4/1 Aerated cultures of (5:30 AM)

Wg16 pr⁻ – for pen. run

K-12 – λ trip.

1655 – for transduction retrial

Culture of K12 for NZ phage effect (Boyd)

K-12 L4?

Tube.	1	2	3	4
λ dil	1-10	1-100	1-1000	1-10000
ml K-12 overnite culture	1.0	1.0	1.0	1.0
Pen.	10	10	10	10

In both at 9:45

at					
10:15	granular appear	granular appear	turbid	turbid	overall turbidity the same
11:30	"	"	"	"	"
2:10	"	"	"	"	"
3:00	"	"	"	"	"

discarded

1655 + 882

16 colonies picked from streakings of 2/30 – tested against 1655

all appear to be phage free 4/2

1655 – Transduction

tube	1	2
(11:30 AM) 1655 cult.	5.0 ml	5.0 ml
λ L6	0.5 ml	–

Centrifuged – resuspend in saline (5.0 ml each) ~~[illegible]~~

~~[illegible]~~ + ~~[illegible]~~ Bucks soln. (PF stock dil. 0.1 + 10 ml)

Plate 0.1 ml on D(0)

4/2 – plate with phage has heavier background than without. due to exposure to broth

4/3 – " " " " "

discarded

K-12 L7

aerat. culture 1:15

centrifuged resusp in sal – mod 35 sec

Incubation at 1:45

cleared at 4:00

Eliminates requirement for DW-D buffer in dwff – use saline.

4/2

(52)

Wg 16 Pt - Pen run

Colonies

Plated 1-10 } EMB(0) -
and } -

Wg 14 pr - trypt - x 58-161
grown overnight in Pen in mixed culture.
plated out - D-O (two plates)

4 colonies on 1 plate
5 colonies on other -

K-12 on agar -

K-12 cells scraped off EMB(0) to Pen
Aerated 8:15 out at ———
Centrifuge resuspend in saline - irradiate after dil. to 10^8 cells/ml

1655 0.5ml + 1.0ml 1.5

Dose	Count/ml	Phage/ml		Survival	K-12 as plaques
0	256x10 = 2560	5x3x10 = 150	0	1.0	0.06
5	195x10 = 1950	11x3x10 = 330	180	0.76	0.13
10	123x10 = 1230	39x3x10 = 1170	1020	0.48	0.46
15	76x10 = 760	52(?)x3x10 1560	1410	0.30	0.61

1681 - Overnight culture - retested for rad. resistance and Luff.
Culture aerated (1.0+1.0ml Pen) 8:15 - Centrifuged
Dilute to

1681 cross streaked on 1655 - no plaques observed

Dose	Count/ml	Phage/ml	Survival
0	35	0	1.0
10	21	0	0.60
20	19	0	0.54
30	7	0	0.20
40	8	0	0.23 out

is this culture lysogenic? for 882?

0.54
35 | 19.0
175
150
140
100

Wq 14 pr⁻ Pen survivors -
replicated to D(0) +Pr and EMD(0)

4/7 Off 40± colonies picked and replicated & failed to grow on D(0) + pr - Wq14pr⁻x⁻ transferred to broth and D(0) + pr + tryptt.
48 additional survivors picked

4/8 Aerated culture of W267 in D(0) + Lac [0.1 ml (10%) + 10 ml D(0)]

1.0 ml overnight aerated D(0) + Lac culture + fresh D(0) + Lac (8:30 AM)

Centrifuge - resuspend saline · dilute to ca. 10^3 cells/ml

Dose	Counts EMB lac
0	1.
	2.
	3.
10	1.
20	1.
30	1.
40	1.

discarded apparently overgrown with lac + prototrophs

W518 λ^s

Aerated culture from overnight culture in Pen (8:30 AM) (1.0 ml + 10 ml)

Add 0.1 ml λ (ca 10^9 particles) to aer. cult (ca 1.5×10^8 cells/ml) Phage/cells ≠ ca 1.0

Incubate ~~20-30~~ 15 minutes.

und.	—	Plate on EMB lac ~~solid growth~~
Dilute 1-10	—	many colonies - all lac⁻, some pinkish showing lysis of cells
1-100	—	" " " " " " " " "
1-1000	—	few " " " " " " " "

W1931 $882^+ \lambda^s$ - Dread

Aerated culture from overnight unaer. 1.0 + 10 ml Pen (8:30 AM)

Centrifuge resuspend in sal. dil to ca 10^3 cells/ml

Dose	Count EMB lac	Survival
0	252	1.0
10	176	0.70
20	86	0.34
30	68	0.27
40	64	0.25

(more resistant than K-12)

4/9 Wed.

Induction of lysis in K-12 by cold shock?

Aerated culture 0.5 + 10 ml — 10:30AM to 1:30

Culture ca 5×10^8 cells/ml

Pre-shock

Cell titer $10^2 - 10^4 - 10^6$ → plate 0.1 ct 300 = 3×10^9

phage titer → 1.0 ml + 1.0 ml 518 $\xrightarrow{0.1}$ plate 2 = 2×10^7

Post shock

cell titer $10^2 - 10^4$ → plate 0.1 ml ct 3000? 3×10^8 (?)

phage titer $10^2 - 10^4 - 10^5$ → 1.0 ml + 1.0 ml 518 $\xrightarrow{0.1}$ plate 4 = 4×10^6

Results not conclusive either way.

1831 10:30AM → 1:30

Centrifuged aerated cult. resusp in saline

Dilute to ca 10^3

Dose		Survival
0	242	1.0
10	138	0.57
20	33	0.136
30	15	0.062

U.V. resistance similar to that of K-12

1831+882 as 1831

Dose		Survival
0	1105	0.00
10	589	0.51
20	353	0.307
30	149	0.13

U.V. resistance appears higher than K-12

1831+882 cross streaked on 1831

no reaction

H267 - Original culture.

Streaked on EMB / EMS to recover diploid

discarded

Start again

W916 pr surviving replicated to D(0) 48

near of course — ... on D(0) + pr

4/10

Wq 16 pr survivors (see previous attempt) 48 in non
replicated to D(0) + pr -
1 failed to grow - transferred to start
for later identification - Wq 16 pr⁻ x⁻ ②

K-12 Repeat on induction of lysis in K-12 with cold.
Vigorously growing culture (two transfer in aerated Pen)
sampled immediately and placed in ice bath c̄ aeration 10 min
Sampled 10 min - returned to 37c

Immed. Cell count - 86
" Plaque " - 3

Post treat cell count - 116
" " plaque count - 1

no evidence of sensitivity to cold shock
no evidence of induced lysis.
no gross lysis
[illegible]

Wq 14 (Pr⁻) Aerated culture - washed twice in saline
ca 10^5 cells added to:
D(0) + pr + trypt + pen (0.2 ml) - plated out
D(0) + pr + trypt - growth

undil. = 14
1-10 = 0

4/12
Wq 14 (Pr⁻) Survivors of Pen run of above
replicated to D(0) + Pr
2 of 14 colonies possibly diaudotrophs
picked and streaked for further replication

4/14

Aerated cultures W67 and 1177 started 8:15

(57)

H267 Radiation - culture in D(0) + lactose 36 hours -
dilute to 10^3 cells/ml

Dose	Count EMB		Phage TSA 1655 (0.5ml + 0.5ml 1655)	Survival
0	341 316 365	cu 10 lac/plate	0	1.0
10	307	10 lac	7% 12 ·2 = 24	0.90
20	187		—	0.55
30	98		—	0.29
40	60		84 ·2 = 168	0.18

Wg 14 pr - Pen survivors - 2 possible diaux. - 10 colonies picked from streakings of each - streaked on EMB-D

replicated 4/15 to D(0)+Pr, EMB lac.

4/16 - situation same as before - apparent reversion in replica ⁘ D(0) ● EMB - Discarded

2 wgs of W67 + 1177

Cells centrifuged. resuspended in H_2O (10 ml)

Irrad. 35 sec

10 ml + 10 ml Pen

Incubation with air 1:30

cleaning at 4:00

partially clear following morning

4/15

(58)

Aerated cultures of Wg 14 pr⁻ for pen run
W1678 for sensitivity
W1682 " "
} 10:00 AM

W1678 Twoff and Survival

aerated culture above - dil to 10^3 cells/ml

Dose	Count/plate	Phage/plate (0.5 ml + 0.5 ml 1661)	Survival
0	338	0	1.0
10	259	—	0.77
20	107 (235)	143·2 = 286	0.32
30	40		0.12
40	17	—	0.05

Wg 14 pr⁻ Pen run -

aerated culture - centrifuged - resuspend in saline -
centrifuged, resuspend in saline.

add 0.1 ml to:

D(0) + Pr + trypt + 0.5 pen -
" " " —— -

H267 - culture in broth (from H267-I) 2.0 ml + 5.0 ml Pen (L1)

Incubate 60 minutes.
Centrifuge, resuspd in H_2O - mod. 35 sec - add 5 ml Pen
Incubate - 2:45 PM

partially clearing 4:00
" cleared next morning.

~~Wg 14 pr⁻ Pen run~~ -

4/16

Aerated cultures started

1661
1662 } 8:15 AM
1736

Wg 14 Pen Survivors
plated to EMB lac. – undiluted
plate 1 – >1000
2 – "
3 – "
– 40 colonies picked, tested – all grec 4/19, 4/20

H267 Culture #2
Dilute to ca 10^7/ml

Rose	+	Count/plate	Pure Lac⁻	Sag.	Survival
0		306	12		1.0
5		317			1.04
10		271			0.89
15		259			0.85
20		221			0.72
25		144			0.47
30		86			0.28

~~λ-titers~~ ~~EMB-01~~ titer

~~λ-K-12-LT~~ ~~Tab~~

H267 — 10^6 → 0.5+0.5 1661 → 0.1 — 2 — 4×10^7

W1177 same >134 $> 1.3 \times 10^9$

W67 " 0 : $< 2 \times 10^7$

Mod. of cultures – centrif. resuspd in sal. – incub. 35 sec. add 3.0 ml + 10 ml Pen – incubated
after 2 hours turbid → W1661 – plated on 1661 – no plaques by itself
~~[illegible]~~ ← no plaques on 1661 ~~[illegible]~~ by mod. of 1661

clearing – W1662 – $40-80 \times 10^7$

clearing – W1736 – $50-100 \times 10^7$

4/17

1736 – } 2 plates | 4/20 no(+) colonies | 4/4 = | 4/20 ca 50 papillae(−)

1736 + λ (0.1 ml) Lb } each | no+ colonies | = | ca 50+ papillae (−)(+)?

aerated K-12

4/20

H267 - Prep #2 Plated on EMB-maltose

dilute to ca 10^3 cells/ml - irradiate, count cells, plaque

Mol Seg or (-)	%	Dose	Count/plate	cells killed	Phage/plate (0.5ml + 0.5ml 1985) → 0.1ml		Survival
23	(5)	0	461	0	3 · 2 : 6	(1.2%)	1.00
Δ 39 62	(14)	5	445	15	4 · 2 : 8	(1.8%)	0.97
49 72	(21)	10	342	119	7 · 2 : 14	(3.0%)	0.74
65	(24)	15	274	187	38 · 2 : 76	(16.5%)	0.60
66	(25)	20	260	201	60 · 2 : 120	(26%)	0.56
85	(43)	25	196	265	140 · 2 : 270	(59%)	0.43

4/21 Wg14 pr- Pen survivors - (see previous)

(no auxotrophs among 40 colonies picked)

on 1 plate EMB lac a pink colony - picked and restreaked on EMB lac

(a lac+ Wg14?)

50 additional colonies picked

on each of the 3 survivor plates. 3-4 more opaque, raised colonies - picked and examined.

4/22

H267 above -

5 mol - colonies picked from each mod. unit.

and streaked on EMB lac to find chart.

0 5/5 lac, 1/5 appears mixed upon basis of opacity.

5 5/5 lac, 0/5 mixed

10 5/5 lac, 1/5 mixed on opacity of colonies

15 5/5 lac, 1/5 mixed on opacity of colonies

20 5/5 lac, 1/5 mixed on opacity of colonies

25 5/5 lac, 1/5 mixed on opacity of colonies

4/22

K-12 L8 Two 10ml Pen cultures (100ml + 10ml Pen)
aerated ~~[illegible]~~.
Washed & suspd. in saline. Inad 35 sec.
Add 10ml Pen - incubate with air -
Inc. 2:30 PM - out ca 5:00 cleaning.
Centrifuged on following morning - one appears viscous - K-12 L8 (viscous) 1
K-12 L8 (non visc) 2

Continuation of Transduction of W1736 by L6 of K-12

	1.	2.	4/25 1.	4/25 2.	Ave
4/23 Plates with 0.1ml Pen	83 colonies* (14 lac+)	91 colonies (4 lac+)	110 lac+	132 lac+	123 lac+
Plates with 0.1ml L6 of K-12	90 colonies 17 (lac+)	70 colonies (28 lac+)	152 lac+	151 lac+	152 lac+

*really papillae on heavy background growth
incubation continued.

Wg 14 pr - second step -
1. All 40 colonies picked after Penicillin selection grew - discarded.

2. of the lac+ Wg 14 pr - two colonies picked and streaked EMB lac-
on 4/22 - 1st day doubtfully lac+
on 4/23 - 2nd day strongly lac+
lac+ #1 #1 smooth glistening raised
lac+ #2 #2 rough dry, odd scattering appearance ● - segregation of lac-? delayed fermentation?
lac+ #3
lac+ #4
Both transferred to agar slants from single typical colony.

3. Papillating and lac+ colonies tested in D(o) + pr + trypt } ten colonies in all
D(o)
- all grew in supplemented and not in D(o)
- on the original streak plate - 3 colonies lac+ - appeared less + on second day after refrigeration - 2 appear to be the same (solid light purple) one mixed.
one of each type transferred to agar slants
→ lac+ #3 solid
→ lac+ #4 mixed

opaque mutants(?) forgotten -

Cultures started
1736 - transduction repeat
1485 - [illegible]
[illegible]

4/23 Wednesday -

Titration of K-12 λ8-1 and K-12 λ8-2

dilute 10^7 —0.5 ml→ + 0.5 ml 1485 —0.1 ml→

	plaques/plate	dil.	titer
K-12 λ8-1	105	$2 \cdot 10^8$	2.1×10^{10}
K-12 λ8-2	152	$2 \cdot 10^8$	3.0×10^{10}

- Aerated culture of 1932 started 9:50 - 1.0 ml + 10 ml Pen. out at 1:05 for Luria and survival. - dil to 10^3 cells/ml. used.

Time	Count/plate (EMB)	Plaques/plate	(0.5 ml + 0.5 ml 1485) TSA	Survival
0	107	0 0	0/107	1.0
5	92	0 0	0/107	0.86
10	60	11x2 = 22	22/107	0.56
15	(65) pipetting/plate	24x2 = 48	48/107	-(0.61)
20	33	53x2 = 106	106/107	0.31
25	8	64x2 = 128	128/107	0.075

mix up here - probably sampled twice at 10 - thereafter 15, 20, 25 samples are really 10, 15, 20

U.V. Resistance similar to K-12

- 1736 transduction - on EMB here

1736 cells from unaerated culture -

λ from K-12 λ8 Plate (EMB)	λ'	1736 cells	4/24	4/25	4/28		
1	0	0.1	no papillae	papillae appearing	30+ papillae	0	
2	0	0.1	"	"	26+ "	0	gal+
3	0.05	0.1	"	"	189+ "	186	25×10^3 gal+/ml = $\frac{1}{187}$ λ
4	0.1	0.1	"	"	274+ "	271	720×10^3 λ/ml
5	0.2	0.1	"	"	372+ "	361	

- Degradation/Survival of 1932 - aerated culture 3 hours - dilute to 10^6 cells/ml with saline.

Time	Counted EMB	Plaque/ml (0.5 ml + 0.5 ml 1485)	Survival
0			
5			
10			
15			
20			
25			

W1821 } aerated cult. w/ at T=15
W1682 } T=0 + 10 min

T=15 plus

Thursday
4/24

(1.0 ml + 10 ml Pen)
Aerated cultures of 1682 and 1831 started 9:15 AM

- Irradiation survival, and Lwoff of 1682 (λ^+ 882$^+$)
  Aerated cultures.
  Dilute to 10^3 cells/ml (Plaques = 0.5 ml + 0.5 ml sens. cells) → 0.1 ml

Dose	Col/Plate (EMB loc)	Plaques on 1485 (= λ+882)	on 1831 (= 882)	Survival
0	134	0	0	1.0
5	125	—	—	0.93
10	83	—	—	0.62
15	50	0	0	0.37
20	33	0	0	0.25
25	16	0	0	0.12

Slightly more resistant than K-12 1/3
No lysis effect observed in the gross (see below) or by plate for either λ or 882 -
is λ really present?

- 1682 for gross Lwoff obs. - cells remaining from above centrifuged, resusp in sol. - read. 35 seconds.
  add 10 ml Pen - aerate.
  in - 1:10 PM
  no clearing 4:00
  no clearing following morning.

- 1682 λ^+ 882$^+$ cross streaked on 1831 λ^+ for the purpose of determ presence of 882 - Plaques and lysis of 1831 obs. indicating 882 present.

4/25 Friday

- Aerated cultures of 1485 and 1831 started 8:30

1 - Survival of 1485 λ^s — out at 10:45 culture diluted to ca 10^3 cells/ml c̄ saline

Dose	Count/plate EMBlac
0	20
5	46
10	50
15	2
20	15
25	12
30	7

This exp of no value - previously due to the friction or variability of EMBlac plates used - they were poured 4/24 - avoid in future.

- Survival and Lw off of 1831 λ^+ - aerated cult. diluted to 10^3 cells/ml

Dose	Count/plate EMBlac	Plaques 0.5 ml + 0.5 ml 1485 on 1485
0	63	1 x 2 = 2
5	41	–
10	27	–
15	5	40 x 2 = 80
20	3	24 x 2 = 48
25	5	28 x 2 = 56

not a good exp. can be said that 1831 undergoes Lw off -

- Streak - 1682 colonies across 1681 in order to determine if λ is present -
  5 colonies picked from 0 plate of 4/24 irrad.
  none showed evidence of ability to lyse 1681

+ H267 - Culture #3 dilute to 10^3 cells/ml - irrad. for Survival - also to attempt isolation of reconstituted dip.

Dose	Counts/EMBlac plate	S.F.
0	192	1.0
5	134	0.70
10	27	0.14
15	19	0.10
20	4	0.02
25	2	0.01

unusual survival curve - sensitivity

more sensitive than H-12

Monday 4/28

λ transduction of 1736 of previous pages

# papillae recorded on original page.	4/29	4/30	Replicad to galt	5/1
10 papillae (+) from control, picked, streaked EMB-loc	7/7 loc	7/7 loc		6/7 galt
17 " " " O/ml λ plate " " " "	17/17 loc	17/17 loc		16/17 galt

exceptions may be due to heavy contam with baked cell of original plate.

4267

5 (+) centers from previous incid. picked and streaked on EMB loc, EMS loc

streaking are lact on EMS loc
either mixed or segregating on EMB loc

Cultures
1485
1736
W914 pr-typ

Tuesday 4/29

Wg 14 pr - trypt -
diluted and plated out for lac+ reversions -

	plates EMB lac -	#30	3/1
10^6 →	#1	ca 150 (-)	∞ 4/30
	#2	"	"
	#3	"	"

- W1736 - mad - Lwoff, and examination of plaques for presence of phage other than λ
- aerated culture, dil to ca 10^7 cells/ml

Dose	Colonies/EMB plate	Phage (0.5 ml + 0.5 ml 1485)
0	18	2
5	20	—
10	6	2
15	5	3
20	7	8
25	3	7
30	5	—

insufficient col count - appears odd in addition

- H267 Repeat on survival of prep #3

Dose	Colonies/EMB lac plate	SF.
0	296	1.0
5	168	0.57
10	106	0.36
15	92	0.31
20	22	0.07
25	4	0.01
30	6	0.02
35	2	0.007

odd? appears to check previous run with this prep re sensitivity over grown c̄ lac+ prototrophs?

lac - col.	seg. col.
8	48 (?)
46	74
32	46

- Penicillin runs

Wg 14 pr - trypt - → Pen aerated culture → wash → D(0) + pr + trypt aerated cult → add ca 10^6 cells/tube

tube 1 - Pr + trypt + D(0) + Pen (0.2 ml) - 4/30 no growth apparent
2 " " " - showed no growth - why?

Wg 16 → Pen aerated culture → wash → D(0) aerated cult → add 10^6 cells/tube

tube 1 D(0) + Pen (0.2 ml) 4/30 no growth apparent
2 D(0) growth

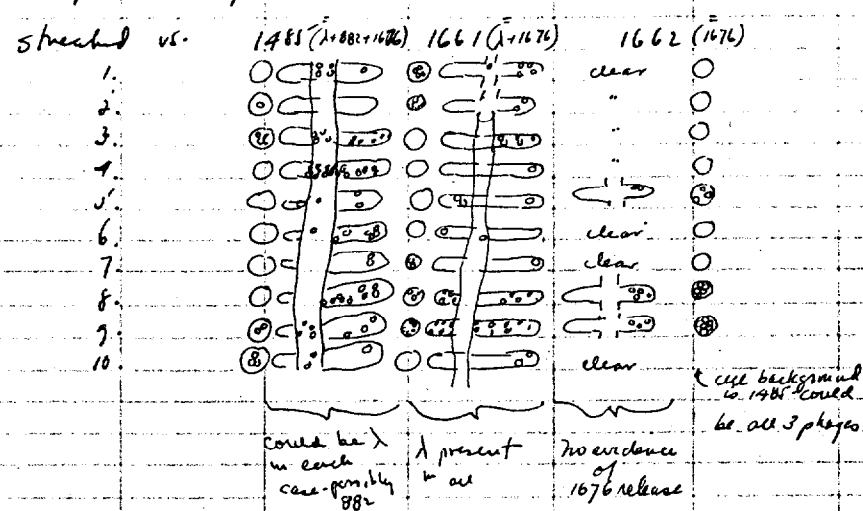
W1661
W1662
W1485

Wednesday 1/30

- Lac+ papillations from the last attempted transduction of 1736 replicated to gal EMB.
- Plate out

		NSA
Wg 14 pi - trypt -	undiluted	>
	1-10	>
	1-100	150-200 col (was something lacking in lac tube? - control failed to grow
Wg 16	undil.	16
	1-10	2
	1-100	–

- Plaques following 1736 mad.
  10 picked to pen broth (0.2 ml) - numbered



Thursday 5/1

- titer of K-12 λ L7
  dilute 10^7 plate 0.1 ml $10^8.2$ 1485 → 78 = 1.6×10^{10}

- replica Wg 16 pen survivors to D(0) on mid 12/18 failed to grow } indicating
  on 1/10 2/2 failed to grow } anaotrophy

- store Wg 14 pr-trypt - until D(0) + Supplements can be made -

- λ from K-12 L7 + L8 pooled and filtered - giving around 50-60 ml
  total volume
  dilution 3, 4, 6, 7 $\xrightarrow{0.1\,ml}$ 427
  ?
  titer = 4.3×10^{10}
  or if dilution error 4.3×10^9

- K-12 - day old aerated culture in D(M) + lac (10% 0.1 ml) - diluted to 10^3 cells/ml - mod.

Dose (sec)	Col/EMB lac plate	Plaques on 1485 (0.5 ml + 0.5 ml 1485)		Survival	K-12 as plaques = %
0	373	-		1.0	-
5	282	-		0.76	-
10	234	-	total	0.63	-
15	197	89.2 = 178	375	0.53	0.48
20	149	-		0.40	-
25	108	138.2 = 276	384	0.29	0.74
30	86	-		0.23	-

- H-267 as K-12 above.

Dose	Col/EMB-malt plate	Plaques on 1485 (0.5 + 0.5 ml 1485)		Survival	K-12 as plaques = %
0	285	-		1.0	
5	277	-		0.97	
15	168	-	total	0.59	
25	116	86.2 = 172	288	0.41	0.60
35	59	116.2 = 232	291	0.21	0.82

Cultures.
776 - 882 (882)
1485
H267 from H267 #1 D(M) + lac
K-12 in D(M) + lac

Tuesday
~~Saturday~~ 5/6

- Aerated cultures 8:00 1503 λ^+
  1808 Wg 31
  1959 Wg 31 λ^+
  K-12

- K-12 Phage plating on 518 - For possible recovery of both phage and irradiated cell

Dose	Count/EMB lac Plate	Phage/518 plate (0.5+0.5 ml 518)		Survival	%K-12 as plaques
0	206	—	total	1.0	—
5	79	—	phage+cells	0.38	—
10	53	58.2 = 116	169	0.26	0.56
15	27	55.2 = 110	137	0.13	0.53
20	18	68.2 = 136	154	0.09	0.66

- 1954 = Wg 31(λ^+) - Observation to see if λ is twoff-able in different strain

Dose	Counts/EMB lac Plate	Phage/518 plate (0.5+0.5 ml 518)		S.F.	1954 as plaques
0	216	—	total	1.0	—
5	163	—	phage+cells	0.76	—
10	102	52.2 = 104	(206)	0.47	0.48
15	8	120.2 = 240	(248)	0.046	1.0
20	10	107.2 = 214	(224)		1.0

- 1972 = $\lambda^+\Omega^+$ Examination for twoff and Recombination of λ and Ω

Dose	~~Plaques~~ on EMB lac	Plaques on 518 (0.5+0.5 ml 518)	Plaques on 133
0	8	—	—
5	8	—	—
10	2	10.2 = 20	—
15	0	18.2 = 36	—
20	0	11.2 = 22	0

No apparent reason for low cell no. - apparently large amt of free phage (= λ) in cult. turbidity was expo to 5-10 [illegible]

- Transduction of 1736 - Gal
  Unaerated overnight cult of 1736 in Pen
  + λ on Gal and Lac plates = 0.05 ml 1736 added/plate.

Not done?

#	EMB lac Plates	λ added	EMB gal Plates	λ added	5/8 Papillae
1	1	0	1	0	
	2	0	2	0	
	3	0.05	3	0.05	
	4	0.1	4	0.1	
	5	0.2	5	0.2	
	6	λ alone 0.1	6	λ alone 0.1	

- 2 Plaques 882/1485 picked to lightly inoc. aerated cult of 1503 - incubated overnight - turbid following day

- Cultures 518
  1898 } rad. sens.
  1655 }
  K-12 5 yr
  H267 (from #1) 5 yr

Tuesday 5/6

- Survival of 1503 - λ^s - aerated cult from overnight. dil to 10^3/ml

Dne	Cl/EMBlue plate	Survival
0	311	1.0
5	231	0.74
15	114	0.37
30	28	0.09
45	3	0.0097

Wednesday 5/7

- W518 λ^s Examination of u.v. resistance - Aerated culture - del. to 10^7 cells/ml

Dose	Col / EMB Blue Plate	Survival
0	327	1.0
5	186	0.57
15	91	0.28
30	40	0.12
45	8	0.024

- W1898 λ^s Examination u.v. resistance - same as above -

Dose	Col/EMBlue Plate	Survival
0	100	1.0
5	88	0.88
15	62	0.62
30	11	0.11
45	4	0.04

- W1655 λ^s Examination of u.v. resistance - as above

Dose	Col/EMBlue Plate	Survival
0	74	1.0
5	48	0.65
15	47	0.64
30	21	0.28
45	24	0.32

don't understand

- K-12 survivors(?) from Tuesday continued - D(●) plates

1485 u.v. sens
1736 trans
1177 trans

Thursday 5/8

- Transduction of W112 Lac - λ from K-12

EMB lac plate	λ from K-12	λ from K-12 boiled 10 min	W112 cells* unaerated overnight cult.	# papillae 5/11
1.	—	0.1 ml	0.1 ml	ca. 200
2.	—	0.1 ml	0.1 ml	ca. 250
3	0.1 ml	—	0.1 ml	ca. 300+ *
4	0.15 ml	—	0.1 ml	ca. 250+ *
5	0.2 ml	—	0.1 ml	ca 300+ *
6	λ from W1177 ⊔ 0.1	—	0.1 ml	ca 300+

* ~~[illegible]~~ plaques (50+) also on these plates.

Original cult. may be cont. - papillae not really (+) some pink with zones as if contaminated c̄ phage and lysis

- Transduction of W1736 gal - unaerated cult

EMB gal plate	λ from K-12	λ from K-12 boiled 10 min	No papillae (+) 5/11
1	—	~~[illegible]~~	17
2	—	0.1 ml	22
3	0.1 ml	—	324
4	0.1 ml	—	345
5	λ from W1177 0.2 ml	—	Thin plated dried up

λ from L6 probably

Gal 4 - $\lambda = \frac{3.0 \times 10^3 \text{ t/ml}}{2.0 \times 10^{10} \text{ λ/ml}}$ $2.1/10^{-7}$

- λ in 1736, 1177 } aerated cult. centrifuged - resusp in 10 ml sal.
mad. 40 sec. ~~[illegible]~~ 10 ml Pen - incubate with air 2:20 PM

- K-12 survivors in D(0) from previous - tests to phage + cells survive
mad. of K-12?
centers of 101 plaques picked and streaked on D(0) agar
on 4/8 1 streaking shows good growth - incubation continued

4/9 1 " " " "
2 " show single colonies

5/9 Friday -

- Transduction of 1736 on gal with λ from K-12 page 69

Plate	λ added	no gal + papillae
1	0	38 } 25
2	0	12
3	0.05 ml	153
4	0.1 ml	302 ← $\frac{2.75 \times 10^3 \text{ t/ml}}{72.0 \times 10^{10} \text{ λ/ml}} = 1/1.3 \times 10^{-7}$
5	0.2 ml	620
		L6?

Gal 4 -

why the diff.?

Transduction W1736 on lactose with λ from K-12 page 69

Plate	λ added	
1	0	201 } 185
2	0	171
3	0.05 ml	262
4	0.1 ml	344
5	0.2 ml	600

Should be noted that papillae counts are lower than real value since counting is difficult and many small papillae are skipped -

- Plate 1 of 1736 Transd on lac - 98 papillae picked to gal - 26 grew - 24/26 were gal -

- Plate 5 of 1736 transd. on gal - 98 papillae picked to lac - 22 grew 22/22 were lac -

This finding suggests that papillae on EMB lac control are different from papillae on EMB gal - that the gal transduction effect does not now appear as a release of negative inhibition - against gal plus colonies

lac plates were discarded unfortunately -

Monday 5/12

- K-12 Synthetic culture #2 - dil to 10^3 cells/ml

Dose	Count/EMB Con plate	Survival
0	330	1.0
5	220	0.67
15	211	0.64
30	155	0.47
45	83	0.25

more resistant than previous

- H267 Synthetic culture #4 dil. to 10^3 cells/ml

Dose	Count/EMB Con Plate	Survival
0	215	1.0
5	222	1.0
15	148	0.69
30	50	0.23
45	14	0.065

different from previous

- Titration of K-12 λ L7 + L8

dil	0.05 + 0.5 ml 1485
10^8	67
10^8	68

$68 \times 2 \times 10^8 = 1.3 \times 10^{10}$

-

Tuesday 5/13

- Titer of 882/1485 on 1485

dil. $10^2 \to 0.5 + 0.5$ 1485 $\longrightarrow$ no plaques
$10^4 \to 0.5 + 0.5$ 1485 $\longrightarrow$ "
$10^6 \to 0.5 + 0.5$ 1485 $\longrightarrow$ "

- Transduction of Serine or Glycine in 1678

	D(0) plate	1678*	heated λ	λ	5/14	5/15
air + pr	1	0.1	–	–	colonies around spot of deposit of pr	contam.?
	2	0.1	0.1	–		1 col
	3	0.1	–	0.1		3 col

* unaerated culture

- Titration of L2 of 1736 - from usual exp. - centr., resp'd sal, mad 35 etc. incubated E air overnight - nearly clear AM

dil $10^8 \to 0.5 + 0.5$ 1485 $\to$ no plaques

- Transduction of 1736 on Lac and gal (done on Monday 5/12)

Gal EMB plate	1736 Aerated cult ca 5×10^8 cells/plate est. λ from K-12 L7+L8	heat λ from L6	1736 Unaerated cult ca 10^8 cells/plate est. λ from K-12 L7+L8	heated λ from L6
1	0	0.1	0	0.1
2	0.1	0	0.1	0

Papillae on Gal 1 unaerated 5/15 = 25
Gal 2 unaerated 5/15 = 36

What happens?

Gal 1 aerated 5/15 = 29
Gal 2 aerated 5/15 = 425

$4.3 \times 10^{-}$ = 2 [illegible]
$2.[illegible] \times 10^{10}$ λ
gal 4 $\boxed{1/5 \times 10^{-6}}$

Lac EMB plate	Unaerated 1736 heated λ (K-12 L6)	unheated λ (K-12 L7+L8)	Papillae (-) not yet (+) 5/15
1	0.1		121
2	0.1		150
3	0	0.1	182
4	0	0.2	218

Little or no effect of λ on unaerated cultures of 1736 - fairly large effect on aerated cultures. Meaning not clear. Incubation of Lac plates continued to increase size of papillae to make picking more successful.

Thursday 5/6

- transduction of W1736 Leu - aerated culture (1.0+10.0 ml Pen) from 2 day unaerated culture - Cells centrifuged resp'd in saline

D(0) Plate + TB1	1736 cells	heated λ (L7+L8)	λ	colonies 5/17
1	0.1 ml	—	-	0
2	0.1 ml	0.1	-	ca. 50 (contam?)
3	0.1 ml	0.1	-	12
4	0.1 ml	-	0.1	0
5	0.1 ml	—	0.2	1 col
NSA plate 6	—	—	0.1	0

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H267 #4 Dilute to saline 10^4 - mod 5 seconds - Add 1.0 ml + 10 ml Pen/lac. Incubate with air at time 0

Time	Plate on EMB Mal	Pure Mal+ col	total no colonies per plate	Dark Mal+ colonies
Pre-mod 0'	1	10	168	5
Post-mod 0'	2	8	162	17
11:00 60'	3	29	256	14
11:30 90'	4	22	268	18
12:00 120'	5	23	302	16
12:30 180'	6	27	513	36

Thick plate reaction defined to are

- Ice box cleaned up.

- Culture on vial min. of 1832 Pen^r received from J.L. to ~~resolated~~ resolated - Pen broth inoculated - } 1 to DG, 1 to J.L. 5/17

- Wg 16 auxotrophs from page 68 picked to EMB plate, preliminary to determ requirements

- Galt papillae of 1736 Transduction of Page 74 - from aerated cult + λ
  35 picked to EMB lac for indentification

5/17 all grew - slowly - all appear lac^-, most spots are sectored light and dark (opacity) - some appear to be giving pink papillae.

instability of W1736

- Wg 14 pr- trypt - lac^+(?) col - streaked out

5/16 slight pinkish

5/17 dark red - probably slow lac^+

Slant made from single col -

Wg 14 pr- trypt- lac^+.