DATE: 5/11/55 REF:

HFT 7⁻ ×6⁻ 342 D – Of 44 P.E. (−) picked for (+), 4 were true (−), 20 P.E. (−)

Procedure as in 343

	(+) at. seg?		/6	Segregant /7	/4		
P.E(−) #1	0	–	+	+	+	–	–
2	+	7	0 ✓	+	+		6⁻
3	+	10	~~0~~	~~0~~	~~+~~		lost
4	+	3	0 ✓	0	+		6-7⁻
5	0	–				–	–
6	+	12	0	0	+		6-7⁻ ← 344-6
7	+	7	0	+	+		6⁻
8	+	4				P.E.(−)?	
9	0	–					–
10	+	4				P.E.(−)?	
11	0	–					–
12	+	9	+	+	+	P.E(−)?	
13	0	–					–
14	0	–					–
15	0	–					–
16	+	4	0	+	+		6⁻
17	+	5	0	+	+		6⁻
18	+	730	0	+	+		6⁻
19	+	750					
20	+	10				P.E(−)?	

13 seg – one lost
7 nonseg.

5 6⁻
2 6-7⁻
4 P.E(−)?

(A) 344-6 above, the 6-7⁻, × K12λ 7/27/54
no. cols = 0
o. line λ = 44 see p. 325 for previous use of this lysate
many small pap.

Seg.	/6⁻	/7⁻	/4	Phenotype
1	0	0	+	6-7⁻
2	0	0	+	"
3	0	0	+	"
4	0	0	+	"
5	0	0	+	"
6	0	0	+	"
7	0	0	+	"
8	0	0	+	"
9	0	0	+	"
10	0	0	+	"
11	0	0	+	"
12	0	0	+	"
13	0	0	+	"
14	0	0	+	"
15	0	0	+	"

344B = [illegible] (+)
±– –
2⁻ + +

(B) 344-6 × 2580
no. cols = 10
o. line = 215

Segregant	/2	/6	/7	
1	0	+	+	2⁻
2	0	+	+	2⁻
3	0	+	+	2⁻
4	0	+	+	2⁻
5	0	+	+	2⁻
6	0	+	+	2⁻
7	0	+	+	2⁻
8	0	+	+	2⁻
9	0	+	+	2⁻
10	0	+	+	2⁻
11	0	+	+	2⁻
12	+	+	0	2⁻
344-13 → 13	+	0	+	6-7⁻
14	0	+	+	2⁻
15	0	+	0	6-7⁻ 2⁻?
16	NA+	0	+	
17	0	+	0	2-7⁻
redo 9/18	0 0	0 +?	0 0	[illegible]
344-18/19	0	+	+	2⁻
20	0	+	+	2⁻
21	0	+	+	2⁻

18 2⁻
2 6-7⁻
1 2-7⁻?

DATE: 5/21/55 REF:

	1	2		3	4	5	6	7	8	9	10
				342C1							
	HFT 4⁻ → 6⁻		24	P.E. (-) picked	3 segments.	7 were	true (-)	17 P.E. (-).	to m	344,343	
		(total seg?	no. seg.	/4	/6	/+					
	P.E. (-) #1.	+ eyes	13	+	o	+					
	2.	+	5	+	o	+					
	3.	+	6	+	o	+					
	4.	+	12	+	o	+					
	~~5.~~	~~+~~	~~10~~								
	5 6.	o	–								
	6 7.	+	3	+	o	+					
10	7 8.	+	24	+	o	+					
	8 9.	+	13	+	o	+					
	9 10.	+	11	+	o	+					
	10 11.	o	–								
	11 12.	+	14	+	o	+					
	12 13.	+	28	+	o	+					
	13 14.	+	11	+	o	+					
	14 15.	+	6	+	o	+					
	15 16.	+	9	+	o	+					
	16 17.	+	6	+	o	+					
20	17.	o	–								
		14 seg				14 6⁻					
		3 non-seg.									

DATE: 5/11/55 REF:

342E1

HFT 6 — x 4 — 24 P.E.(−) picked, 1 real(−), 27 P.E.(−). as in 345, 44, 43.

Lyt. Lyt. using culture pert in Lyt

1	2		3	4	5	6	7	8
	+obt. seg?	mut	/4	Seg. /6	/+			
P.E.(−) #1	0 = no	−						
2	0	−						
3	0	−						
4	0	−						
5	+ = yes	9	−	−	−	P.E.(−) Lp		
6	0	−						
7	0	−						
8	0	−						
9	0	−						
10	+	3	0	+	+	Lp^s	Gal	
11	0	−						
12	0	−						
13	+	1	+	0	+	Lp^s	Gal6 −	[illegible]
14	+	1	+	0	+	Lp^s	Gal6 −	
15	0	−						
16	0	−						
17	0	−						
18	+	1	0	+	+	Lp^s	Gal4 −	
19	+	2	0	+	+	Lp^R	Gal4 −	← 346−19
20	0	−						
21	0	−						3 Idn / 2Lp^s 1Lp^R
22	0	−						
23	0	−						2 Gal
	6 seg, 17 nonseg.		strk II	strk III				

This is Lp^R/s. Use Lyt

SEE 347

342E2 II — 5/26/55 Picked to Pen-Gran testerlead

1	2		3		4			6	
	+obt. seg	mut/strk	Seg.	seg +	seg for II, III				
✓ 1	0	−						−Gal6 − ?	
2	+	1	+	nd	[illegible]			Gal6 −	
3	+	7	−	0 nd	+		+	Gal4 −	I
4	+	1	−	0 nd	0		+	Gal4 Gal6 −	II
5	0	−	+	nd	− − +			Gal4 − Gal6 −	
6	0	−	+	nd	− + +			Gal4 −	
✓ 7	0	−	−	−					
8	+	8	−	0 nd	+		+	Gal4 −	I
9	0	−	+	nd	− + +			Gal4 −	
10	+	1	−	+ nd	0		+	Gal6 −	I
11	+	1	+	0 nd	+		+	Gal4 −	I
12	0	−	+	nd	+ − +			Gal6 −	
13	0	−	+	nd	+ − +			Gal6 −	
✓ 14	0	−	−	−					
✓ 15	0	−	−	−					
✓ 16	0	−	−	−					
✓ 17	0	−	−	−					
18	0	−	+	nd	− − +			Gal4 Gal6 −	
✓ 19	0	−	−	+	+ − +			Gal6 −	
20	0	−	+	nd	+ − +			Gal −	
21	0	−	−	−					
22	0	−	−	−					
23	0	−	−	+	+ − +			Gal6 −	
	17 nonseg, 6 seg		↑ 13 Total dis seg	16				8 Gal6 −, 5 Gal4 −, 2 Gal4 Gal6 −	

W3163
346-4 Ho ✓
5/16/56 found 4-6-7
← 346-4

5/27/55 Lysate of 346-4 x 2580
no odd = 14
0.1 ml = 71

Seg.	/4	/6	/+	Gen
1	0	0	+	4-6-
2	0	0	+	−
3	0	0	+	"
4	0	0	+	−
5	0	0	+	−
6	0	0	+	"
7−16	0	0	+	"
16	all	4-6-		

These results + x 346-4 →7

All apparently stable + were picked and spotted on W2589 (Gal2 Mal+ S^r) on B gal ahuplongai, mod. 20 sec, mucus. 2 days. No. papillae at spots observed — indicating NFT

W2589 found Gal − Mal −, λ-? resist

Won't mature?

5/19/55. After streaking III, remaining 7 sel (from broths) were spotted on W2175 on S gal + mod. 20 sec. After 2 days all show papillae — suggesting HFT

DATE: 5/25/55 REF:

1	2	3	4	5	6	7	8	9	10

1. Continuation of 346.

A. 346-4 × K12 7/27/54 $\frac{++}{--}$ = + ?

no add = 0

0.1 ml = 46 of 344-6

B. results on P. 346 under 346-4 × 2580

C. 346-4 × 2580

1. 1st 24 segregants were 2⁻
2. Rpt. streaking of above yielded 16(−) = 15 gal_2^-, 1 gal_6^- } lysate of a gal_6^- in this culture?
3. Rpt. " " " 15(−) = 13 gal_2^-, 1 gal_6^-

See 418 4. Rpt experiment using same lysate and applying to 2580 20⁻ 19 Gal_2^-, 1 Gal_6^-

Retest 346-4. See 346 for retest

Note, this 6⁻ requires 2+ days to be clear, have the tests of 346-4 been allowed to go long enough?

DATE: 6/6/55 REF:

285-1 Rpt 1Lam of 4⁻ x 1⁻

Cells removed from stab, streaked to obtain P.E. (−), restreaked to give 24 P.E. (−), from these 24 Gal+ obtained.

Gal+	Seg.?	(−)/total	/1	/4		
1	0					
2	0					
3	0					
4	+	2	0	+	1⁻	
5	+	5	0	+	1⁻	
6	0					
7	+	8	0	+	1⁻	
8	0					
9	0					
10	0					
11	+	6	0	+	1⁻	
12	+	10	0	+	1⁻	
13	+	5	0	+	1⁻	
14	+	3	0	+	1⁻	
15	0					
16	+	1	0	+	1⁻	
17	+	5	dark	dark	?	P.E. (−) and Gal +
18	+	5	0	+	1⁻	
19	+	+	0	+	1⁻	
20	θ		0	+	1⁻	
21	+	2				
22	0					
23	0					
24	0					
	+ = yes				11 1⁻	
	0 = no					

12 stable

12 seg.

DATE: 6/13/53 REF:

1. Do mixtures of Cel - ferment galactose?
NS13 + 1.0% Cel + ferm tubes vals

Reading after 2 days

1.	W 7750	Cel$_1$-	small bubbles
2.	W 911	Cel$_4$-	no bubble
3.	W2580	Cel$_2$-	small bubble
4.	W750+W2580	1+2-	small bubble
5.	W750+W911	1+4-	small bubble
6.	W911+W2580	4+2-	no bubble

} Apparently the fermentation in mixtures by the 1's

2. Tested by Inc
293-12J - Cel$_4$- 1HFT. Found equivalent or better than 2478-1

DATE: 6/20/55 REF:

1. Testing for segregation of λ_p in isolated lysogenic. Also effect of temperature shift on frequency of transduction.

A. Overnight cultures 2279, 2790 518 at 37C in incubator.
1 cultures divided and 1 portion placed at room temp for ca. half hour.
RM temp culs., 37C culs.

B. 1. Lysates
Gal_2 - λ for 2279 518 = 2342 3/30/54
Gal_1 - λ for 2790 = 2346λ
a. Lysates 0.4 ml ~~plated~~ in 2 small tubes, caps off incubated at 37C for half hour.

C. Technique.
0.4 ml lysate + 0.1 ml cult ← overnight cult { 0.1 + 10 Pen 2790 2279 / 0.5 + 10 Pen 518 }
↓
incubated 37C 10 min
↓
diluted
0.1 + 10,
1.0 + 10
↓
0.1 ml plated
Expt'l Counts

Control counts.
↓
0.1 + 10,
0.1 + 10,
↓
0.1 ml plated

37C Cultures	Plate 1	portion (1) 2	3		Cultures	Control Counts. RM temp	37C
W518	1/443	3/460	0/428	← these cols. also plaqued	W518 =	112, 103 (1.1×10^7)	119, 130 (1.3×10^7)
W2790	1/460	1/372	1/422	← also contaminated with plaques	W2790	35, 42 (3.9×10^6)	173, 184 (1.8×10^7)
W2279	8/173	1/166	5/142	← just about every colony has plaque - multiplicity too high	W2279	180, 144. (1.6×10^7)	338, 290 (3.1×10^7)

No gal + on these plates

The multiplicities in this experiment were too high to be used for the purpose of testing for segregation.

RM temp. Cult.	Plate 1	portion (1) 2	3	
W518	2/ca. 50	3	4	← these plates have contaminant - no count of total made.
W2790	3/263	1/292	0/262	
W2279	1/55	3/56	0/50	← these colonies all are plaqued. mult. too high

DATE: 6/23/55 (the day was written over; it may have read 6/27/55) REF:

1	2	3	4	5	6	7	8	9	10

1. Observations on segregation rate:

A. Culture: 348 idio type
B. Streaked out onto single pure col. inoculated into Pen-amon overnight
C. Diluted $10^2 - 10^4 - 10^6 - 10^8$

10. 0.3 ml samples incubated 37C 8:30 AM → 1:00 PM

Plated 0.05 ml of

1.	361
2.	61
3.	219
4.	122
5.	855
6.	1279
7.	441
8.	676
9.	319
10.	336

Below 60 min by. div every 30 min, from 9:30AM → 11:30 (12:00)
2^5 – leave until 1:00PM
2^8 = (256X)

5 0.1 ml samples plated B gal

1.	100
2.	108
3.	134
4.	136
5.	108
	586 / 5 = 117

Therefore ave. of 1.2 cells/0.1 ml ↑ = 3.6 cells/tube

1.05 ml

0.6 cell

6/24/55

2. Repeat experiment using 750tKR-1
A. Overnight culture from stock.
B. diluted $10^2 - 10^4 - 10^6 - 10^8$

5 0.1 ml samples incubated in horizontal 0.2 ml pipette at 37C. 9:15 AM
20 in number

Plated 1:45 PM
(and [illegible])

Assay 0.05 ml

1.	98,	(2-)
2.	112,	(5-)
3.	106,	(4-)
ave	106	(4-)

212 (8-) / 100 = 2 cells/0.1 inoculum, 0.08 [illegible]

(1)

Samples incubated in pipette	(–)	total
1	*	*
2	6	1169
3	3	595
4	4	251
5	0	237
6	*	*
7	23	1252
8	* 103	* 2730
9	319	1622
10	↓	↓
11		
12		
13		
14	15	1/4 = 141
15	9	1113
16	all (–)	
17	22	1966
18	15 (19)	1/4 = 479
19	est. 100	est. 2000–3000
20	19	897

(2) At the same time 5 0.1 ml ~~pipettes~~ samples plated in drop form on B gal, respread after platings ca. 2:15PM by adding 0.1 ml broth + spreading

	(–)	(+)	
1.	0	89	2 seg. colonies
2.	0	52	2 seg. colonies
3.	7	1295	
4.	22	1222	
5.	52	730	

Calculations on next page – also rates –

*. These platings have ca. 3000 cells

The no. (–) appears to vary from ca 10–300

$a = 0.602 (\text{no. mutants}) / N \log N$

DATE: REF:

	2 Incubation in cm^2 plates	3	4	5	6	7	8	9	10
	No. (-)	Total cells	N log N			0.602 R	5/3	Recalculated	
	6	1169	1169 (3.068) = 3580			3.612	1.0×10^{-3}	1.0×10^{-3}	
	3	595	595 (2.77452) = 1650			1.806	1.1×10^{-3}	1.1×10^{-3}	
	4	251	251 (2.39967) = 600			2.408	4.0×10^{-3}	4.0×10^{-3}	
	23	1252	1252 (3.09776) = 3880			13.846	3.6×10^{-3}	3.6×10^{-3}	
	9	1113	1113 (3.05308) = ~~3400~~ 3391			~~5.418~~ 5.418	~~1.6×10^{-3}~~	✓ 1.6×10^{-3}	
	19	897	897 (2.95279) = 2650			11.438	4.5×10^{-3}	4.3×10^{-3}	
	0	237	—	—		—	—		
10	B. Regrowing								
	7	1295	1295 (3.11227) = 4030			4.214	1.0×10^{-3}		
	22	1222	1222 (3.08707) = 3780			13.244	3.5×10^{-3}		
	52	730	730 (2.86332) = 2085			31.304	1.5×10^{-2}		
	0	89							
	0	52							
	103	2750	2750 (3.439) = 9430			62.00	6.6×10^{-3}	6.6×10^{-3}	
	319	1622	1622 (3.21005) = 5200			~~197~~ 192.	3.7×10^{-3}	✓ 3.6×10^{-2}	
20	22	1966	1966 (3.29358) = ~~8~~ 6460			13.2	2.0×10^{-3}	2.8×10^{-3}	
						$\frac{3.20}{1}$	$2 < \times 10^{-3}$		

Estimated P_f.

$a = \frac{2.3}{200} \log \frac{1}{\frac{1}{10}} = (0.012)(\log 10) = (0.012)(1.08) = 1.3 \times 10^{-2}$

Using 200

30

Using 1024 $a = \frac{2.3}{1024} \log \frac{1}{\frac{1}{10}} = \frac{2.3}{1024} \log 10 = \frac{2.3}{1024}(1) = 2.2 \times 10^{-3}$

40

50

DATE: 6/22/55 REF:

Yield of HFT λ from heterogenote

1. Culture 241-14 (([illegible])) - streaked from stock and 10 colonies picked, tested for HFT against 2279 - Colonies stored [illegible] fig

O O O O O
O O O O O

2. Reaction not apparent 24 hours - select most likely appearing col and proceed. Broths incubated 24 hours - Estimated titer 10^9

3. Expt.
   a. dilute 1.0 ml prep to Blank ca. 1000 col / ml in Saline. Irrad 35 seconds and plate on 6B gal with 750 col. - Make pre irrad. count
   b. Inc. 2 hours - reappread 3 plates

4. Results:

Pre irrad count / 0.1 ml =		67				
		50	ave: 53			
		46				
Post irrad plaques / 2279				Exp.	PAP [illegible]	3.5 + A [illegible]
0.1 ml:		22		5	3	(1) 3/5 +^u
		12	ave 16	8	4	(2) 5/8 +^u
		14		5	2	(3) 2/5 +^u
				352B stk 1		
Post irrad. pg / 737	(1)	2		1 0/2 +^u		
NOT SPREAD	(2)	5	ave 3	2 1/5 +^u		
	(3)	2		3 1/2 +^u		
				352C (stk 1)		
	(1)	3		(1) 3/3 [illegible]		
SPREAD	(2)	2	ave 2	(2) 1/2 [illegible] ←		
after 2 hours	(3)	3		(3) 2/3 [illegible], 1G		

DATE: 6/22/55 REF:

1	2	3	4	5	6	7	8	9	10

For multiplicity reduction, - does λ segregate?

1. Culture - 2279 overnight
λ - 241-14 (10/12/54) titer 5.3×10^8 plaques, 1.3×10^8 hazard } see p 300

(a) 0.4 ml 2279 + 0.1 ml HFT 2⁻ ; " 0.1 ml broth }

(b) incubate 37C 10 min.

(c) dilute $10^3, 10^4, 10^6$ → plate 0.05 ml
λ - 6 plates

(1) Control 3 plates — 53, 47, 49 on Gal - $50 \times 20 \times 10^6 = 1.0 \times 10^9$

(2) Expt. plates

(+)	total
1	66
0	54
0	40
0	41
0	54
0	56
(1)	311

no (-) found colonies with λ or partial lysed.

tested against 2279 and found not lysogenic

353-1

Rpt.

1. Culture - 2344 JL's δ stock Gal_2^- Ly^s Hfr
taken from his stock culture labelled δ

2. Lysate - 750HKR-1 1/28/55 titer = trnsd. titer - 2.6×10^7 see 315

3. Expt. 0.1 ml lysate + 0.1 ml cells inc. 37C 15' - 10 ml Pen added, diluted $10^2, 10^4$ → 0.1 ml

Results -

A. control plating - exposure to broth

Plate 1. 237; 2. 326; 3. 283 } $282 \times 10 \times 10^4 \times 10^2 / 2 = 1.41 \times 10^9$ cell/ml overnight cult.

B. Lysate exposed

	+ *	(-)	(-) partially lysed	total
1.	3	92	0	95
2.	2	84	4	90
3.	0	76	1	77
4.	3	71	4	78
5.	1	61	0	62
6.	0	94	1	95
7.	0	96	2	98
	9		12	595

from Pen-ase titer estimated 750 HKR-1 5.6×10^7

* the + appear to be in two categories (1) Appearing ca. 18 hours and represented by intact (not lysed) colonies that are almost all (+) (2) appearing in (-) colonies, λ contaminated and partially lysed, after ca. 24 hours. In the above there were 5 were early, 3 late.
This correlation was noted.

Pattern	Lysis grid 10⁸ strd.	Fraction Gal +
early	2/5	predom. (+)
late	3/3	" (-)

} are trnsd. not purified - total = 8

C. Plates - primary streaks of (+) given to EML.
1-5 early - 2/5 Ly pos
6-8 late - 3/3 ↗

EML 900
Give letters A-H

DATE: 6/30/55 REF:

Crossover rate in P.C. heterozygote
342E2 6 → 4
Expt as in 357 with 750CKR-1

1. Overnight culture from single colony (inoculated 9:30 → 2:30)
2. Dilute $10^2 - 10^4 - 10^6 - 10^8$ → 20 0.1ml samples

3 plates for estimate of ratio (cells/0.1ml)

	(+)	total	
1.	4	67	(8 col-)
2.	4	78	(9 col-)
3.	6	78	(8 col-)
		ave 71	= 0.71/0.1ml

Using null method and N = 5900 (from culture #4)

~~$a = \frac{2.3}{5900} \log \frac{1}{12/18}$~~

~~$a = (0.00039)(0.176) = 7.0 \times 10^{-5}$~~

$a = \frac{2.3}{2.9 \times 10^3} \, 0.176 = \frac{0.405}{2.9 \times 10^3} = \frac{4.05 \times 10^{-4}}{2.9} = 1.4 \times 10^{-4}$

These all low because log values used

	(+)	total	Max Prob
1.	0	0	—
— 2.	0	1406	1.3×10^{-4}
3	0	ca. 3000	6.0×10^{-5}
— 4	0	1/8 = 193 = 1544	—
— 5	0	1/16 = ca. 193 3088	6.0×10^{-5}
6	0 contaminated		—
7	ca 1000	ca 3000	—
8	0	ca. 150	—
— 9	4	1/8 = 643 5144	ca. #3 1.3×10^{-4}
10	0	ca 4000	ca. #3 4.1×10^{-5}
— 11	2	1/8 = 841 6728	5.3×10^{-5}
— 12	5	2266	4.0×10^{-4}
13	0	0	—
— 14	1	1/16 = 370 5920	3.0×10^{-5}
— 15	0	ca. 7000	2.5×10^{-5}
16	ca 1000	ca 2000	—
17	0	0	—
— 18	0	2245	—
— 19	0	748	"
— 20	0	34	—
← total		29128/10 = (2913)	

B Reciprocal 342C1 As above – except stab picked to dilute per and then grown overnight

		(+)	total
10:30 AM	Control 1.	0	577
1:45 PM	2	2	595
	3	0	556
			583 = 5.8/0.1ml

~~Using null method, and N = 1200 (4A)~~

~~$a = \frac{2.3}{1200} \log \frac{1}{17/20} = (1.9 \times 10^{-3})(0.13)$~~

~~$= 2.6 \times 10^{-4}$~~

$a = \frac{2.3}{779} \log \frac{1}{17/20} = \frac{2.3}{7.79 \times 10^2} \log 1.2$

$= 2.4 \times 10^{-4}$

	(+)	total	each	
1.	2	728	276 (100%)	4.0×10^{-4}
2.	0	818	62 (810)	
3	0	1/16 = 110	660	
4.	0	1/8 = 86	688	
5.	0	1/8 = 80	640	
6	0	1/8 = 49	392	
7.	0	1/8 = 80	640	
8	0	1/8 = 91	728	
9.	0	1/8 = 83	664	
10	0	1/8 = 149	1192	
11.	0	1/8 = 62	496	
12	0	1/8 = 83	664	
13.	0	1/8 = 137	1096	
14	0	1/8 = 74	592	
15	0	1/8 = 90	720	
16.	0	1/8 = 152	1216	
17.	1	185	185	
18	0	1/8 = 87	696	
19.	1	1249	1.4×10^{-4}	
20.	0	1/8 = 62	496	
		20 | 15588	1559/2 = (779)	

(353)

DATE: 6/30/53

REF:

1924 → 2346 λ to look at single hole counter

1 0.1 ml overnight culture + 10 ml

0.1 ml + 0.1 ml 2346 λ 11/13/54

↓

incubated 37C - for 10^{-1}

↓

add 10 ml Pen → dil 10^{-2}, 10^{-4} → 0.1 ml samples

2. Broth - centrifuge

2346 λ 11/13/54 - expt.

DATE: 7/6/55 REF:

Segregation from w2730

1. Split as before - see page 357

2. Overnight cult

$10^{-2} - 10^{-4} - 10^{-6} - 10^{-8}$ → 20. 0.1 ml sample.

	0.1 ml (+)	total
1.	17	122
2.	19	168
3.	21	151
	19	441/3 = 147

= 1.5 cells/0.1 ml

Using mull fraction

$$a = \frac{2.3}{100}\left(\log \frac{1}{p_0}\right) - \frac{2.3}{100}\left(\log \frac{1}{0.5}\right) = 0.012$$

$= 1.2 \times 10^{-2}$

Original water dilution

Size → 40, 80, 200, 150, 40, 30, 150, 100, 30

ave at 100

	(+)	total	
1.	all	323	8.1×10^{-3}
2.	2	176	3.2×10^{-3}
3.	0	0	8.9 —
4.	8	1669	5×10^{-4}
5.	3	317	2.2×10^{-3}
6.	52	1236	8.1×10^{-3}
7.	0	10	—
8.	0	0	—
9.	ca. 300	ca. 700	1 —
10.	36	1055	6.6×10^{-3}
11.	0	0	—
12.	0	0	—
13.	3	299	2.4×10^{-3}
14.	0	0	—
15.	0	0	—
16.	6	386	3.6×10^{-3}
17.	0	0	—
18.	0	0	—
19.	55	1965	5.1×10^{-3}
20.	0	0	

46.2 = 5.1

8 failed to grow 9

1 contaminated Gal+

10 Contaminated both Gal+ Gal-

DATE: 7/13/55 REF:

1. Rpt 234/HI (SL stock) X from HFT 4
Lysate = 293-125 —

2. 0.3 ml lysate + 0.2 ml overnight cult.
↓
add 10 ml broth
↓
1-10², 10⁴
↓
0.1 ml

broth as control.

Plates bad (too wet)

3. Result. A. Control. 4/10 · 10³ · 10² · 10 = 6X10⁷

1. 457		?		
2. 540	5.0X10⁹		m(+)	
3. wet plate				

B. Expt.

	(+)	total (−)	contam. (−)	Decay.
1.	2	322	2	0 ? 0 1
	1	ca 300	0	0 3

Rest of plates (−5) have been in acute counts — above the same as above — after 2 days some (+) appeared — may be reversions — The culture seems to revert more than other Gal₂ —

DATE: 8/10/55 REF:

Pevonsay Scott Cot tubes – Blank = water – PC-4 filter

λ	OD	Crystal Cult ca. 10^9/ml)
650	0.051	0.74
600	0.071	0.81
550	0.11	0.90
500	0.19	1.04
450	0.365	1.23
400	0.630	1.45

Lysates	1210	K12	2236
10:50AM 0	0.58	0.73	0.80
1:10 PM 2:20	0.75	0.52	0.80
1:25PM 2:35	0.69	0.44	0.64
1:45PM 2:55	0.67	0.40	0.53
	0.67	0.40	0.48

$\frac{0.2}{20.0} \times 5$ 0.05% RNA

9/25/55.

750EK12-1 grown on B gel + 0.05% RNA

1. no RNA 5 gal – + ca 10 sey + ca 60 gal +
2. RNA 6 gal – + ca " + ca 70 gal +

} No effect of RNA at this conc

1210 λ – × 750 in search of HFT δ

no odd 2
0.1ml 18 per lysate

HFT test	No. tests	against?	HFT?
I	10	750, 2341	possibly no/2341 – lysate made 358-1
2.	10	750, 1210	possibly no/750 358-2

] now HFT – reversion of bin made – both stable

2279 + 750 ← K12 8/10/55

1. 750 dil: 142×4: 568
2. 2279 0.100 426

← Rather low titer on lys – This lysate has been at room temp since it was made

no Control

DATE: 8/17/55 REF:

1	UV	3	4	309-1 = (HFT ?) 6	7	8	9	10

Assay of irradiated HFT by sate - Irradiated since time ago - Preliminary assay.

(359A) ← ← irradiated at later

UV Dose	1-100 dil /750 (0.1 ml ?)		0.05 750 /2307	spontaneous + = 5 frequency ← 0.5 ml 10⁴ 2580	.05 ml 10⁴ 2790	plaque	0.1 ml /2790 ref	1.6×10⁷ purified 2790
0 min	21	0	13	257	37	ca. 500+	196	
1 ..	273	252	14	729	190	ca 500+	297	
2 ..	517	496	6		227	584	994	ca 1000
3 ..	593	572	6		341	436		
4 ..	885	864	10		413 (1st plate)	234	← at max 413×20×10⁵ = 8.3×10	
5 ..	950	929	11		175 (with plate)	16		
6 ..	809	888	11		148	7		
			spont = 8	sp = 22	sp 14	sp 17		

1-100 dil for conc. frequency on 2790 - dil at least 10² more

← RPT = 359/2

these with precarious plates

See 359A →

359A test ← λ^{test} rev against 2279 →

λ /2790 + = lysogen 0 = not lysogen; s = sens, r = resist

ns = not segregating

	Spont (+) λ /2790	0 λ /2790	1 λ /2790	2 λ /2790	3 λ /2279	4 /2279	5 λ /2279	6 λ /2279
1.	s ns 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
2.	s 0	r s 0	r? ns 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
3	s 0	r s 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
4	s 0	s ns 0	r ns r+	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
5	s 0	s ns 0	r s r+	s ns 0	s ns 0	r ns 0	s ns 0	s ns 0
6	s 0	r s 0	r s 0	s ns 0	s ns 0	r ns +	s ns 0	s ns 0
7	s 0	r s 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
8.	s 0	r s r+	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
9	s 0	s ns 0	r s 0	s ns 0	r ns 0	s ns 0	s ns 0	s ns 0
10	s 0	r s 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
11	s 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
12	s 0	s ns 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
13		s ns 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
14		s ns 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
15		r s 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
16		s ns 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
17		r s r+	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
18		r s r+	r s 0	s ns 0	s ns 0	r s 0	s ns 0	s ns 0
19		s ns 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
20		r s 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0	s ns 0
21		r s 0	r s 0	s ns 0	s ns 0	r s 0	s ns 0	s ns 0
22		r s? 0	r s 0	s ns 0	r s 0	s ns 0	s ns 0	s ns 0
23		s ns 0	s ns 0	r s 0	s ns 0	s ns 0	s ns 0	s ns 0
24		r s 0	r s 0	—	s ns 0	s ns 0	s ns 0	s ns 0
	12/12 lp^s	9 lp^s, 3 lp^+, 12 lp^r	3 lp^s, 2 lp^+, 19 lp^r	22 lp^s, 1 lp^r				
		−5.6 lp^s	−1.7 lp^s	−1.44 lp^s				
	12 ns	9 lp^s ns, 3 lp^+ s, 12 lp^r s	3 lp^s ns, 1 lp^+ ns, 1 lp^r ns, 1 lp^+ s, 18 lp^r s	22 lp^s ns, 1 lp^r s	23 ns − 22 lp^s, 1 lp^r; 1 s lp^r	22 ns − 20 lp^s, 1 lp^+, 1 lp^r; 2 s lp^r	24 ns	24 ns

↑ The surviving growth from 16 of these streaks, streaked on B gal - found pure Gal+ - adding λ does not restore stability

50

359A

2915 used for λ^x

DATE: 9/28/55 REF:

Rpt. of 359 - 2' dose /2750 48 papillae picked streaked out single col. tested /λ 1229 streaked out on B gal for stability. This dose taken because less likelihood of contam. from nearby plaques - Early doses require more dilution to avoid

Rev of second streak	#	Dose 2' /2779 (359A2')	Gal-ρ λ	359A0 /2750	359A1' /2915
ws	1	s ws o		r s o	r s o
s	2	r s o	Lp^s	r s +	r s +
ws	3	s ws o		r s o	r s o
ws	4	s ws o		r ws +	r s o
ws	5	s ws o		r s o	r s o
s	6	r s o	Lp^s	r s o	r s o
w	7	s ws o		r s o	r s o
s	8	r s o	Lp^r	r s o	r s o
ws	9	s ws o [illegible]		r s +	r ws o
ws	10	s ws o		r s o	r s +
ws	11	s ws o		r s o	r s o
ws	12	s ws o		r s o	r s o
ws	13	s ws o		r s o	r s o
ws	14	s ws o		r s +	r s o
ws	15	s ws o		r s o	r s o
—	16	no growth		r s o	r s o
ws	17	s ws o		r s o	r s o
ws	18	s ws o		r s o	r s o
ws	19	s ws o		r s o	r s o
ws	20	s ws o		r s o	r s o
s	21	r s o	Lp^s	r s o	r s o
ws	22	s ws o		r s +	r s o
ws	23	s ws o		r s o	r s o
ws	24	s ws o		r s o	r s o
s	25	r s o	Lp^s	r s o	r s o
ws	26	s ws o		r s o	r s o
ws	27	s ws o		r s o	r s o
ws	28	s ws o		r s o	r s o
s	29	r s o	Lp^s	r s +	r s o
ws	30	s ws o		r s o	r s o
ws	31	s ws o		r s o	r s o
ws	32	s ws o		r s o	r s o
ws	33	s ws o		r ws +	r s o
ws	34	s ws o		r s +	r s o
s	35	r s o	Lp^s	r s o	— —
ws	36	s ws o		r s o	— —
ws	37	s ws o			
ws	38	s ws o		8 Lp^+	2 Lp^+
ws	39	s ws o		78 Lp^r	32 Lp^r
ws	40	s ws o			
ws	41	s ws o		2 ws Lp^+	33 s Lp^{+r}
ws	42	s ws o		34 s Lp^{+r}	1 ws Lp^r
ws	43	r s o	Lp^s		
s	44	r s o	Lp^s		
ws	45	s ws o			
ws	46	s ws o			
ws	47	s ws o			
total ws	48	s ws o			

$Lp^+ = 0$
$Lp^r s = 9$
$Lp^s w = 38$

10 Lp^r s, 60 Lp^s ws — total

Recessive 10/3/55
0.1 ml 10^4 dil /2915

	c	= 25	ca 5 pl : 1 t
359A0	0	= 355	V_{co}
359A1	1	= 662	ca 2 pl : 1 t
	2	= 678	ca 1 pl : 1 t

Assay of these for pract. sig., $λ^x$ ors ←

359B

DATE: 10/6/55 REF:

Continuation of 359 359A

Plating of UV irrad 309-1 lysate on 2580 (lp+) - 0.1 ml 10^5 dilution of irradiated lysate.

UV dose	Papillae	het	Assay/2341 (same dose)
control	31	het	?
0	75	44	35
1	183	142	
2	388	347	
3	576	545	
4	710	689	253
5	563	532	
6	412	381	

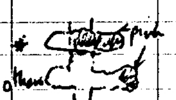
#10 streaked ... -10 ul. V against 2776 all ...

359B(4) 7th seq #10

on plate not work

55 = 54
ns = more seg
+ = lysogenic
0 = non lys

	0 dose /2279		4' dose	Seg	/2279	1st seg	2nd seg	3rd seg	4th	5th	6th	7th	
1	ns	+	r	s	+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+ (wk)	2-lp+	2-lp+	359B1 clear streaks 2 stab
2	s	+	r	s	+ x	2-lp+	2-lp+	2-lp+	2-lp+	2-2-lp+	2-lp+	2-lp+ wk	
3	s	+	r	s	+ x	2-lp+	2-lp+	not obt.	2-lp+	2-lp+	not obt.	2-lp+	
4	s	+	r	s	+ x	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	
5	s	+	r	s	+ x	2-lp+ *	2-lp+	2-lp+ *	2-lp+	2-lp+	2-lp+	2-lp+	
6	ns	+	r	s	+ x	2-lp+	2-lp+	none	2-lp+	2-lp+	2-lp+	2-lp+	
7	s	+	r	s	+ x	2-lp+	contam.	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	
8	s	+	r	s	+ x	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	359B10 clear stab
9	s	+	r	s	+		2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	
10	ns	+	r	s	+ x	2-lp+ *	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+ R	
11	s	+	r	ns	+		none	none	none	none	none	none	
12	s	+	r	s	+ x	2-lp+	2-lp+	2-lp+	not test	2-lp+	2-lp+	2-lp+ wk	1
13	ns	+	r	ns	+		none	none	none	none	none	none	
14	s	+	r	ns	+		none	none	none	none	none	none	
15	ns	+	r	s	+ x	2-lp+ *	2-lp+	2-lp+	2-lp+	2-lp+	not obt	2-lp+ wk	
16	s	+	r	ns	+		none	none	none	none	none	none	
17	s	+	r	s	+ x	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	1
18	ns	+	r	s	+ x	2-lp+ *	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	
19	s	+	r	s	+		not obtained	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	
20	s	+	r	ns	+		none	none	none	none	none	none	
21	s	+	r	s	+ x	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	
22	s	+	r	s	+ x	2-lp+ *	2-lp+ 2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	← 359B-22
23	s	+	r	s	+		contam	none	none	none	2-lp+	none	
24	s	+	r	s	+ x	2-lp+ *	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	2-lp+	
	6 ns 18 s	24+		5 ns 19 s	24+								



	1st seg	2nd seg	3rd seg	4th	5th	6th	7th	
	14 endo 2 exo	14 endo 1 exo 1 amphi	16 endo	16 endo 1 exo	16 endo 1 exo 1 amphi	17 endo	17 endo 1 exo	110 6 2

55 17
2 1
1 2

DATE: 8/19/55 REF:

1924 × HFT 1⁻ to study lysogenization + crossing over

α-λ$^{\varepsilon}$ × 1-λ^{+}

1. Transd. plates not recorded - ca. 300 colonies/plate
   no gal+ in control, lysate agreed.
2. After 2 days on the lysate plate several papillating colonies
3. Result:
   Papillating colonies streaked out and from the primary streaks 8(−) and 8(+) tested for lysogenicity against 2279

	8 gal+/gal?	Gal+ seg?	Gal-lys?	dry ± plain?
Pap. col. 1	no lys	no seg	no lys	—
– 2	no lys	no seg.	no lys	yes.
– 3	no lys	no seg.	no lys	—
4	no lys	no seg	no lys	—
5.	no lys	no seg	no lys	=
6.	no lys	yes	no lys	yes.
7	no lys	yes	no lys	yes

} discarded

Gal+ 1984 360-2 Aug 26 discarded

4. 360-2 - 24 Gal+ colonies from 24 diff. ± colonies tested /2279 for lysogenicity
   all found not lysogenic.

Unaerated cultures (ov. nite) read. and incubated without air density ca. 10⁸

	stbl?	/2279	-seg/2279	HFT /1	HFT /4	Genotype	Confluence spotted /2279	Gal+ stbl?
1	stbl	non lys						
2								
3								
4								
5								
6			not done					
7								
8								
9								
10								
11								
12								
13	unstbl		non lys	0	0	1-4⁻	ca. 1000 plaques	— ← 360-3
14				0	0	1-4⁻	"	— 360-4
15				0	+	1⁻	"	(1) 1 found stbl
16				0	+	1⁻	"	(1) 1 found stbl
17				0	+	1⁻	"	(2) 2 found stbl
18				+	0	4⁻	"	(9) 8 found stbl
19				+	0	4⁻	"	(8) 7 found stbl
20				0	0	1-4⁻	"	—
21				0	+	1⁻	"	(0) —
22				— (E(−))	—	—	"	[illegible]
23				+	0	4⁻	"	(5) 4 found stbl
24				+	0	4⁻	"	(9) 8 found stbl

4 prods
4 ess
3 amphi

DATE: 8/21/55 REF:

1	2	3	4	5	6	7	8	9	10

Preliminary Expt. on seg freq. from $\frac{-\ +}{+\ -}$ both Lp^+ and Lp^R/L

1. Culture of W2868 $\frac{4^- 2^+ Lp^S}{4^+ 2^- LpR}$

A. Overnight culture diluted $10^2 - 10^4 - 10^6 - 10^8 \rightarrow$ 10 0.1ml sample

	(+)	(−)	total	
assay 0.1 1.	40	3	43	mean/sample
2.	76	2	78	
3.	52	3	55	$= \frac{59}{100} = 0.6$
	$\frac{168}{3}$ 56	8/3 = 3	$\frac{176}{3}$ = 59	

B. Sample 10 (0.1ml) plated after 5.25 hours

1.–6 no growth

		(−)	(+)	
361-7	7.	5	1208	
stored 361-8	8.	1	462	← this seg tested + 0 + Lp^S – Gal – Lp^S ↑ 4 $+^R$ obtained, 3 full +, 1 intermed. 4/4 not segregating
361-9 (tested + three out of 100 were)	9.	63	648	
	On a plate streaked from 10.	ca 19	1/16 ca 185	

This a colony – almost pure (−) noted = ①

Analysis of this colony is on 362-B

Reversions – the (−) seg from 361-8, 361-9, 361B8 streaked on B gal and 4 $+^R$ obtained from each – examined for Gal stability – 10 tested

1+	2−	4−	Lp
+	−	+	S
+	−	+	R
+	−	+	S
+	−	+	S
+	−	+	S
+	−	+	R
+	−	+	S
+	−	+	S
+	−	+	S
+	−	+	S

Lp S 4 $+^R$ from each of 5 ~~Lp~~ Lp^S:
1. 3/4 intermed., all not seg
2. 3/4 int. (+), all not seg
3. 2/4 int. (+), all not seg
4. 3/4 " " " "
5. 3/4 " " " "

(2−S = 8)
2−R = 2
1. 3/4 int (+) 1(+) seg.
2. 3/4 int (+) 2(+) seg

2 Culture of W2869 $\frac{4^- 2^+ Lp^+ ?}{4^+ 2^- Lp^+ ?}$

A. Overnight cult. diluted $10^2 - 10^4 - 10^6 - 10^8 \rightarrow$ 10 0.1ml sample

0.1ml assay	+	−	total	
1.	18	7	25	
2.	22	8	30	0.33/0.1ml
3.	22	20	42	
	62, 3 = 21	35, /3 12	97/3, 33	

B. sample 10 (0.1) plated 5.21 hours

1.–5 no growth

		(−)	(+)	
	6–7	all (−) 1 c	ca 2000 col	1 c ca 500
361-B8	8.	4	361	← these 4 seg; 1+ 1 2− 0 4− result = 4− ~~[illegible]~~
	9.	14 = 66	ca 101	
	10.	ca 1500	ca. 700	

1. 4/4 int. (+) none seg
2. 2/4 int (+) none seg
3. 2/4 int. (+) none seg
4. 4/4 int. (+) none seg

SEE 362 ALSO

Using inocula from above – 0.1ml of 10^6 dil in each case added to 7ml Pen and incubated 20 hours at 42C

2868 inoculum ratio $56^+/3^-$ 20 hour ratios: $167^+/10^-$, $155^+/10^-$, $175^+/12^-$

2869 " " $21^+/12^-$ " " " 214/176 +?−/ +?−/

DATE: 8/24/55 REF:

Repeat study of segregation from $\frac{2^x 4^-}{+ \; +}$ Lp^+ - as on 361 - W2869

Culture streaked out and single (+) colony selected. Overnight culture from this colony diluted:

$10^2 - 10^4 - 10^6 - 10^8 \rightarrow$ 20 0.1ml samples incubated 4.5 hours, plated, ppt washed with 0.1ml broth

Cell assay (ΦI) made next day

	Sample	(−)	(+Hal)	allele ✓			(−)	(+Hal)
	1–5	no growth						
	6	0	354					
	7	1/4 = 157	ca. 300 × 4	discarded	2^-4^-	1	14	140
~~1(−) picked~~ →	8	19	1209	8 tested HFT + 8 +	+ 0 = 8(4⁻)	2	2	136
	− 9	2	464	both Gal⁻		3	16	118
	− 10	2	540	Gal⁻ HFT +	2^-4^-		32/3 = 11	394/3 = 131
~~8(−) picked~~	11	19	1089	4 tested 1 / 3	+ 0 + = 1 2⁻ / + + 0 = 3 4⁻			
	12	17	1357	discarded			HFT + 5 4⁻	
6(+)	13	297	933	10⁻ picked	2 contain E(+)		8 + + 0	= 8(4⁻)
these are ⅞	− 14	4	428	4 Gal⁻				
	15	12	840	discarded				
	16	11	1525	"				
	17	33	796	discarded	7 12⁻ 14⁻			
12(−) picked 6(+)	18	441	690	"	12(−) + + 0 = 12 4⁻			
	19	494	1722	"				
	20	18	604	"				

<u>362B</u> the ⓟ colony of 361-9

1. the ⓟ colony streaked out gave almost all (−)

2. from #1 10 (−) colonies tested 7 12⁻ 14⁻ all 2⁻ Lp^s

from #1 4 Gal+ colonies obtained and streaked out - Examine (−) seg from each to test for secondary non disjunction - 6 Gal+ picked from each of the 4 and streaked to obtain 6 separate segregational events

Gal+ #	observation	single seg		
			?	s
1	4/6 no (−) seg	2/2 Gal_2^- Lp^R	2	0
2	1/6 no (−) seg	5/5 Gal_2^-, 2/5 Lp^s, 3/5 Lp^R	2	2
3	1/6 "	5/5 Gal_2^-, 2/5 Lp^s, 3/5 Lp^R	3	2
4	0/6 "	6/6 Gal_2^-, 2/6 Lp^s, 4/6 Lp^R	4	2
		6 Lp^s + 12 Lp^R	12	6

DATE: 8/25/55 REF:

Observation of erotic induction

1895 ⊗ 2790 Aerated, antifoam added, cultures 3 hours from overnight

1.0 ml + 5.0 ml + 4.0 ml Pen - aerated no antifoam (4.1×10⁹ Hfr / 2.08×10⁹ F⁻)

$10^2, 10^4, 10^6$ → 0.5 ml — 1. 206, 2. 206, 206

Samples taken — ml — diluted $10^2, 10^4$ — 0.05 ml plated — 1. /2279 on B Gal — 2. / D(o)

$10^2 - 10^4, 10^6$ → 0.05 ml — 1. 209, 2. 187, 396/2 = 198

Optical Density Read / 10 ml Pen at 650

Time	O.D.	Plaques/2279	Proto. D(o)	λ	λp
0	0.95	—	—		
15	—	7 *	3	35	contam. with phage
30	1.10	19	1	15	
45	1.7	36	0	—	
60	2.2	69	3	35	
75	2.0	147	11	115	
90	2.2	ca 170	6	65	

Ratio of Hfr/F⁻ : 20.6/99 = 1/5

412×10⁷ 4.1×10⁹ 4.00×10⁹

* since these plated on B gal estimation of 1895 Gal+ component can be made appear to be ca 100 at 0 and no reduction in number up to 90 min noted.

7×20×10⁴ = 140×10⁴ = 1.40×10⁶ λ/ml at Zero

150/7 × 1.4 = 29 × 10⁶ = 2.9×10⁷

21 = 1.4, 84, 21, 29.4

2.9/41 = 7%, Hfr

0.07, 41 | 2.900

2341 ⊗ 2308 Mal- Ind. 2341 to look for translocations and seg behavior of Clogents

1. 2341 aerated cult. irrad. 60 sec. - Mixed equivalent volumes of Hfr & F- incubated 30 min 37C diluted 1/200 and plated B gal, 5 Mal- 5 Mal plates discarded - difficult to split from Mal-.
2. 20 Gal+ prototrophs on 5 Gal picked and tested /2279 for lysogenicity all non lysogenic

The 20 Gal+ are divided into several groups on degree of Gal-ness
12 full Gal+ — these do not segregate
3 internal Gal+ } all appear to be seg.
5 weak Gal+ — these contain a few full Gal+ }

363 B →

DATE: REF:

Segregant diploidy. Test by reverting segregant, & stability

Culture.			Rev. stable	0 = no	+ = yes
1	518E1612		0		
2	518E750		0 (wk +)		
3	518E902-1		0		
4	518E902-2		0		
5	2175E811-1		0 (wk +)		
6	2175E811-2		—		
7	750E1612-1		0		
8	750E1612-2		0 (wk +)		
9	750E902-1		—		
10	750E902-2		0		
11	811E902-1	all +	no		
12	811E902-2	all +	0 (wk +)		
13	286-1		[illegible]		
14	286-2	failed to grow	—		
15	2175E750-1		0 (wk +)		
16	2175E750-2		—		
17	290-1		—		
18	295-2		—		
19	302-2	7+2−	—		
20	3448	2+6−7−	0		

10/10 successful tests no seg = 10 seg not diploid

311-2 = HFT 6− =

streaked out 2 Gal+ reversions obtained

both segregating Gal+/Gal− ← HFT 6− +R

/+ /2 /6

2 HFT seg obtained both + + 0 = 6−

309-1 = HFT 7−

streaked out 8 Gal+ reversions obtained

2/8 segregating Gal+/Gal− HFT 7−

2874 looking for Gal_2^- in 583 line

1. first 8 seg = Gal 7−
   2nd 6 " = "

DATE: 8/31/55 REF:

Segregation – non disjunction? Obtain clone with early seg and test the Gal+ component to see if it segregates 50% homogenote.

7. Culture. W2869 – from single (+) colony → overnight lec. dilute

10^7 – 10^6 – 10^4 – 10^2 (10 13.3/0.01ml)

→ 0.1 ml for Gal+ : Gal- ratio

↓ Spot 0.01 ml samples B Gal, 0.01ml incubate 3 hours and respread. – look for clones with many (–)

	Gal	total
1.	19	117
2.	17	142
3.	19	139
	55/3 – 18	398/3 – 133/0.1ml

(–)

Respreaddun

1. failed to grow or regrowth – 19 — $\bar{x} = 1.3$, $e^{-x} = 19.4$ — obs 19
2. are gal+ – 32 (clone size 10 to 20) — Gal+ $\bar{x} = 1.2$, $e^{-x} = 21.6$ — (19+6) = 25
3. are gal– – 6 — Gal– $\bar{x} = 0.18$, $e^{-x} = 59.76$ — 13 c Gal–
4. mixed Gal+ + Gal– – 15 (72)

Nos used for seg. studs

Discarded — 4 Gal+ picked to obtain clones — TEST OF ORIGINAL SEGREGANT COUNT

	total clone size	#			Discarded	Gal+ picked	1	2	0	
1	14	1.	(1–) : 13+	no other (–) on plate		(1) +	+	+	0	Gal₁–
	46	2.	8– : 38+	(–)	+					
2	11	3.	(2–) : 9+	no other (–) on plate		2 +	(2) +	+	0	2 Gal₁–
	45	4.	8– : 37+	same plate	+					
3	8	5.	(1–) : 7+	same plate		3 +				
	39	6.	15– : 24+		+		+	+	0	Gal₁–
	24	7.	10– : 14+	(–) same plate	+					
	41	8.	10– : 31+	(–)	+					
4	33	9.	(1–) : 32+	(–)		4 +	+	+	0	Gal₁–
5	11	10.	(1–) : 10+	(–) same plate		5 +	+	+	0	Gal₁–
	9	11.	7– : 2+	(–)	+					
	35	12.	13– : 22	(–)	+					
6	12	13.	(1–) : 11+	(–) same plate		(6) +	+	0	+	Gal₂–
	5	14.	4– : 1+	no other (–) on plate	+					
—	12	15.	1– : 11+	no other (–) on plate	+					

○ 2 Gal+ reversions selected from each – all found Gal stable

Localization of clone on plate and relationship to other Gal–, in case of spattering from one streak to another

Segregant from three Gal+

#6 col A = 1 (2–), 5 (4–)

B = 3 (2–), 3 (4–)

C = 2 (2–), 1 (4–) 1 no test

D = 1 (2–), 4 (4–)

tube agent +, 2–, 4–

Segregant from #1 Gal+ 1

A. 8 (2–) 5 (4–)

B. 2 (2–) 3 (4–)

C. 4 (4–)

D. 5 (4–)

STREET ↓

A1 (2–), D2-4 (4–) } A repeat: 1-4 (4–) Σ 3 4–; 6 seg analysis: 5-6 (2–) 3 2–

B1 (4–), B2-4 (2–), B5-6 (4–)

Gal+ Reversions (2) obtained from these and tested for stability

2 lev of A1 unstable – (16+) col. tested / 750 — 4 contain (+), 5 NFT, 7 HFT

A2-A6 stable, B1 (–) stable, ½ B2-4 unstable, and stable B5-6

10(+) col. tested 1 HFT / 811

– NFT segregant obtained 6/6 Gal₂–

10^{13} /80 plated

Gal

C1 – (4–), C2-3 (2–) } C repeat: 1 = 4–, 4 (4–); 5 seg anal.: 2,3 = 2–, 4,5 = 4–, 4 (2–) — 6/6 stable

D1-2 (4–), D3 – (2–), D4-5 (4–) → 10/10 stable

DATE: 9/25/55 REF:

A

293-12 (which are) gives increased frequency of HFT Gal$_4^-$
Use to test for dispersal of distal markers as a result of C.O. between Gal's.

1. A mal$^-$ obtained by λ^2 resistance on B mal.
2. 12 seg. obtained, tested for HFT against 2580, 8U

control	2580	8U
o o ⊙ o	o o o ⊕	o o ⊙ o
o o o o	o ⊕ o o	o o o o
o o o o	o ⊕ o ⊕	o o o o

⊙: contam
⊕ = HFT

4/12 HFT 4$^-$ malt populace

Total
13 HFT 4$^-$
37 +
17/49 or 16/49

(often mal +^{R} Gal allele)

3. Streak HFT 4$^-$ on B mal. v mal +^{R} stbl – from seg. colonies
#4 – 6/6 malt^R stbl, 3/3 Gal+^{R} stbl; #6 6/6 malt^R stbl, 3/3 Gal+^{R} stbl; #10 6/6 mal+^{R} stbl, 3/3 Gal+^{R} stbl; #12 6/6 malt^R stbl, 3/3 Gal+^{R} stbl

A1

4. Test these seg /2$^-$ /4$^-$ /+ (these are mal$^-$)
A Despite mal reaction – after 3 days at 37C, reactions obtained – all appear to be transduced by +, 2$^-$, but not by 4$^-$ – means all Gal$_4^-$ – Gal$_4^-$ is in exogenote
B. Reversions of #4, 6, 10, 12 obtained to check HFT.

The HFT above (A1) and 3 from A2 grown with 1177 24 hours and plated on SMal Sm – no growth – indicating these cultures not Hfr.

Reversions above say cultures not homogenote. Why?

5. Rpt. on 12 additional seg.

control	2580	8U
o o o o	o o ⊕ o	o o o o
o ⊙ ⊙ ⊙	o o ⊕ o	o o o ⊕
o o o o	o o o o	o o o o

A2 (the Gals are from Hfr? mal^R)

	#3	#6	#7	#8
A2 Malt^R	6/6 +^{R} stbl #3	6/6 +^{R} stbl #6	6/6 +^{R} stbl #7	6/6 malt^R #8 stbl
Gal +^{R}	5/6 " "	5/6 stbl	6/6 " "	2/2 stbl 4
	1 unstable	1 unstable		

CONTINUED PAGE 366

6. 293-12 irradiated on Bxyl, Bara, Bmlt. – 1 xyl$^-$ obtained, 1 mtl$^-$ (?) not Gal+/Gal$^-$

2852 = 365B = Gal$_1^-$ Gal$_4^-$ × 2$^-$ to obtain (– – –) for expt. to show crossing over between genotes, as: $\frac{+}{2^-}$ | 1^+ 4^+ = not Gal+, c.o. between genotes to give $\frac{+}{+}$ yields Gal+.

B

1. 2852 streaked / HFT 2$^-$.
2. 10 streaks made from cross-lined.
7 yielded 4p$^+$ from 8.
3 " 4p^R/4p^S same seg 1$^-$ 4$^-$ 4p^S
3. Obtain 1$^-$ 4$^-$ 52p^S]] 1$^+$ 4$^+$ 2$^-$ 4p^r (one)
a. seg. 11 1$^-$ 4$^-$ 4p^S
1 2$^-$ 4p^S
1 2$^-$ 4p^r → sweated B gal → 7 seg obt. 6 = 1$^-$ 2$^-$ 4$^-$ 4p^S, 1 1$^-$ 4$^-$ 4p^S

→ Reversion 365B-1 and 365B-2 next

DATE: 10/1/55 REF:

1. 1578 – to obtain F⁻ 1⁻4⁻ and F⁻ 1⁻ for EML

1578 X — 2346 X (Gal₁⁻) – Exposed 0.1 ml O.N. 1578 to 0.3 ml 2346 – incub. 10'
dilute 10⁻², 10⁻¹, 10 0.1 ml / Plate – No contra-selection.

	Plate	(–)	2 days pos.	
Segregation from #1	1.	14	0	366-1
11 (stbl)(+)	2.	15	2	(1 mosaic appearance)
12 seg.	3.	17	1	366-2
Segregants tested	4.	12	1 lost	
1 1⁻4⁻	5	8	0	
6 4⁻		66	4	(90) 4/70
1 1⁻			contrl = 3, no lost	
2 P.E.				

2 ml to tubes — 366-2 stock gal⁻ duplicated Sept 1961

F⁻ Gal₁⁻ Gal₄⁻ Lys⁺
= 366-3
W2994

K12 lysate 9/28/55

Assay 10/1/55 = 108 × 10⁸ – 1.1 × 10¹⁰
10/4/55 = 108 × 10⁸ – 1.1 × 10¹⁰

365A3 continued. 293-12 mal – → seg. tested for mal diploidy, etc.

A3 9 additional seg.

	/2550	/811		/2550	/811
HFT test			Not		
phenotype of clone	o o ⊕ o	o o o o	individual	o o ⊕ o	o o o o
exceptions no different from	o ⊕ o o	o o o ⊕ 20		o ⊕ o o	o o o ⊙ 2
non exceptions or B gal →	⊕	o		⊕	o

This growth, streaked out formed +, also mosaic colonies

THESE APPARENT HFT SEGREGANTS ARE OBSERVED WITHOUT UV TESTING.

these were not numbered. 4 mosaics obtained from 3 were from one. Which?

A3 Gal⁺ᴿ ← of above (1) 2/4 +ᴿ seg; (2) 1/1 +ᴿ seg; (3) 3/4 +ᴿ seg.

	Malt⁺ᴿ from Gal+/–	Malt+ Gal+/– ?	→ Gal–	gave together Malt/Gal+/–
#1	6/6 stbl	6/6 Gal–	6/6 stbl	
#2	5/5 stbl	2/5 Gal+/–	6/6 stbl	
#3	5/5 stbl	1/5 Gal+/–	6/6 stbl	

Do these seg. heterogenotes have a greater tendency to fall apart? Should have effect on seg? Of 16 malt⁺ᴿ, only 2 were stable Gal +/–

Return to 293-12 – are the cosegregants really Gal₂⁻? 365 suggests no.

/2550	/811
⊕ o o ⊕	o o o o
o o ⊕ o	o o o o
o o ⊕ o	o o o o
o o ⊕ o	o o o o
o o	o o

tested against HFT 2⁻, 4⁻ all Gal₄⁻

DATE: 10/8/55 REF:

1	2	3	4	5	6	7	8	9	10

2341 Lp^r/Lp^s 2/2 x —— (+) 750 C112-1

1. 2 day cult. 2341 → 1-10 ↓ 0.1ml + 0.1ml 750C112-1 → 10^2, 10^4 → 0.1ml inc. 10' 37C

2. same with broth as control ——→

		(−)	(+)
Control	1.	150	0
	2.	164	0
Expt.	1.	173	1
	2.	158	2
	3.	162	2
	4.	123	2
	5.	189	2

} (2-4) several partially lysed colonies/plate

One streaking test Gal. /λ, 2271

	Gal+	Gal−	1st strk
1	Lp^s	Lp^s	→ Lp^s+, −s
2.	Lp^r	Lp^r	→ Lp^s+, −s
3	Lp^s	Lp^r	→ Lp^s+, −s
4	Lp^s	Lp^r	→ Lp^s+, −s
5.	Lp^r	Lp^r	→ Lp^s+ −r
6	Lp^+	Lp^+	→ Lp^s+, −+
7	Lp^+	Lp^+	→ Lp^s+, −+
8	Lp^+	Lp^+	→ Lp^s+, −+
9.	Lp^+	Lp^+	→ Lp^s+, −+

(−), (+) picked tested (λ, 2271

[These discarded]

A

365B2 = M− Gal1− Gal4− Gal2− Lp^s x —— HFT 7 to see quadruple (−)

1. Procedure as with 2341 above. 309-1 lysate used.

			(−)	pap. col.
1.	Control	1.	428	0
		2.	454	0
		3	503	0
2.	Expt	1.	171	15
		2.	189	27
		3.	196	20
		4.	206	33
		5.	200	46

} These are low, perhaps 30% most (90%) colonies partially lysed

B

$N_0 = 43 \times 30 \times 10^3 = .6 \times 10^{?}$

$N_s = 2 \times 20 \times 10 = 4 \times 10^2$

DATE: 10/10/55

REF: $\frac{N_s}{N_0} = \frac{4}{.6} \times \frac{10^2}{10^?} = 0.5 \times 10^{-2}$ $= 0.5\%$

241-14 – (a colony picked for HFT) HFT λ yield

1. dilute to obtain 10^4–10^5/ml saline (10, 10^3 serum) → dil, 10^2, 10^1 → orig.
2. Irradiate 60 seconds.
3. 1.0 ml + 10 ml Pen. ca 10^4/ml

↓ 0.05 ml sample → plated 1. 1 col. 2. 3 col ← forgot to add 3. 3 col./7/3 – 2 all lost!

N_0 = 1. 36 (1 year) 2. 40 3. 53 — 129/3 = (43)

4. /750 — PAP /2279 — PLAQUES /2279

Time	/750	/2279 PAP	/2279 PLAQUES		
0 (1:50)	52	79	>62	Plaques small	4. 33
15	30	81	>168	″ ″	5. 35
45	32	93	>107	″ ″	6. 34
55	43	88	>280	″	102/3 (34)
65	50	86	>334	wet plate	
90	56	93	>374	″ ″	

} Replace

HOPELESS

SEE RESPREAD →

253/6 = 42

Gal × August 26 1964 Mc Card

For EML – a Gal⁻ in F⁻ 750 lin.

2902 × HFT 4

2 heterogenotes (P.E. –) obtained

368-1

368-2 ← 4p°/4p°

plaques → Gal⁺ 2902/4

18/22 + ~~stab~~

↑ 13 Gal⁺ lysog to λ 2279 3 4p°, 10 4p°

4 any del. 3 eb. del. 1 P.E.(–)

2915 × HFT 7 (309-1) – Do 4p°/4p° heterogenotes come from clones were there 11 also (4p+)λ⁺? Recovery of 4 strands.

1. Freshened culture of 2915 ca. 10^8 ~~dilute~~ 0.1 ml + 0.4 ml 309-1 λ (prep. incubated 10′ 37°C, dil 10^2, 10^4, 10^1 → 0.1 ml plated B Gal

		(+)	(–)	(–)/ly sec	total
1. Broth control	1.	0	92	0	92
	2.	0	94	0	94
	3.	0	96	0	96
2. Lysate spotted.		lysed colony			
	1.	4 (3)	ca 80 wet plate	12	ca 16
	2.	3 (2)	ca 60 wet plate	14	ca 77
	3.	6 (6)	65	10	71
	4.	2 (1)	41 wet plate	9	52
	5.	5 (3)	ca 62 wet plate	ca 16	ca 83

CONTINUED →

3. testing the heterogenotes – streaked / 2902, streaked out B Gal.

	2902 lysogenicity				
1	+	Gal seg	11	+	Gal-
2.	+	″	12	+	Gal seg
3	+	″	13	+	″
4	+	″	14	+	″
5	+	″	15	+	″
6	+	Gal-	16	+	″
7	+	Gal seg			
8	+	″			
9	+	″			
10	+	″			

} these last 4 came from intact col., the others from partially lysed colonies.

DATE: 10/18/55 REF: 368 A

(bridge)

Continuation of 368 - Do lp^{r}/lp^{s} transductions come from λ^{+} particles?

1. Examination of the heterogenotes - 6 gal+, 6 gal- taken from the 1st streaking out of the isolated (single colony) heterogenotes, and streaked against λ, 2279

		Het # 1	2	3	4	5	7	8	9
Gal+	1	+	+	+	R	+	+	+	+
	2	+	nt	+	R	+	+	+	+
	3	+	+	+	R	+	+	+	+
	4	r ?	+	+	R	+	+	+	+
	5	+	nt	+	R	+	+	+	+
	6	+	+	+	R	+	+	+	+
Gal-	1	+	+	s	s	s	+	+	+
	2	s	s	+	s	+	+	+	+
	3	s contam	s	+	s	s	+	+	s
	4	+	+	nt	s contam	nt	+	+	nt
	5	nt	+	+	s	nt	+	+	+
	6	s contam	s	+	s	nt	+	+	s contam

+ = lp^{+}, r = lp^{r} s = lp^{s} nt = no test

Reason of plate on which these were tested mostly shows good lysis

Re. #16, the plate shows self lysis and [illegible] plaques.

		#10	#12	#13	#14	#15	#16	#18
Gal+	1	+	+	+	+	+	R	+
	2	+	+	+	+	+	R	+
	3	+	+	+	+	+	R	+
	4	+	+	+	+	+	R	+
	5	+	+	+	+	+	R	+
	6	+	+	+	+	+	R	+
Gal-	1	s	s	+	nt	+	s	+
	2	s	s	+	s	s	s	+
	3	+	s	+	s contam	s	s	+
	4	+	+	+	s	s	s	+
	5	+	s	+	s contam	+	s	+
	6	+	s	+	s	+	s	nt

(SEE 371)

FOR #4 #16: The original colony (from the first plate) grown up in broth and spotted (post centrifugation) on 2279. One set of plates irrad., one not. Both showed λ, the irrad. = 0 induced. (?)

FOR A Reddish (?)

To AR. new (W) Hfr 3 x Gal₂⁻ (241-14) W

also Gal₄⁻ (247R-1) x

From the Gal₂⁻ x 4 Gal₂⁻ Blend. 3 lp^{s} 1 lp^{r} were found lp^{+}