}{18}$$

$$\frac{2724 \times 10^4 \text{ d/nd}}{1.8 \times 10^3 \lambda/\mu\text{l}} = \frac{2.7 \times 10^4}{1.8} = \frac{27}{1.8}$$

Handwritten calculations and diagrams:

$$\frac{9724}{4036} = 2.4$$

$$\frac{12760}{2} = 6380$$

$$2 \times \frac{6380}{1360} = 23$$

$$2 \times \frac{4072}{220} = 37$$

$$\frac{1710}{622} = 2.7$$

K-121 5/30

Inactivation of K-121 - plaque survival

Dose	dilution	plaque counts	liters	survival
0	$10^7 \rightarrow$	357, 370	3.64×10^{10}	1.0
7	$10^7 \rightarrow$	337, 282	3.10×10^{10}	8.5×10^{-1}
3	$10^7 \rightarrow$	122, 114	1.18×10^{10}	3.2×10^{-1}
5	$10^6 \rightarrow$	548, 506	5.27×10^9	1.4×10^{-1}
7	$10^5 \rightarrow$	853, 749	8.00×10^8	2.2×10^{-2}
9	$10^5 \rightarrow$	202, 612	4.07×10^8	1.1×10^{-2}
diluted 12 1-10 repeatedly 15	$10^4 \rightarrow$	1033, 897	9.65×10^7	2.6×10^{-3}
	$10^5 \rightarrow$	69, 36	5.3×10^7	1.4×10^{-3}
	$10^3 \rightarrow$	103, 140	1.22×10^7	3.3×10^{-4}
18	$10^3 \rightarrow$	18, 3	1.1×10^6	3.0×10^{-5}
21	10^2	6, 6	6.0×10^4	1.4×10^{-6}
24	10^2	0		
27	10^2	0		

1.8×10^{-3}

8.2×10^3
 2.8
 43.6
 2.46
 $2.093 \times 10^3 = 3.1 \times 10^3$