More

Exp I (1)

U.V. 10/5/51 Culture R (811)

Diluted culture from — hour unaerated culture. 0.C. aerated — hours at 37C. Prob. titer 1.5×10^8. Diluted 1:10.0 with Penassy broth blank. 11 ml vol. Shelter set at 30 -

Dose (Sec.)	Dilutions	Plates	Approx Col. Count	Titer	Survival
✓ 0	100, 10 →	R-0-4	300	3×10^7	1.0
✓ 20	100 →	R-20-3			
	↘ 10 →	R-20-4	400	4×10^6	0.133
✓ 40	100 →	R-40-3			
	↘ 10 →	R-40-4	400	4×10^6	0.133
✓ 60	100 →	R-60-3			
	↘ 10 →	R-60-4	30 *	3×10^5	0.10
✓ 80	100 →	R-80-3			
	↘ 10 →	R-80-3			
	↘ 10 →	R-80-4	150 *	1.8×10^6	0.06
✓ 100	10 →	R-100-2			
	↘ 10 →	R-100-3			
	↘ 10 →	R-100-4	10 *	1.0×10^6	0.03

* one lysed colony -

More (2)

U.V. 10/5/51 Culture S (518) Expt I

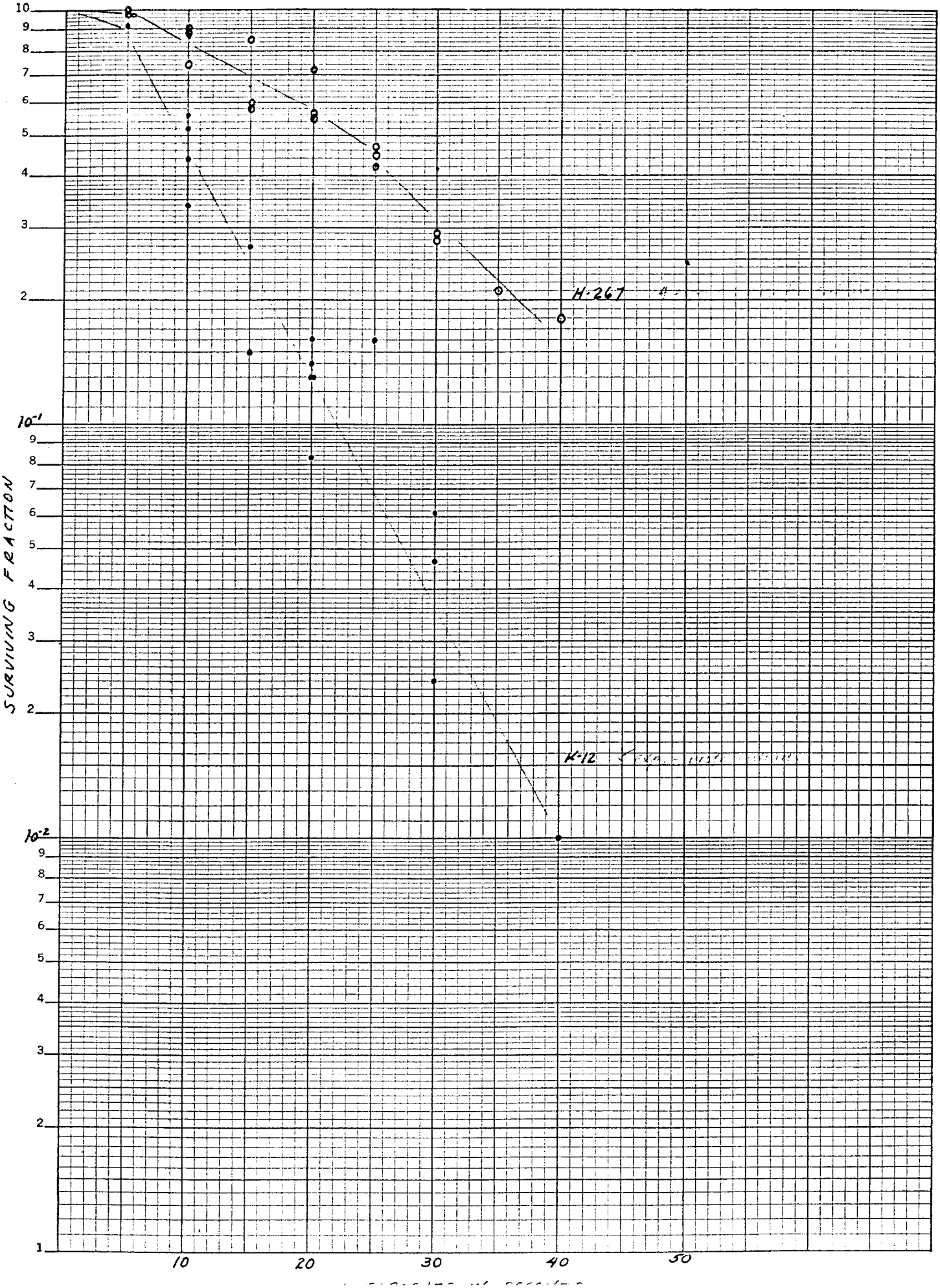
Diurnal culture from ____ hour macerated culture. 1.C aerated ____ hours at 37°C. Prod. Titer 1.5×10⁸. Diluted 1+100 with Penassay Broth - final vol. 11 ml volume shaken @ 30

Dose in Seconds	Dilutions	Plates	Approx. Col. Count	Titer	Survival
~~1×10⁶~~ ✓0	100, 10	→ S-0-4*	500	5×10⁶	1.0
✓20	100	→ S-20-3	~~400~~		
	↘ 10	→ S-20-4	400	4×10⁶	0.8
✓40	100	→ S-40-3			
	↘ 10	→ S-40-4	900	9×10⁶	0.8
✓60	10	→ S-60-2			
	↘ 10	→ S-60-3	300	3×10⁶	0.6
✓80	10	→ S-80-2			
	↘ 10	→ S-80-3	2500	2.5×10⁶	0.5
✓100	10	→ S-100-2			
	10	→ S-100-3	200	2.0×10⁶	0.4

* dilution = 10^4

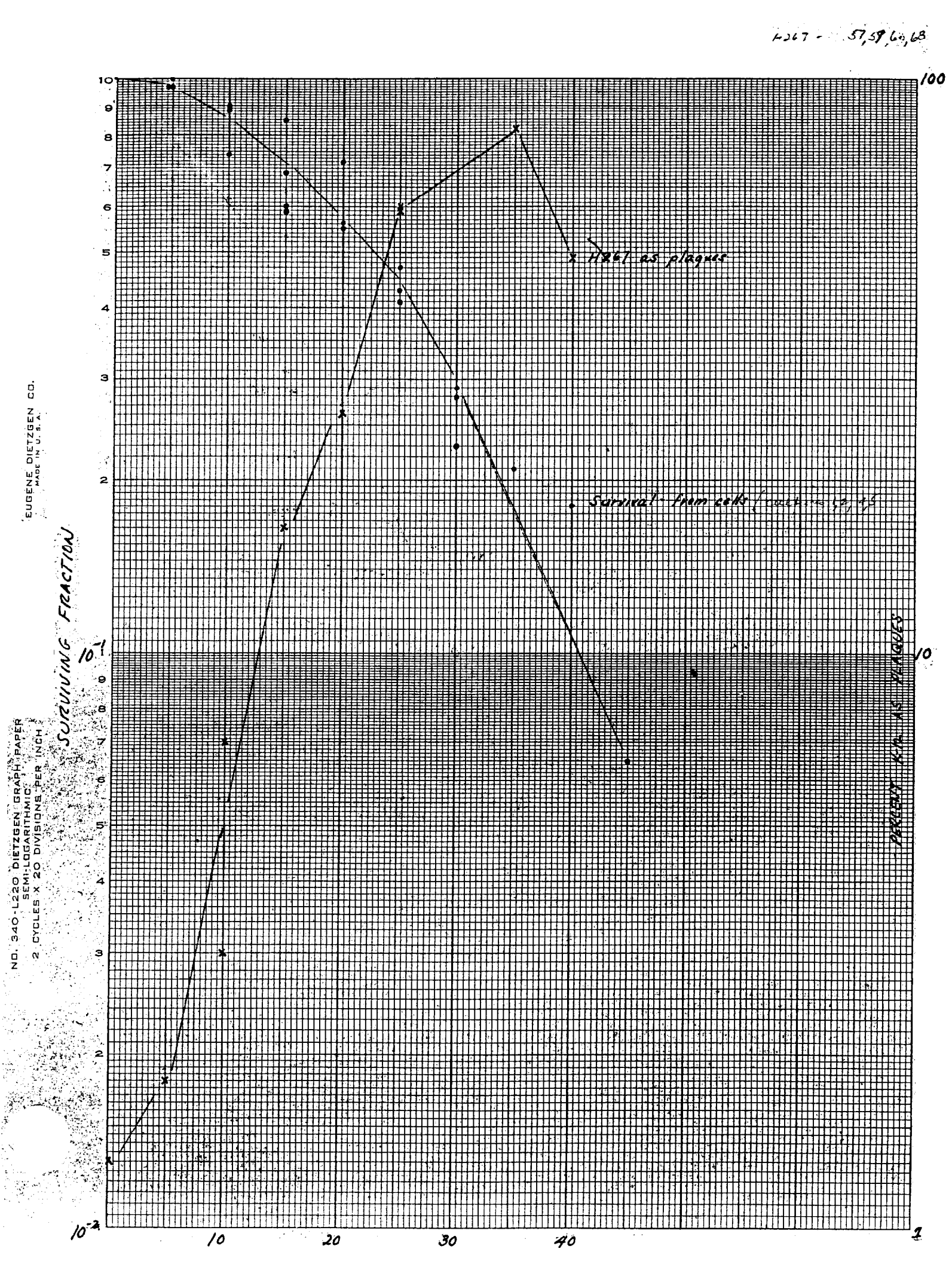
S-20-4

S = sensitive; 20 = exposure seconds; 4 = dilution



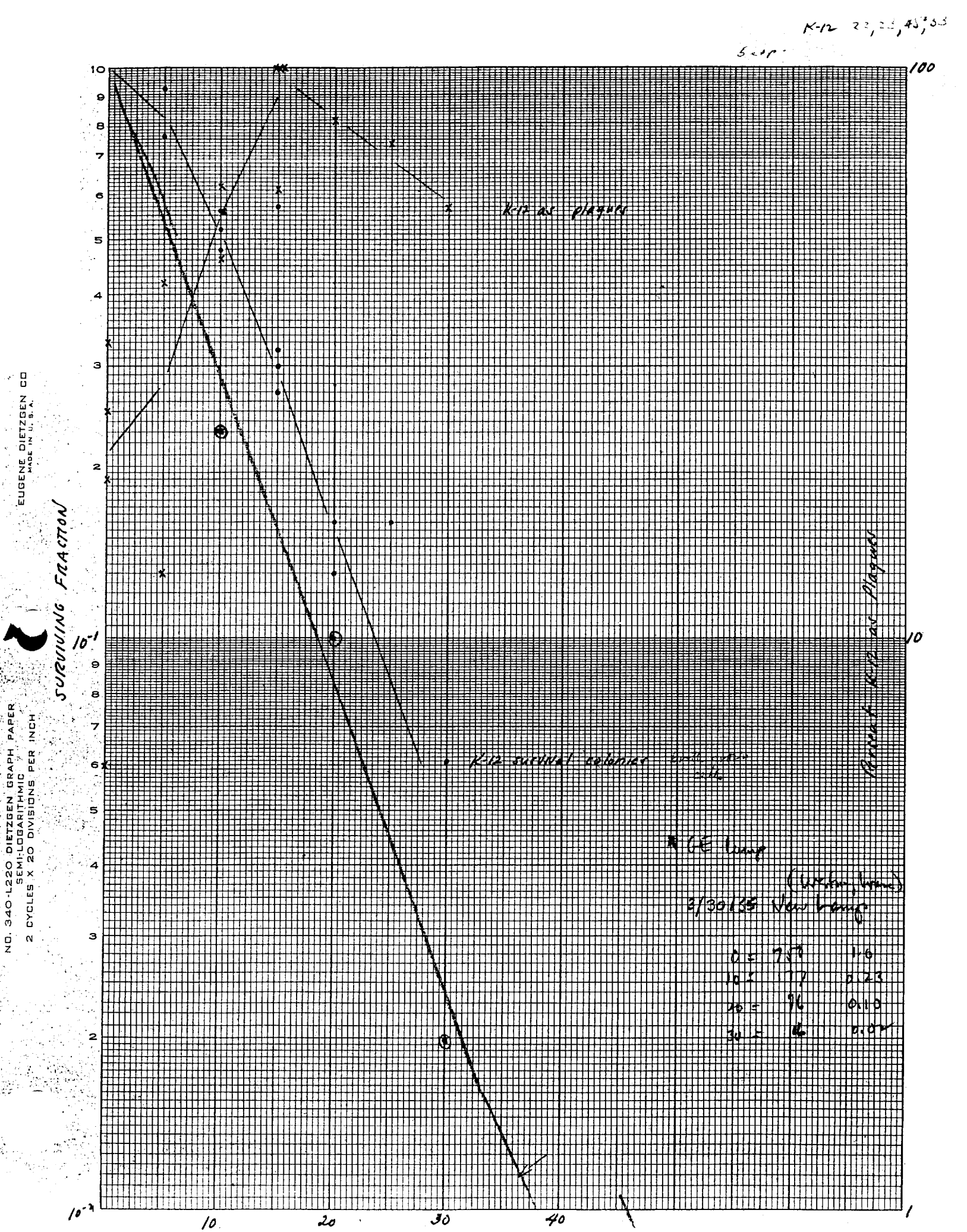
KEUFFEL & ESSER CO., N. Y. NO. 358-77
Semi-Logarithmic, 3 Cycles X 10 to the Inch, 5th lines accented.
MADE IN U.S.A.

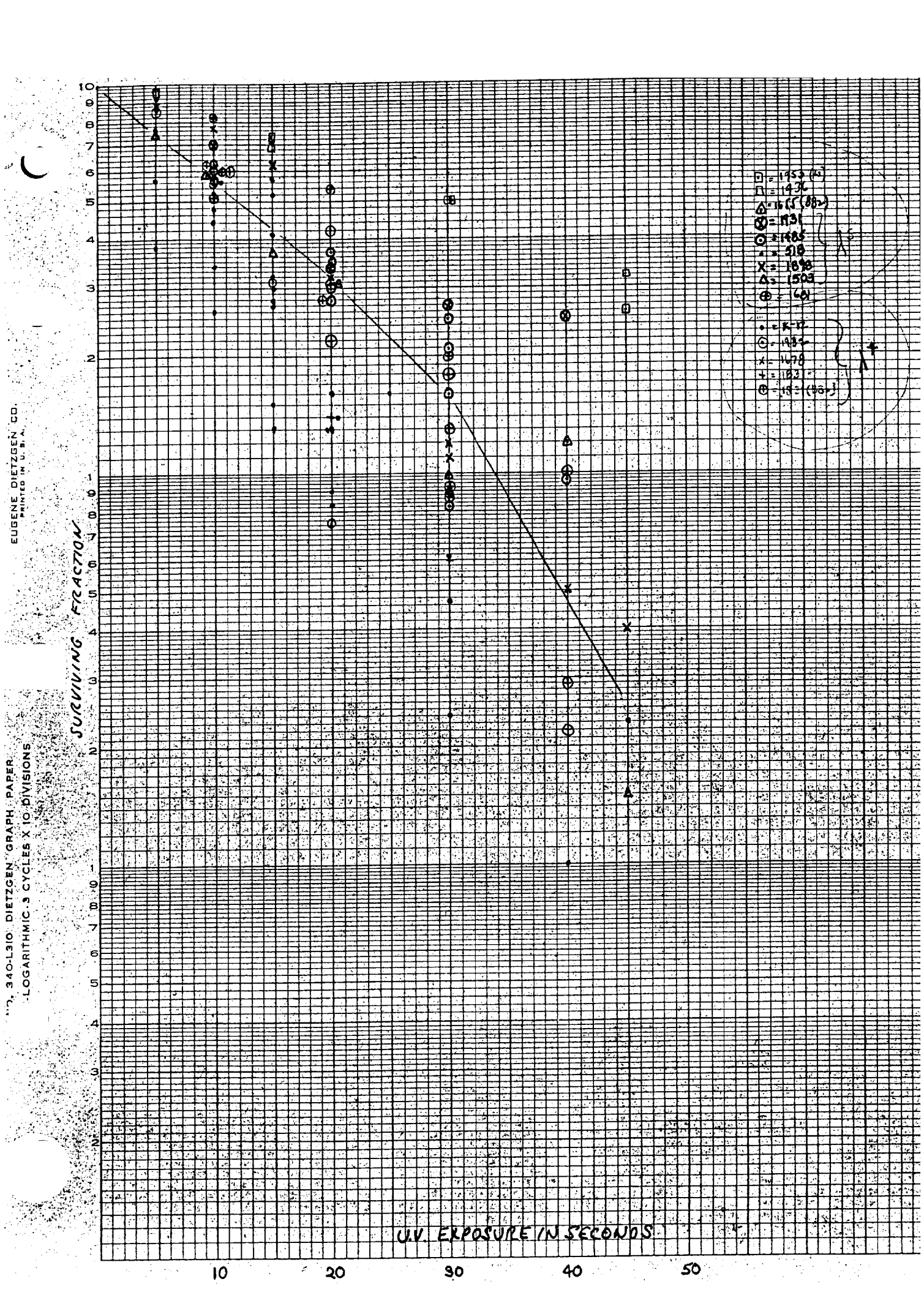
A267 - 57, 59, 60, 68



K-12 23, 25, 43, 53

Surv.





EUGENE DIETZGEN CO.
PRINTED IN U.S.A.
340-L310 DIETZGEN GRAPH PAPER
LOGARITHMIC 3 CYCLES X 10 DIVISIONS

Irradiation - U.V - Exp II

(6) Morse 12/26/51

10 ml fresh culture (2 hours from overnight unaerated) diluted 1-10 in W-D buffer after centrifugation and resuspension in 10 ml W-D buffer.

Culture	Dose (sec)	Dilutions (0.1 ml plated)	Count (on EMB-Lac)	D.F.	Irrad. titer	Surviving Fraction (titer ÷ diluted 1-10 before irradiation)
A (W-1655)	0	10^{-6}	174	10^{-7}	1.74×10^8	
B (58-161)	0	10^{-6}	174	10^{-7}	1.74×10^8	
C (W-1681	0	10^{-6}	92	10^{-7}	9.2×10^7	
D (W-1682)	0	10^{-6}	102	10^{-7}	10.2×10^8	
A	60	10^{-4}	995 (EMB-Lac)	10^{-5}	9.95×10^7	5.7×10^{-1}
B	60	10^{-3}	57 (EMB-D)	10^{-4}	5.7×10^5	3.2×10^{-3}
C	60	10^{-3}	1937	10^{-4}	1.937×10^7	0.21×10^0 ± 2.1×10^{-1}
D	60	10^{-3}	1081	10^{-4}	1.081×10^7	1.0×10^{-1}

(7) More

Exp III 12/27/51

Stock suspension from Exp II used.
Diluted 1-10 ē PO$_4^{=}$ W-B before Irrad.

Culture	Dose	Dilutions	(almost lethal) D.F.	Counts (all on EMB-Lac-)	Titer	Irrad Titer (1-10)	S.F.
A	0	2, 4, ⑥	10^7	202	2.0×10^9	2.0×10^8	
B	0	2, 4, ⑥	10^7	304	3.0×10^9	—	
C	0	2, 4, ⑥	10^7	77	7.7×10^8	—	
D	0	2, 4, ⑥	10^7	69	6.9×10^8	—	
A	100 secs.	2, ③, ⑤	10^6	3	3.3×10^6		$\frac{3.3\times10^6}{2\times10^8} = 1.6\times10^{-2}$ (0.704)
B	100	1, ③, ③	10^5	93			
C	100	② ③ ④	10^3	—			
D	100	② ③ ④	10^4	—			
			10^6	—			
			10^4	—			
			10^5	—			
			10^4	—			

Stock suspension

No colonies

Exp II (8) Mine
12/20/51

Stock suspension from Exp II used
Diluted with PO$_4$ = W-D 1-10 before inoc.

Culture	Pore		Dilutions	(0.1ml/plate) D.F.	EMB Lac Count	Titer	Tyrod. titer (-10)	S.F.
A W-1685	0	Stock susp into tryptone broth 1st dilution	2, 4, (6)	10^7	111	1.1×10^9	1.1×10^8	
B 58-164	0		2, 4, (6)	10^7	198	1.9×10^9	1.9×10^8	
C W-1681	0		2, 4, (6)	10^7	69	6.9×10^8	6.9×10^7	
D W-1687	0		2, 4, (6)	10^7	62	6.2×10^8	6.2×10^7	
A	80 sec.		2, (4)(5) →	10^5	81	8.1×10^6	—	7.3×10^{-2}
B	80		(1)(2)(3) →	10^2	142	1.4×10^4	—	7.4×10^{-6}
C	80		(2)(3)(4) →	10^5	7.3	7.3×10^6	>	1.1×10^{-1}
D	80		(2)(3), (4) →	10^5	52	5.2×10^6	—	8.4×10^{-2}

Lac
Colonies picked and streaked EMB lac
cultures purified and coded
in stocks 180.5
180.6
180.7

58-161

Exp V (4) duplicate

Dose	Phage Titer/ml $\times 10^6$	Viable bacteria/ml	Phage / viable cell	Phage / killed cell
0	66	1.8×10^8	$\frac{6.6 \times 10^7}{1.8 \times 10^8} = 3.1 \times 10^{-1}$	—
20	61	3.5×10^7	$\frac{6.1 \times 10^7}{3.5 \times 10^7} =$	—
40	109	4.6×10^6	$\frac{10.9 \times 10^7}{4.6 \times 10^6} =$	4.5×10^7
60	73	3.6×10^5	$\frac{7.3 \times 10^7}{3.6 \times 10^5} =$	
80	46	1.0×10^4	$\frac{4.6 \times 10^7}{1.0 \times 10^4} =$	
100	6	1.2×10^3	$\frac{6 \times 10^6}{1.2 \times 10^3} =$	

58-161 (10) Mme

Survival of B- Suspensions of Exp II 12/29/31 Exp I

Diluted with PO4 = W-D. 1-10 before Irrad.

Dose	Dilutions	0.1 diluted D.F.	Emulsion Counts	Irrad. rate	S.F.
0	2, 4, (6)	10^7	176	1.8×10^8	—
~~10~~					
20	2, (4), (5), (6)	10^6	83	3.5×10^7	2.0×10^{-1}
~~30~~					
40	(2), (3), (4), (5)	10^5	46	4.6×10^6	2.6×10^{-2}
~~50~~					
60	(3), (3), (4)	10^4	3.6	3.6×10^5	2.0×10^{-3}
~~70~~					
80	(1), (2), (3)	10^2	101	1.0×10^4	6.1×10^{-5}
~~90~~					
100	4, (1)	10^{11}	253		
		10^2	12	1.2×10^3	6.6×10^{-6}

(A) W-1655 λ

Survival of (B) 58-161 λ^t and Luroff Effect

(II) Illinois

Exp VI

New stock susp. prepared 1/2/52 from overnight unaerated y.e. broth culture by resusp. in W-D PO4 ≡ - Before Irrad. dil 1-10 ≡ W-D PO4 ≡

	Dose	Dilutions	Counts EMB loc 0.1ml	Titer	S.F.
	0	1, 2, (4) →	~~7~~	4X10^5 → 1.9X10^6	1
(B) 58-161	20	1, 2, (3) →	208	2.1X10^6	1.3X10^1
	40	1, (2) →	349	3.5X10^5	2.5X10^-1
	60	(1) →	435	4.4X10^4	3.1X10^-2
	80	(1) →	167	1.6X10^4	1.1X10^-2

(1) dilutions made into TSB - incubated 40 minutes then diluted and plated - Keeping 0 dose

(A) W-1655	0	(1) 2, (4) →	4	4X10^5 → 1.6X10^6	1.0
	20	1, 2, (4) →	28	2.8X10^6	
	40	1, 2, (3) →	~~164~~	1.6X10^6	1.0
	60	1, 2, (3) →	122	1.2X10^6	6.6X10^-1
	80	1, 2, (3) →	~~20~~ 141	~~2.0X10^5~~ 1.4X10^5 → 1.7X10^5	1.0X10^-1

4/2/52 (12) Mimm
Exp VIII

Radiation Sensitivity of $\lambda^{+/-}$

Fresh suspensions - Made from overnight un aerated broth cultures resuspended in W-0 PO4 - Plated 1-1H before irradiation with W-0 PO4

Pair			Count EMB-Lac (0.1ml)	Titer	S.F.
E W-1485 λ^{r} F K-12 λ^{+}	E-0	2, (4) →	122	1.2×10^{7}	
	E-80	(2) (3) →	20	2.0×10^{5}	1.1×10^{-2}
		→	77	7.7×10^{4} $>1.4\times10^{5}$	
V	F-0	2, 3 (4) →	261	2.7×10^{7}	
	F-80	un (3) →	6	6.0×10^{1}	2.2×10^{-6}
G W1673 H W-1678 λ^{+}	G-0	2, (4) →	82	8.2×10^{6}	
	G-80	2, (2) →	74	7.4×10^{4}	9.0×10^{-3}
	H-0	2, 3 (4) →			
	H-80	un, 1 →			

no colonies - cells kept out? too few?

J-W-1177 λ^{+} K-W-1603 λ^{-}	J-0	2, (4) →	not done insufficient time
	J-80	un, 1 →	
	K-0	2, (4) →	
	K-80	2, (1) →	

Exp VIII-A (13) more 1/3/52

Radiation Sensitivity and Turn Off Effect in W-1177 λ+ (J) and W-1603 λ− (resistant) (K)

Stock suspension from Exp VIII diluted 1-100 c W-0 PO4 ≡

Strain				Titer/ml	Corrected Titer	S.F.
J	J-0	in TSB 1, 3, 4 →	68	6.8×10^6	6.8×10^6	1−
W-1177 λ+		→ ca 500				
	J-60 immed. plat.	und, 1, 2 →	39	3.9×10^4	3.9×10^4	5.7×10^{-3}
	1.0 ml + 10 ml Pan + YX Broth - Incub. 40 min @ 37 in WB	und, 1, 2 →	27	2.7×10^3	2.7×10^4	3.9×10^{-3}
		→		dil 10^3	plaques in 1 ml: 142	titer/ml: 1.4×10^5
		→		10^4	16	1.6×10^5

from 6.8×10^6 titer of cell
2.7×10^4 viable cell

K	K-0	in TSB 1, 3, 4 →	73	7.3×10^6	7.3×10^6	~~8.7×10^{-2}~~
W-1603 λ−		→				
	K-60 immed. plating	1, 3 →	64	6.9×10^5	6.4×10^5	8.7×10^{-2}
	K-60 1.0 ml + 10 ml Pan + YX broth Incubation 40 m @ 37 WB	2, 3 →	79	7.9×10^4	7.9×10^5	1.1×10^{-1}
		→				

Mouse 1 (51) (51) (6)

Recapture Exp. II

10 ml culture of each (A, B, C, D, D_A) centrifuged and resusp. in W-O PO$_4$ = Diluted 1-100 = W-O PO$_4$ = before inoc. (ca 10^6 cells/ml)

Diluted 1.0 ml + 10 ml Pen + Fx - Inc. 40 minutes @ 37C

Culture	Dose	Dilution	0.1 ml on EMB	Titer	S.F.	
A (W-1655)	0	1, (3) →	620	6.2×10^6	—	
B (58-161)	0	1, (7) →	768	7.7×10^6	—	
C (W-1641)	0	1, (3) →	821	8.2×10^6	—	
D (W-1682)	0	1, (7) →	919	9.1×10^6	—	
D_A (58-161 c 82)	0	1, (7) → ↑ TSB	486	4.9×10^6	—	

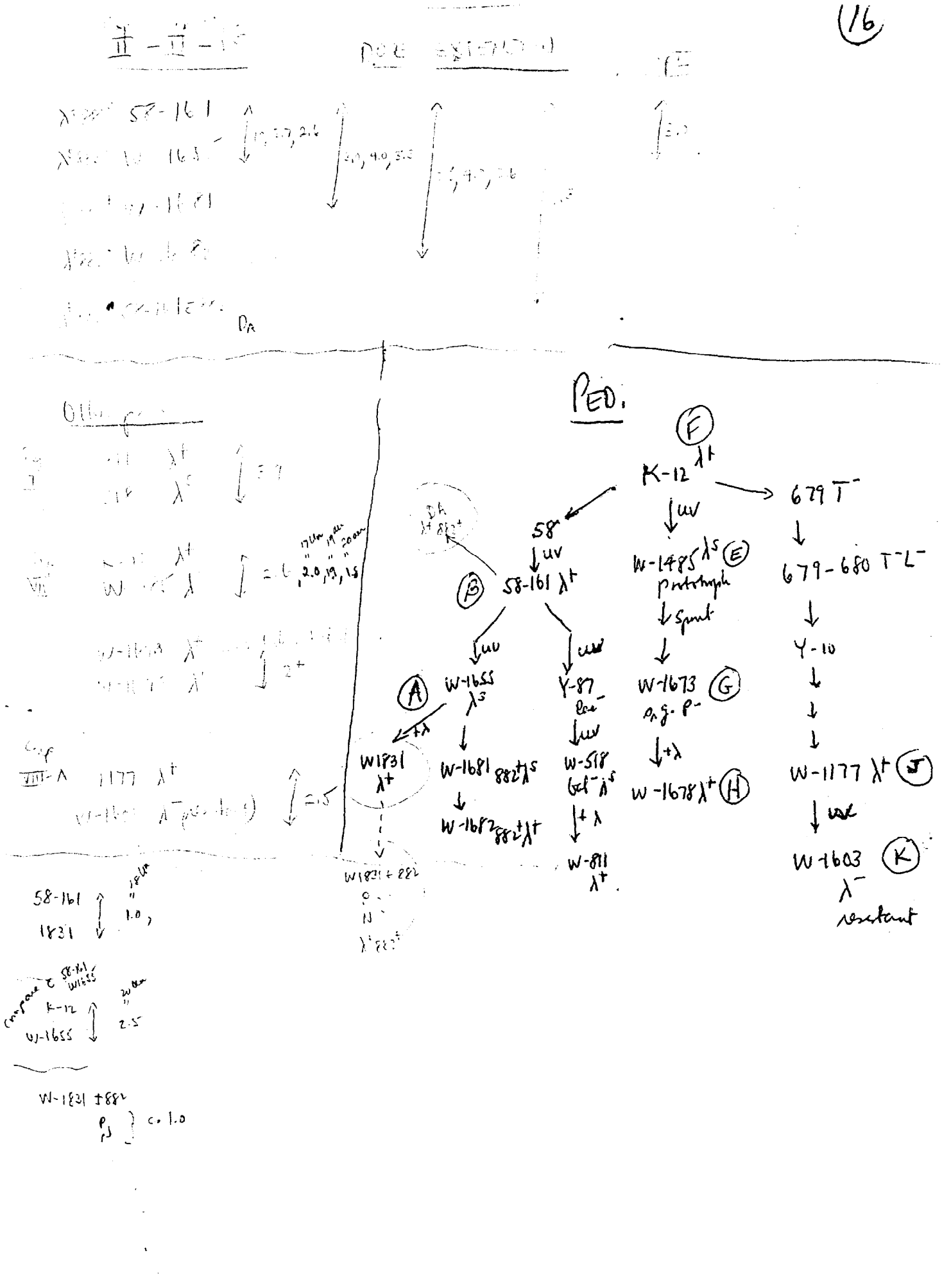
These diluted 1-10 further than above because of nitrofurazone

A	~~40~~	(1) →	147	1.5×10^6 / 1.3	$\frac{1.3 \times 10^6}{6.2 \times 10^6}$	$= 2.9 \times 10^{-1}$ v
		(2) (3) →	11	1.1×10^6		
B	~~40~~	(1) (2) →	48	1.8×10^5	$\frac{4.8 \times 10^5}{7.7 \times 10^6}$	6.2×10^{-2} v
C	~~40~~	2 (3) →	35	3.5×10^6	$\frac{3.6 \times 10^6}{8.2 \times 10^6}$	4.4×10^{-1} —
D	~~40~~	(2) (3) →	28	2.8×10^6	$\frac{2.7 \times 10^6}{9.1 \times 10^6}$	2.9×10^{-1}
		→	246	2.5×10^6 / 2.7		
D_A	40	(1) (2) (3) →	6	6×10^5	$\frac{5.5 \times 10^5}{4.9 \times 10^6}$	1.1×10^{-1}
		→	50	5×10^5 / 5.5		

↑ dilution for growth but considered in S.F.

(1) (2) (3) (4) (5) (6) (7) (8) (9) (10) (11) (12) (13) (14) (15) (16) (17) (18) (19) (20) (21) (22) (23) (24) (25) (26) (27) (28) (29) (30) (31) (32) (33) (34) (35) (36) (37) (38) (39) (40) (41) (42) (43) (44) (45) (46) (47) (48) (49) (50) (51) (52) (53) (54) (55) (56) (57) (58) (59) (60) (61) (62) (63) (64) (65) (66) (67) (68) (69) (70) (71) (72) (73) (74) (75) (76) (77) (78) (79) (80) (81) (82) (83) (84) (85) (86) (87) (88) (89) (90) (91) (92) (93) (94) (95) (96) (97) (98) (99) (100)

10^6/ml → 1, 3 →


PED.
F
K-12 λ+
679 T-
679-680 T-L-
58
uv
58-161 λ+
B
W-1485 λs
E
protrophs
spmt
W-1673
G
ag. p-
+λ
W-1678 λ+
H
Y-10
W-1177 λ+
J
W-1603
K
λ-
resistant
A
W1831 λ+
W-1681
W-1682
Y-87
W-518
λ+
W-811 λ+

(F) = (E) = 2/5/51 (17)

Comparison of K-12 1st and 1485 1s

Unaerated broth cultures - centrifuge and resuspend in W-0 buffer. Dilute 1-100 with W-0 Buffer before irradiation - Make last dilution into P-X broth-inoculate this dilution tube for 4C, 5 minute sample.

Expected Survival No. of cells

Dose	K-12 Inc.	K-12 Unirrad.	1485 Inc.	1485 Unirrad.
0	10^8	10^7	10^8	10^7
20	10^5	10^6	5×10^6	5×10^6
40	10^3	10^5	5×10^5	10^6
60	10^2	10^3	5×10^4	5×10^5
80	0	10	10^4	10^5

K-12	Dose	Charact	Dilutions	Plate	Count	
	0	unincubated	2, 4 → 0.1 ml	F-0-4	80	
✓		incubated	2, 4 "	F-0-4I	123	$\times10^5$
	20	unincubated	2, 3 "	F-20-3	154	
✓		incubated	3 "	F-20-3I	206	$\times10^4$
	40	unincubated	2 "	F-40-2	141	
✓		incubated	2 "	F-40-2I	367	$\times10^3$
	60	unincubated	1 → undil	F-60-1	781	$\times10^1$
		incubated	1 →	F-60-1I	58	$\times10^2$
	80	unincubated	und. →	F-80-1	41	$\times10^1$
		incubated	Δ →	F-80-1I	0	$\times10^2$

Surviving Fractions

K-12 Dose	Incubated S.F.	Unincubated S.F.	
0	1.0	1.0	✓
20	1.7×10^{-1}	8.9×10^{-1}	
40	3.1×10^{-2}	1.8×10^{-2}	
60	4.7×10^{-4}	9.6×10^{-4}	
80	0	5.1×10^{-5}	

W-1485

	0	unincubated	2, 4 →	E-0-4	160 $\times10^5$
		incubated	4 →	E-0-4I	229
	20	unincubated	2, 4 →	E-20-4	75 $\times10^5$
		incubated	4 →	E-20-4I	110
	40	unincubated	2, 3 →	E-40-3	290 $\times10^4$
✓		incubated	3 →	E-40-3I	353
	60	unincubated	2, 3 →	E-60-3	*193/2 = 97 $\times10^4$
		incubated	3 →	E-60-3I	140
✓	80	unincubated	2, 2 →	E-80-2	73 $\times10^3$
		incubated	2 →	E-80-2I	147

W-1485

Dose	Incubated S.F.	Unincubated S.F.
0	1.0	1
20	4.8×10^{-1}	4.7×10^{-1}
40	1.5×10^{-1}	1.8×10^{-1}
60	6.1×10^{-3}	6.1×10^{-2}
80	6.9×10^{-3}	4.6×10^{-3}

Many lysed cells + sectors of K-12, esper 40: 1 col = $1/4 \cdot 1872$

* 0.2 ml Plated

2/7/52

Comparison of U.V. resistance of 58-161 = A ○
W-1655 = B ○
W 1831 = C ×

24 hour unaerated cultures used - centrifuged and resuspended in W-10 buffer - diluted 1-10 with W-0 buffer before irradiation

Expected			Experimental			EHB Plate	Counts	Titr	S.F.
Cult.	Dose	Survival	Cult	Dose	Dilutions*				
A	0	$1.0 = 10^7$ cells/ml	A	0	2, 4	A-0-4	201	2.0×10^7	1.0
	20	$0.1 - 10^6$ cells/ml		20	2, 3	A-20-3	579	5.8×10^6	2.9×10^{-1}
	40	$0.01 - 10^5$ cells/ml		40	2	A-40-2	56	5.6×10^4	2.7×10^{-3}
B	0	$1.0 - 10^7$ cells/ml	B	0	2, 4	B-0-4	142	1.4×10^7	1.0
	20	$0.5 - 5\times10^6$ cells/ml		20	2, 4	B-20-4	35	3.5×10^6	2.5×10^{-1}
	40	$0.1 - 0.5 = 10^6$ cells/ml		40	2, 3	B-40-3	237	2.4×10^6	1.7×10^{-1}
C	0	$1.0 = 10^7$ cells/ml	C	0	2, 4	C-0-4	102	1.0×10^7	1.0
	20	$0.1 - 10^6$ cells/ml		20	2, 3, 4	C-20-3	182	1.8×10^6	1.8×10^{-1}
						C-20-4	27		
	40	$0.1 - 10^6$ cells/ml		40	2, 3	C-40-3	22		
		$0.01 - 10^5$ cells/ml				C-40-2	255	2.5×10^5	2.5×10^{-2}

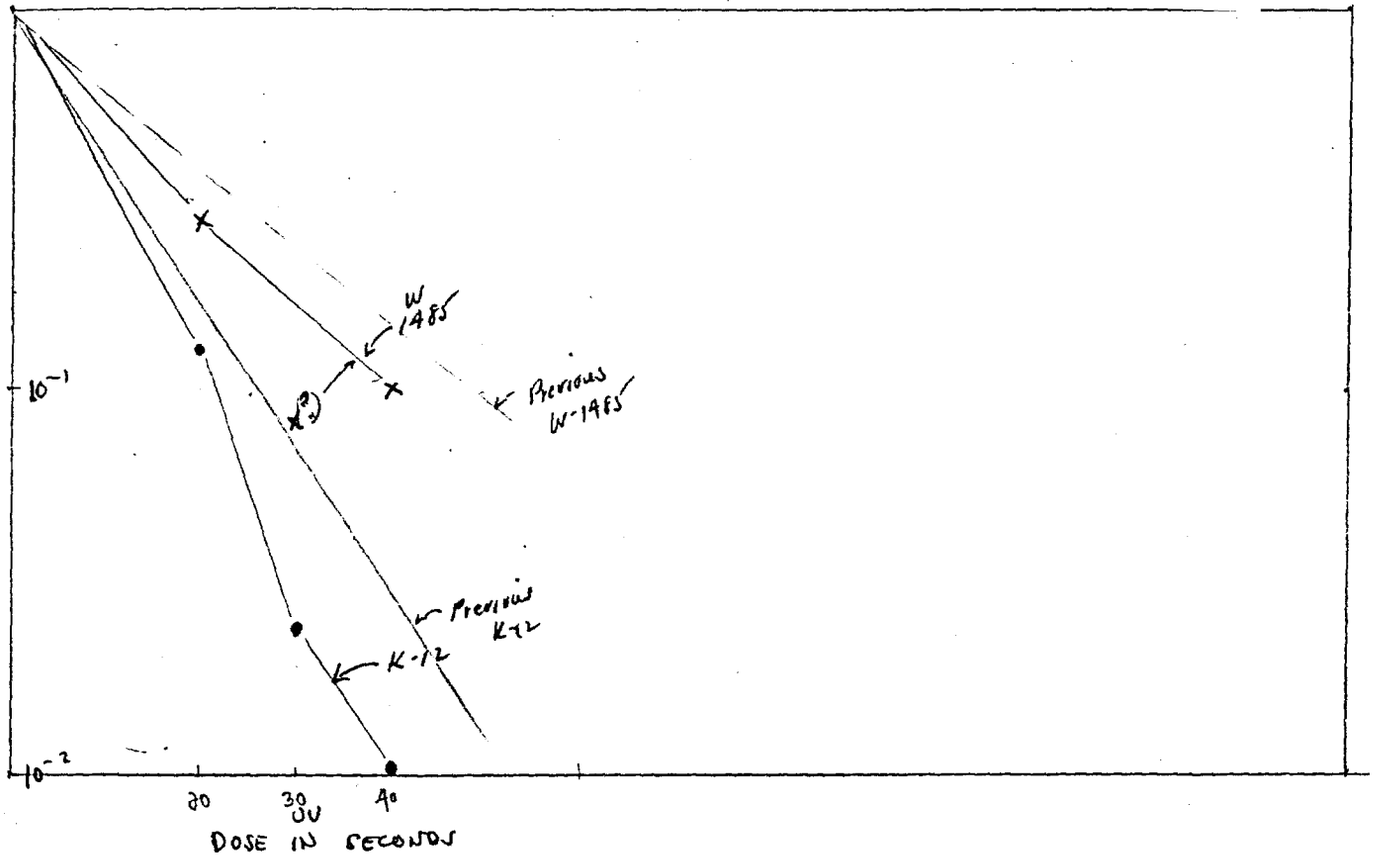
*water

Comparison of K-12 and W1485 —

Aerated cultures = 0.1 ml 24 hour unaerated culture + 10 ml PX broth incubated in air 37C 4½ hours — Centrifuged and resuspended in W-0-buffer-diluted 1-100 before irradiation

K_{12} Dose	Expected Titer	Dilution	EMB Plates	Counts	Titer	S.F.	
0	1×10^7	2, (4) →	A-0-4	– 229	2.3×10^7	1.0	9/229
20	1×10^6	2 (3) →	A-20-3	– 30	3.0×10^6	1.3×10^{-1}	92/229
30	5×10^5	2, (3) →	A-30-2	– ~	~		
			A-30-3	– 56	5.6×10^5	2.4×10^{-2}	data obtained from counts prepared in parallel for the two points
40	1×10^5	2 (3) →	A-40-2	– 238	2.4×10^5	1.0×10^{-2}	83/229
		→	A-40-3	– 21	~		71/229

W1485						
0	1×10^7	2 (4) →	B-0-4	– 277	2.8×10^7	1.0
20	5×10^6	2, (4) →	B-20-3	– ~		
		→	B-20-4	– 80	8.0×10^6	2.8×10^{-1}
30	2×10^6	2 (3) →	B-30-3	– 247	2.5×10^6	8.2×10^{-2}
40	1×10^6	2, (3) →	B-40-3	– 267	2.7×10^6	9.6×10^{-2}



K W 2/10/52.

Comparison of K-12 and 1485

Cultures from overnight unaerated PX broth by 0.1 + 10.0 and aerated 5 hours - Centrifuge and resuspended in W-D buffer - Dilute 10^6 = ca 10^3 cells/ml - Sampled immediately for 0

2, 4, C

K-12	Dose	Plate	Count/plate	Count/ml	Phages	S.F
	0	K0 -	446	4.5×10^3	0	1.0
	10	K-10 -	195	2.0×10^3		4.4×10^{-1}
	20	K-20 -	62	6.2×10^2	No	1.4×10^{-1}
	30	K 30 -	21	2.1×10^2	phage	4.7×10^{-2}

Only with phage

W 1485	Dose	Plate	Count/plate	Count/ml	Indicator	
	0	W-0	626	6.3×10^3	Contaminated	1.0
	10	W-10	392	3.9×10^3	and	6.2×10^{-1}
	20	W-20	234	2.3×10^3	Carrying	3.7×10^{-1}
	30	W-30	129	1.3×10^3	phage	2.1×10^{-1}

Little higher than usual

Examination of W-1655 = C

Unaerated culture - 30 hours centrifuge resuspended in W-D - dilute 10^6 - ca 10^3 cells/ml

Dose	Plate	Counts/plate	Count/ml	
0	C-0	175	1.8×10^3	1.0
5	C-5	~~mutt~~		
10	C-10	138	1.4×10^3	7.8×10^{-1}
20	C-20	92	9.2×10^2	5.1×10^{-1}
30	C-30	64	6.4×10^2	3.6×10^{-1}
40	C-40	56	5.6×10^2	3.1×10^{-1}

Use agreement with W-1661 in W-1661 & 9,5, etc

Examination of W-1655

Undiluted culture - 28 hours PX - Centrifuge, resusp in W-0. buffer - dilute 10^6 - Inoculate

Dose	Plate	Counts
0	W-0 -	?
~~35~~	W-5 -	77
10	W-10	70
15	W-15	60
20	W-20	43
30	W-30	58
40	W-40	-
50	W-50	-

dead exp
fresh plates repeat

1831 + 882 -

Four ~~plaques~~ picked (central growth) and streaked

3 grew -

1482
1837 } to date - single colonies on original EMB last picked - two sets 40 in 1st
16 in 2nd

one possible diploid in 1st 40, more in 2nd

Possible streaked on EMS and EMB - indicated dip.

Three members of EMS dip picked to EMB, inoculated in DM+ lactose (1:0 ml/10ml, 10%)

aerated 10:15 2/19/52

just probable

Comparison of K-12 (K) with 1485 (W)

2/13/52

4 hour aerated culture from overnight incubated - dilute to contain 10^3 cells/ml (2 W-0) - Incubate - Plating for phage on S18

K-12 as Plaque	K-12	Dose	Count/plate	Diff.	Count/ml	Phage/ml	S.F.
0.25		0	278		2.8×10^3	70	50
0.50		10	156	122	1.6×10^3	$\frac{990}{2} = 1560$	5.6×10^{-1}
0.8		20	37	241	3.7×10^2	2240	1.3×10^{-1}
0.57		30	17	261	1.7×10^2	1570	6.1×10^{-2}

1485	0	187	1.9×10^3	S.F. 1.0
	10	106	1.1×10^3	5.7×10^{-1}
	20	66	6.6×10^2	3.5×10^{-1}
	30	30	3.0×10^2	1.6×10^{-1}

Ratio: cells lost / phage gained — Dose

$\frac{122}{156} = 0.79$ — 0-10

$\frac{241}{224} = 1.1$ — 0-20

$\frac{261}{157} = 1.7$ — 0-30

Culture started PX broth

WB-1] project X → 0.25 + 10 ml PX (1-16) 9:15

W1-14, W1-16] for Penicillin Run → 0.25 + 10 ml PX 9:15

K-12, 1485] possible retake on above exp.

2/14/52 (23)

Comparison of K-12 and W-1485

Heavy Effect — 4 hr aerated cultures in PX from overnight unaerated cultures

K-12

Dose	Plate	Colonies/plate	diff.
0	K-0	250	
			18
5	K-5	232	
			120
10	K-10	130	
			182
15	K-15	68	
			210
20	K-20	40	
			210
25	K-25	41	

/ml		Plaque/plate		diff		S.F.	K-12 (no plaques)
2.5×10^3	0	-	<50		0	= 1.0	
					5	= 9.2×10^{-1}	<25
2.3×10^3	5	105	319/3 =	105			
				<55	10	= 5.2×10^{-1}	.92
1.3×10^3	10	155	155	105			
				205	15	= 2.7×10^{-1}	.62
6.8×10^2	15	230	360				1.0?
4.0×10^2	20			134	20	= 1.6×10^{-1}	0.74
4.1×10^2	25	134	384		25	= 1.6×10^{-1}	?

Ratio: cell loss diff. / plaque gain diff.

Dose 0-5: $\frac{18}{<55} = >0.32$

Dose 0-10: $\frac{120}{105} = 1.1$

Dose 0-15: $\frac{182}{205} = 0.89$

Dose 0-25: $\frac{210}{134} = 1.6$

W-1485

Dose	Plate	Colonies/plate	Count/ml	S.F.
0	W-0	251	2.5×10^3	1.0
10	W-10	153	1.5×10^3	6.0×10^{-1}
20	W-20	75	7.5×10^2	3.0×10^{-1}
30	W-30	62	6.2×10^2	2.5×10^{-1}

Cultures

- K-12
- W-1485
- Wq-19
- Wq-16
- WB-1

} are last-tipped ones in incubator

Cultures made - 2/15/52

PX { K-12 - repeat ē incubation
1831 - examine 1831 + FP2 for lysogenic ē FP2
WB-1 - pyocin X.

Sat. 2/16/52 IC-12 aerated culture started 7:50 out at 9:50 0.5+10 $\frac{1}{20}$
WB-1 " " - 7:55 - no lysis 11:10
refrigerat until 2/18

Effect of Post Incubation

K-12 - Centrifuge - resusp in W-D - Dilute to 10^4 viable cells/ml - Post aerad. incub. 40 min at 37C

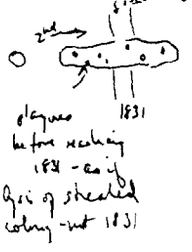
Area	Plate	Count/plate
0	K-0 -	58
	K-0 - Inc	140
15 sec	K-15	25
	K-15 - Inc	20

2/17/52
Lysed WB-1 tube plated at - TSA
10^2 -
10^4 -
10^6 -

Culture started 2/17/52
IC-12
1831
WB-1

WB-1 culture continued at 37C
in 10:00 AM
out 11:05 AM lysis apparently beginning.

20 colonies of 1831 + 882 (from plaques) streaked across 1831 for lysogenicity ē 882 made 2/16/52 examined - no clear cut examples of sensitivity of 1831 to any of streaks noted One streak contained small plaques



Cultures 2/18/52
Started

K-12
1831
1485

2/19/52

Aerated cultures started K-12, 1485 - at 9:15 - aerated overnight
not 12:15 - 0.25 + 10 cc PX

1831 + 882

Z = 1831
W = 1485
P = plague forming - see bottom of L4
N = non-plague forming ⊙

Results 2/20/52

N gave plagues on ref.
lysis 1485 = $N\lambda^+$
lysis 1831 = N^{882+}

P gave no plagues on ref.
lysis 1485 = $p\lambda^+$
lysis 1831 = p^{882+}

K-12 - Effect of post-incubation on Survival

Inoculate - aerated 4:30 hours in PX - Centrifuge - resuspend in MD* dilute ~ M-O to contain 10^9 cells/ml - After incub. 1/10 dilution with PX broth - Incubated and air 40 min at 37C

Dose	Plate	Count/Plate		S.F.
0	K-0	281	-276	1.0
	K-0-I	270		
10	K-10	84		8.2×10^{-1}
	K-10-I	185		6.7×10^{-1}
15	K-15	42		1.5×10^{-1}
	K-15-I	117		4.2×10^{-1}
20	K-20	23		8.3×10^{-2}
	K-20-I	58		2.1×10^{-1}

I = post-Incubated counts.

Suggest that post incubation in aeration deactivates lysogenic effect.

* remainder of this response aerated 25 sec, ca 8 ml PX added and incubated ~ aeration
2:00 PM - 4:00 PM - slight lysis apparent.
left on bench overnight - nearly clear next morning - centrifuged and enough plague (?) for 4 ml
titer: $10^6 \times 43 = ca\ 8.6 \times 10^8 \times 10 = 8.6 \times 10^9$

Cultures started 2/18/52
K-12
1485
1831