N18

DATE: 9/7/56 REF: lac, gal+ / gal− interaction

See EML thesis. W1402 = W811S^r = lac_1^{87} gal_4

W1402 broth from EML.

Cross-streaks on EMBlac against dlac− F−.

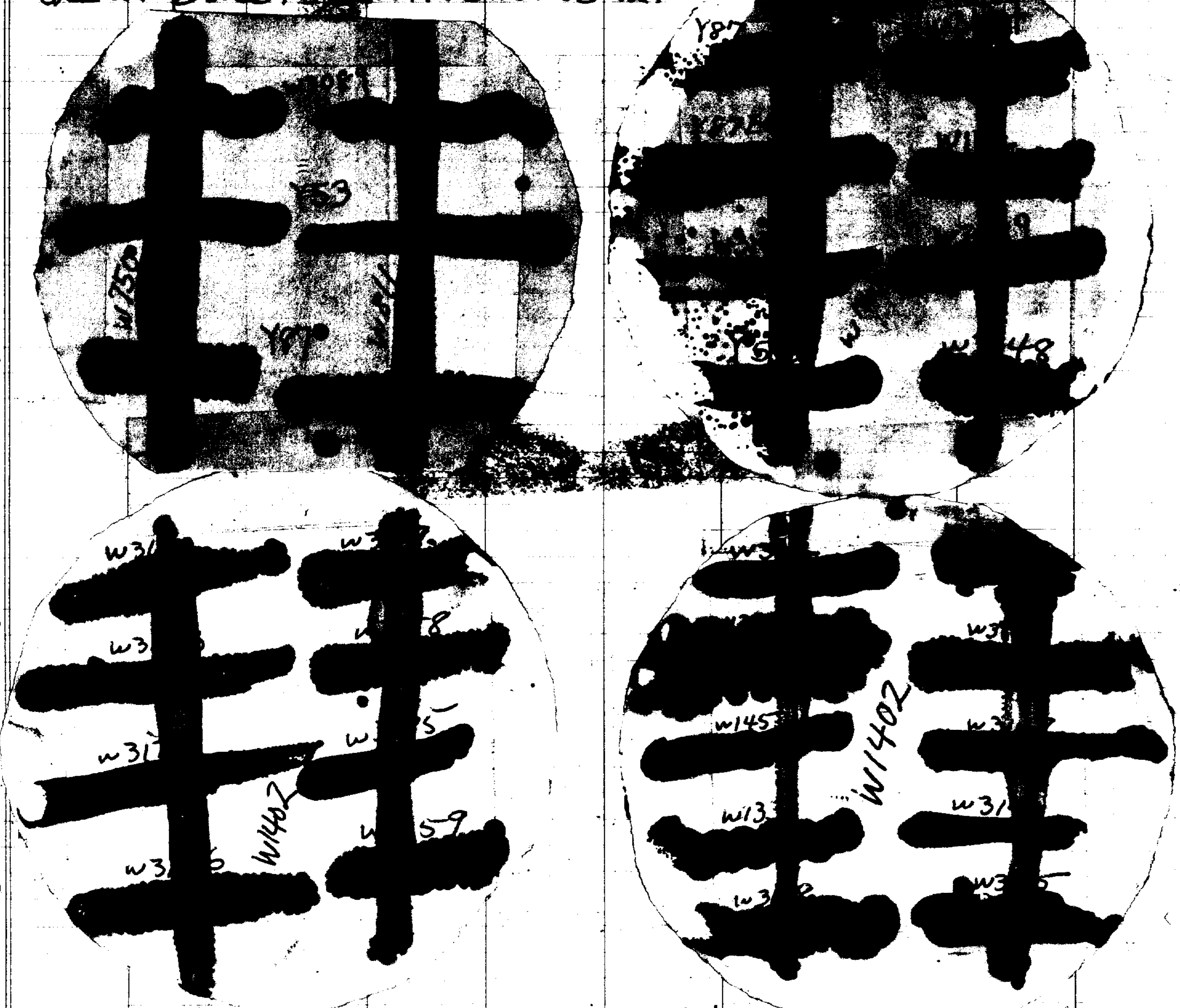
A 9/8 no interaction. Re-incubate.

9/10. Result not clear.

9/11 No reaction. Try W750, W811.

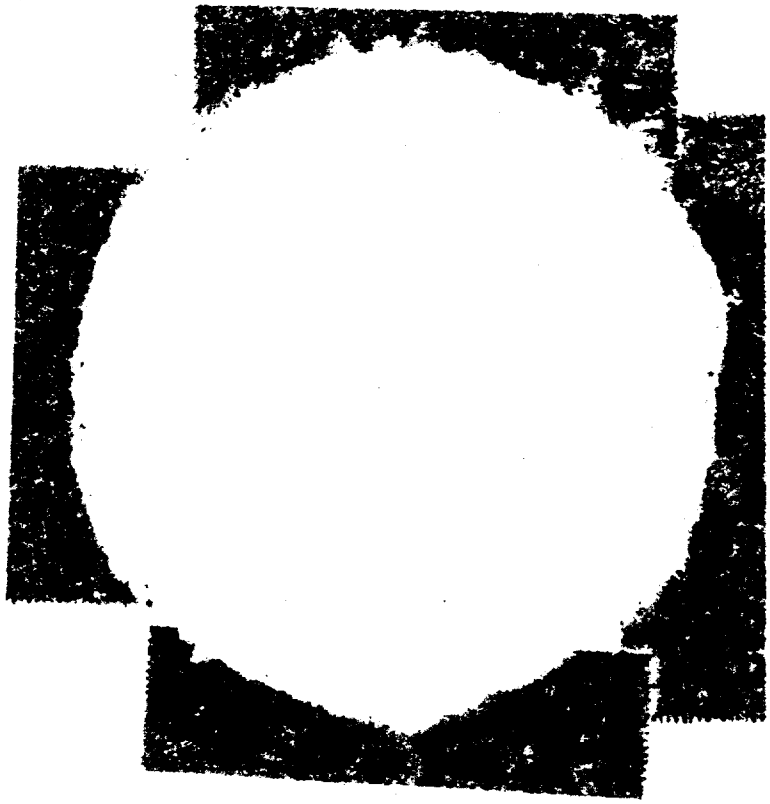
9/12 No reaction. Reincubate.

9/12 Prints made from [illegible] plate, 48 hrs. EMB reaction [illegible] clear on prints than on plates. [illegible] M lac no reaction.



F+ streaks on gal− lac+ see over.

10/7/56. Lac$^-$ Gal$^+$ on gal$^-$ lac$^+$ (w3091).



N19

UV induced lac⁻ from W1895 P⁻

DATE: 9/10/56 REF:

1	2	3	4	5	6	7	8	9	10

Single colonies 1 and 3 each spread on 8 plates B lac @ 1 drop/plate.
Exposed to Hanovia for 8 sec.

9/11 plac⁻ (?) colonies picked, streaked on B lac.

9/12. Restreaked lac⁻.

9/13. One lac⁻ from each colony (1 and 3). Restreaked on B lac.

9/14. Both colonies restreaked on B gal for stab.

9/15. N19-3 is gal⁺. (= W3240) N19-1 is gal⁺. (= W3239) Stabs made for single col.

N20

9/10/56

DATE: Prep. P1 lysate REF:

1	2	3	4	5	6	7	8	9	10

9/10 11 AM. .1 ml. of λ⁻ stocks from perassay to 10 ml. L broth on rotator. (see Lennox paper). Following λ⁻ stocks tested:

* W2859	W3059	W3014
* W3077	W1485	W3019
W2964	W1655	W2915
W3047	* W518	
W3013	W3110	
W3189	* W3010	
W3136	W3017	

2 pm. add .1 ml P1 kc / tube. Observe every hr. to 8 pm.
9 PM. in frig. 9/11. lysates centrifuged, decanted into vials, + $CHCl_3$.
The ones marked * cleared completely, with debris at bottom of tube.

9/19. phage assay. Plate 0.1 from 3rd dilution tube
$\frac{I}{100} \times \frac{II}{100} \times \frac{III}{100} \times 10 = 10^7$

indicator cultures several days old.
- all dil. in H_2O, beginning with W1655 + T6.
- last dil. in L ø of W1655 + T6.
- W 1895 used as T1, T6 indicator
- For P1, the indicator was the stock from which lysate was prepared.

W3014 assays 1×10^8 P1/ml all others less.
W1655 " 4×10^9 T6/ml

T1 lysate not good.

The "T6" lysate lyses W1366 (K_6^r). Other T6 preps. do not. Check the T6 preps. to locate origin of difference.
P_1 cross-streaks show ~~Lo~~ turbid lysis on all stocks.

mapping V_6^r lac_1^{W112} $Prol^-$ V_1^r

DATE: 9/10/56 REF:

~~W112~~ W3236 ~~X~~

$W1366 = F^- T^- L^- B_1^-$ lac_1^{W112} V_1^r V_6^r. From stab coll. into penassay

10 ml L∅ (Lennox broth) + 0.1 ml W3010. On aerator

~~Wash in penassay, centrifuged & resuspended in D(0) + prol. Plate on B + lac.~~ D(0) old Prol and purify ca. 200 colonies on D(0) + Prol.

D = 4 × 10^8 → 50 col/plate.

Prepare T6 and T1 phage stocks on W3010, W1655.

4 PM 1 loop W3010, W1655 in L∅, 5 tubes. 5 pm. 1 loop T_1 or T6

9/11. Lysates centrifuged, decanted into vials, + several drops $CHCl_3$

9/12. Mating on ~~L∅~~ D(0) + prol.

9/13. First streak on ~~L∅~~ D(0) + prol.

9/14 second streak on B gal.

9/15 third streak on D(0) + prol. 9/16 fourth streak on D(0) + prol.

9/17. Spot on D(0) + proline for replica tests.

	1st replica	2nd replica
D(0)	D(0)	B lac
B(lac)	L∅ + T6	L∅ + T1
L∅ + T1		
L∅ + T6		

Scrub block with roccal after each use.

N.B. W1366 was not tested as single colony before use. It is $T1^S$. The T_1, T6 stocks used to score V_1, V_6 are o.K. (they are T6 from EML and T1 (W1485) from stock.

N21A

Counts

DATE: 9/21 REF:

lac+,1, Prol+,? clear. V_1^r & V_6^r ambiguous. Streak out.

plate #.	P+ lac−	P+ lac+	$\bar{P}$ lac+	$\bar{P}$ lac−	Total
2	16	3	5	1	25
3	21	2	2	2	27
4	18	6	4	1	29
5	3	2	2	1	8
6	21	4	—	4	29
1	16	1	4	2	23
Total	95	18	17	11	141

~~[illegible]~~.

lac+ P−

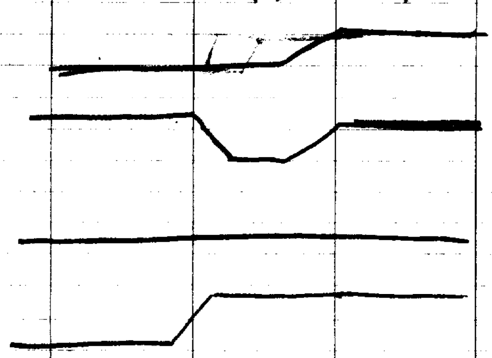
− +. TL−

lac− P+

lac+ P+

lac+ P−

lac− P−



95

18

17

11

$\frac{18}{113}$ .159

$\frac{11}{28} = .393$

~~[illegible]~~.

$\frac{29}{141} = .206$

18	95	113
11	17	28
29	112	141

$\chi^2 = \frac{7.70 \times 10^7}{1.028 \times 10^7} = 7.49$

V_6 Soc Pool

gal V_6 Soc Pool

gal-Soc 25.5

gal-V_6 22.7

gal-Pool 19.1

5.7
2.8
7.8
9.2
25.5

7.8
9.2
4.3
1.4
22.7

18+20=36

P V_6 Soc

Pool V_6 Soc

28+ 20

31 V_6 + Soc

27P

2.8
5.7
9.2
1.4
19.1

V_6

N21 B

DATE:

$V_6^{\sim}$ REF: V_6^{S}

Plate #	2 Loc +3 P+	2 Loc +3 P−	4 Loc −5 P+	4 Loc −5 P−	6 Loc +7 P+	6 Loc +7 P−	8 Loc −9 P+	8 Loc −9 P−	10
1									0
2									0
3									0
4									0
5			1						1
6	0	0	0	0	0	0	0	0	0
1	0	2	15	2	1	2	1	0	23 ✓
2	2 11	4 1	16 16	1 1	1 1	4 ////	0	0	25 ✓
3	1 1	1 1	20 20	2 //	1 1	1 1	1 1	0	27 ✓
4	//// 4	0	//// //// ////	1	2 //	//// 4	1	0	29 ✓
5	1 1	0	/// 3	0	1 1	2 //	0	0	✓ 7
6 (averaged twice on two plates 2 days apart)	0	0	18 //// //// ///	2 //	4	0	3 ///	2 1	29 ✓
	c.o.I	c.o.I	n.c.o.II	n.c.o.I	n.c.o.I	n.c.o.II	c.o.I	c.o.I	
	c.o.II	n.c.o.II	n.c.o.II	c.o.II	c.o.II	n.c.o.II	n.c.o.II	c.o.II	✓
	✓ 8	✓ 4	✓ 89	✓ 9	✓ 10	✓ 13	✓ 6	✓ 2	141
%	5.7	2.8	63.1	5.7	7.1	9.2	4.3	1.4	✓

21 C

DATE: REF:

I II

V_6^r — loc⁻ — p^-

.2?? .2?

	loc⁺	loc⁻
V_6^r	0	20
V_6^s	4	5

c.o. I 5/29 = .17

	loc⁺	loc⁻
p⁺	4	21
p⁻	0	4

c.o. II 8/29 = .276

29/141 = .206

	p⁺	p⁻
V_6^r	18	2
V_6^s	7	2

~~c.o. I + II~~ 9/29 = .310

n.c.o. I & II $\frac{18+0}{29}$ =

doubles I & II $\frac{2+0}{29}$ = .07

p–val V_6^r loc

.21 .142

~~coincidence = 1~~

all data

c.o. I 20/141 = .142

c.o. II 29/141 = .206

Doubles 10/141 = .071

Expected = .029

V_6^r — loc⁻ — p^- — V_1^r — [TL]

.142 .206 ? not small very small?

1366 V_6^r loc, w12 p⁺ ~~[illegible]~~ TL⁻

w1895 p⁻ s +

Prep. of F⁻ prototroph testers

DATE: 9/12/56 REF:

1	2	3	4	5	6	7	8	9	10

✓ 1. W3120 JL X Y10 $\xrightarrow{S\ lac}$ Second streak on S lac
stabbed as W3230.

~~2. Test N7-2 against X10 (W1895 control) [illegible].~~
~~N7-2 in [illegible]assay.~~

✓ 3. Y87 X ~~[illegible]~~ Y10 → 9/17. ~~First~~ second streak on B lac.
Stabbed as 23-3.

✓ 4. W145 X W1895 9/17 mating repeated in broth. ~~my W145 stock~~
my W145 is M⁻. Check stock ~~is M⁻~~ (grows in
~~Stabbed as W3236~~
cold.

5. W3140 ~~W2244~~ X W1895 9/17 mating in broth. Use
= ~~[illegible]7~~ W22 44F⁺ instead.

✓ 6. W133 X W1895 Second streak on S lac. = W3238.
~~Stabbed as W3236.~~
= ~~[illegible]~~3. ~~[illegible]~~.

Resistant Tests of T1, T6, P1

DATE: 9/20. REF:

On Blac: overnight plate.

	T1 strain	T6 (w1655)	P1 (w3014)	
w30140	S+	+	—	
w3110	S+	+	—	
w1655	S+	+	—	
w1366	R	(+)	—	Papilla from w1366 picked; necessary.
w1485	S	+	—	
	O.K.	V?	—	
w3146		+	—	remove's P1KC on the P1 cross-streaks.

New streaks made on LØ with stock T6 & P1kc.

7/23. Colony of w1485 resistant to T6? (w1655) tested against stock T6 and T6 (w1655). It is lysed by T6 (EML stock). not T6 h. Plaque

	T6	T6? (w1655)
w1485	lysis	lysis
w1485R	lysis	no lysis

New T6 lysates made from T6 (EML) on B/1, w1485. Both clear, controls very turbid. Centrifuged, chloroformed, shaken, overnight in frig.

Tests of T1, T6 show some sort of massive confusion in making stocks. T1 (B/6), T6 (B/1), and T6 (1485) are O.K. by comparison with T6 from EML and T1 (1485) from stock on B/6, B/1, w1485. w1366 is T1 S.

w1366 ~~is~~ made resistant to "T6" (w1655) appears to be T5 resistant. Unrecorded error lysate

N24A

Sterility check

DATE: 9/26/56

REF:

1	2	3	4	5	6	7	8	9	10

Phage stocks on B loc.

P1 lac 0

P1 (W3014) 0

T1 (1485) 0

T1 (B/6) 0

T6 (1485) 0

T6 (B/1) 0

T6 (1485) EML 0

P1 (1485) 0

N25

Agar layer plate

DATE: 9/26/56 REF:

	1	2	3	4	5	6	7	8	9	10

10 A.M. 45 ml. L ø agar / plate

2 ml. L. ø top layer agar + 2 drops cells + 1 drop P1.

1 W1485 + P1 lysate

2 W1485 + (W3014) P1

3 W1366 + P1 lysate

4 W1366 + (W3014) P1

old plates 25 ml. { 5 W1485 + P1 lysate

6 W1485 + (W3014) P1 1 ml. P1 lysate (W3014).

3 pm. (W3014) P1 no perceptible lysis. Discard

P1 lysate ~~no growth complete lysis~~ on plate 3, partial lysis on plates 1 & 5. Add 4 ml L broth to plate 3. Discard.

5 pm. add 4 ml L broth to plates 1 & 5.

8 pm. decant, chloroform, store in frig. add one L broth to plates 1 and 5.

9/3/56 Test of titer.

	1485	1485 (P1)
P1 (Lennox)	++	+
P1 (first decant fr. 1485)	±	○
P1 (decanted after 24 hrs.)	±	○

N26

T6 resistance of luc-testers on B(0)

DATE: REF: T6 (B/1)

	1	2	3	4	5	6	7	8	9	10
			+ = lysis		0 = no lysis					
plate 1					plate 2					
	W3229	+	W3133	+	—		W3159	+		
	W3120	+	W3230	+	—		W3089	+		
	343	+	W3134	+	W1941	+	W3148	+		
10	W1950	+	W3157	+	W1945	+	W3152	+		
	W1951	+	W3158	+	W1948	+	W3174	+		
plate 3	W1949	+	W3156	+	plate 4 W3164	+	W3112	+		
	W3146	Resistant	W3175	Resistant	—		W2243	+		
	W1946	+	W3153	+	W3140	+	W2244	+		
20	W3237	+	—		—		W3127	+		
	W3240	+	—		—		W2245	+		
plate 5	—		W3238							
	W3128		W3239							
30	W1940		W3147							
	W1942		W3149							
	W1943		W3215							
	W1944		W3151							
	W1947		W3154							
40										
50										

check on lac⁻ testers N27

DATE: REF:

variegated colonies

plate 1

		A			B	
	W3133	W3230	W3134	W3157	W3158	W3159
W3229	0	7	8	+	+	0
W3120	0	7	2	+	+	0
3143	0	6	5	+	+	0
W1950	12	13	3	+	+	(3)
W1951	2	12	3	+	+	0

(some diversity) — all allelic ✓

plate 2 ✓ 3143 test

		F only	+ Hfr	F-	+ Hfr	
W3089	~~W1441~~	0	0	0	0	~~W3146~~ W3156
W3148	~~W1945~~	0	0	0	5	~~W1946~~ W3175
W3152	~~W1948~~	0	0	0	3	W3153
W3174	~~W1949~~	0	0	0	0	W3133

plate 3 W3174 tester

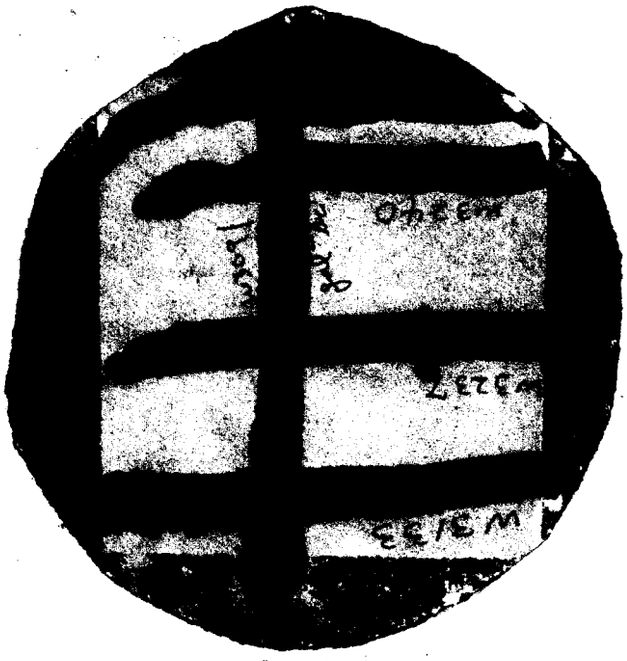
	homologous F	W3174	homologous F	W3174	
W3229	0	0	0	+ (small)	W1941
W3120	6	+		+	W1945
3143	4	0	0	0	W1948
W1950	+	+ (small)	0	0	W1949
W1951	+	(+ small)	0	+	W3146
~~W1941~~			0	+	W1946

plate 4 W3230 test

	homol. F	W3230	W3230 only
W1941	0	+ +	0
1945	0	+ +	0
1948	0	+ +	0
1949	0	+ +	0
3146	0	+ +	0
1946	0	+ +	0

plate 5

	~~1946~~	W1948	1949
3174	0	0	0
3156		0	0
		~~W3089~~	+ W3120
W3159	0		0
W3230	5		14
			3140
W2244	0		0
W3127	+		1



27-8

27-10

27-

27-11

N27A

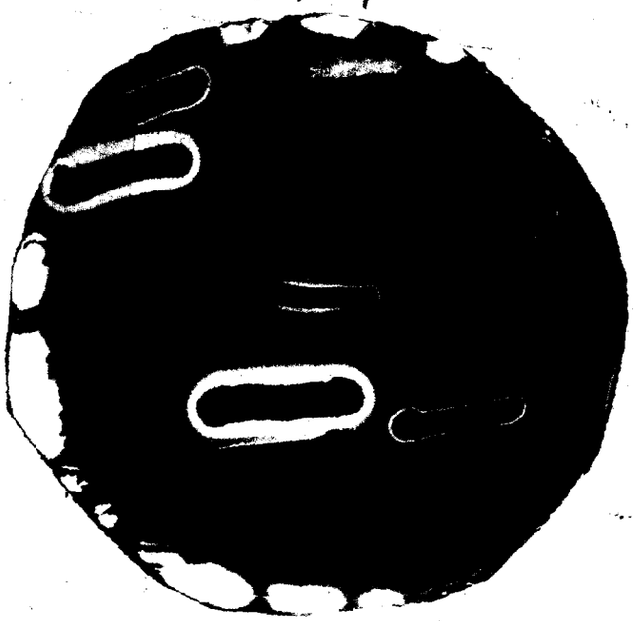
+ 24 hrs

DATE: REF:

	1	2	3	4 6A	5	6	7 6B	8	9	10
	plate 6	w3140	N7-1	w3112	W2243	W2244	w2245	w3238	w3128	w3239
		w3140	O	O	++	O	++	+	±	±
		w3229	O	O	++	O	++	+	±	±
			O	++	+-	++	++	++	±	±
					w3128+w3229					
10	plate 7 —			w3133	w3112	W2243	w2244	w2245	w3238	w3239
	O		3237	++	+	++	+	+	++	++
	O		3240	O	O	++	O	+	++	±
			w3089							
					T6 (B/1)	plate 9			plate 10	
20	plate 8	w811	T6		w811	T6		w811	T6	
	w3133	+		w3089	+		w3153	+		
	w3230	+		w3148	+		w3237	+ (0)		
	w3134	+		w3152	+		w3240	O (0)		
	w3157	+		w3174	+		w3172	+ (0)		
30	w3158	+		w3156	+		w2243	+ (0)		
	w3159	+		w3175	+	Repeated	w2244	+ (0)		
Blue	plate 11			plate 12			plate 13		B mal	
							+,- at 12 hrs.			
	w3127	+		w3147						
	w2245	—		w3149			w3133 +	w3174 +	w3127 +	
40	w3238	+		w3215			w3230 +	w3156 +	w2245 —	
	w3239	+		w3151			w3134 +	w3175 +	w3238 +	
	w3147	+		w3154			w3157 +	w3153 +	w3239 +	
	w1366	+		w3014			w3158 +	w3237 +	w3147 +	
							w3159 +	w3240	w3149 +	
50	plate 14		B mucase				w3089 —	w3112	w3215 +	
	plate 15		xylose				w3148 +	w2243 —	w3151 +	
	plate 16		MTL				w3152 +	w2244 +	w3154 +	
	plate 17		B gal							

N.B. Tomorrow test N13-2 Het against other loci.

27-17



27-16

27-13

DATE: 7/27. Persil readings at 12 hrs. REF:

plate 16 BMTL ~~[illegible]~~ B +- at 12 hrs. MTL

w3133	+	w3174	++	w3127	+
w3230	+	w3156	+	w2245	— —
w3134	++	w3175	+	w3238	+
w3157	+	w3153	+	w3239	+
w3158	+	w3237	wh slow	w3147	+
w3159	+	w3240	wh slow	w3149	+
w3089	+	w3112	wh slow	w3215	+
w3148	+	w2243	— —	w3151	+
w3152	+	w2244	+	w3154	+

plate 14 B sucrose
at 12hrs all negative; slowest growth of w2243.

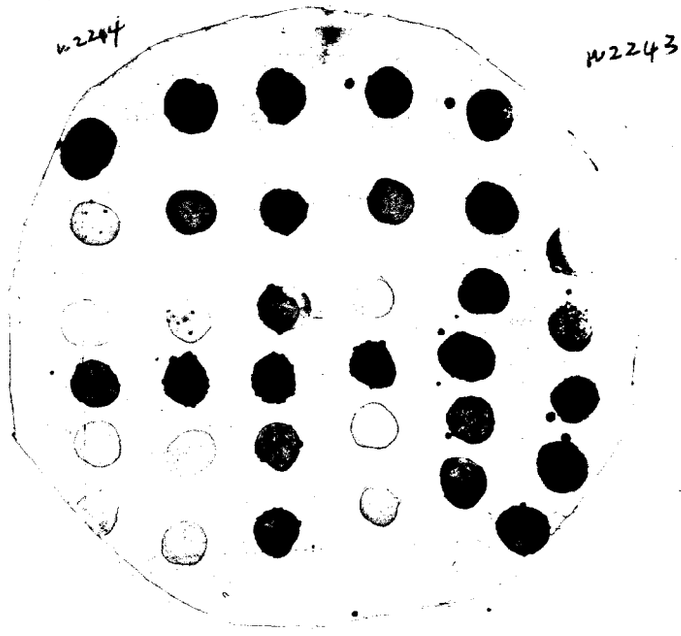
plate 15 xylose all positive; w2245 wh. at 24 hrs all xylose +

plate 17 B gal

w3133	+	w3174	+	w3127	+
w3230	+	w3156	+	w2245	v. slow
w3134	+	w3175	+	w3238	+
w3157	+	w3153	+	w3239	+
w3158	+	w3237	~~wh +~~ slow	w3147	+
w3159	+	w3240		w3149	+
w3089	+	w3112	~~wh +~~ v. slow	w3215	+
w3148	+	w2243	[illegible]	w3151	+
w3152	+	w2244	+	w3154	+

11/29/56
w2243 is glucose —
w2245 is glucose +

28-1B



N28

Test N13-2 against various live loci on M live and test for het

DATE: 9/26/56 REF: N13

	1	2	3	4	5	6	7	8	9	10
	Plate 1.			w2243	w2245	w3112	w3238	w3239	w2244	
			w2243	++	++	++	++	++	++	
			w2245	++	+	+	+	+	6	
			w3164	++	+	0	+	7	0	
10			w3238	++	+	+	+	+	+	
			w3239	++	+	0	+	0	0	
			w3140	++	+	0	+	0	0	
								+ plate 3		
	plate 2			w3133	w3230	w3134	w3157	w3158	w3159	w3089
			N13-2	3	5	5	++	+	1	3
20			w3134	2	7	0	++	+	2	3
			3H3	0	5	3	++	+	0	0
			N23-3	6	10	13	++	+	10	10
			~~N13-2~~							
								plate 5		
	plate 4	w3240	w3148	w3152	w3174	w3156	w3175	w3153	w3237	
30	N13-2	2	2	3	1	3	2	0	6	
	w3134	2	2	2	4	4	0	1	6	
	3H3	0	0	1	0	1	1	2	0	
	N23-3	10	10	10	10	11	10	9	9	
	~~N13-2~~						plate 7			
40	plate 6	w3112	w2243	w2244	w3127	w2245	w3238	w3239	w3147	
	N13-2	8	++	3	5	+	0	++	15	
	w3134	1	++	5	3	+	0	++	2	
	3H3	3	++	0	3	+	0	++	4	
	N23-3	10	++	10	10	+	+	++	10	
50	~~N13-2~~									

+ 24 live

plate 8	W3149	W3215	W3151	W3154
W13-2	8	5	6	1
W3134	1	1	3	3
3H3	1	1	1	3
N23-3	20	8	7	20

DATE: W 3236 P^- Hfr M^- × N6-1 W112 F^- gal $^-$ TL B_1^-

1 2 3 4 5 6 7 8 9 10

~~S gal+M+B_1~~ gal$^+$ (Hfr ? lac$^-$) M^- P^+ ~~lac on S gal+M+B_1.~~

27 A N6-1 in primary. , drop each of W 3236, N6-1 on ~~S~~ S gal + M.+B_1. 29 Replicate on M gal. Spot on B lac.

gal$^+$ lac$^-$ prototrophs cross streaked on Y10 on M lac.

10 One colony N29-6 picked for further tests (Hfr ?).

Streaked on B gal for single colony resolution. N29-2 also picked (Hfr ?).

10/18/56. Test by D(0), D(0) + M.

10/15/56 Both 29-2 & 29-6 are M+ Hfr -1 lac, str$^-$. Purify

20 W29-6 and store as W3221. Purify and retest w29-6.

10/15/56	W3153	Y10	W3089
29-2	+	+	O
29-6	O	+	O
W1946	O	+	+

30

10/18/56. Retest repurified 29-2, 29-6.

	29-2	29-6
W3133	7	O
W3230	+	+
W3089	O	O
W3148	+	+
~~W3189~~		
W3152	+	+
W3174	+	+
W3175	+	+
W3153	+	+

40

Both O.K. = M+ Hfr lac, str$^-$.

50

N. E. Morton
Oct. 4, 1956

Report on lac- study

Preparation of stocks

The plan of this study is to prepare pairs of stocks containing the same lac- allele, the initial member to carry Cavalli's Hfr, M-, and a UV-induced lac-, the other to be a lac- F- prototroph derived from the first by recombination with Y10. For chromosome mapping, each Hfr stock will be modified by selection of V_6^r, a marker closely linked on the left of lac-1. In future, the Hfr stock will also carry P-, one locus for which is reported to lie between lac-1 and V_1^r (Fried, m.s.; her data fit equally well the order P V_6 lac-1). An Hfr P- M- stock W3236 was obtained by UV irradiation of W1895 and is being tested to determine the location of P-. Preliminary tests indicate the order P V_6^r lac or V_6^r lac P.

Pending the development of the P- stock, F- prototrophs and Hfr M- P/ stocks were prepared for the genes lac_1^{y87}, lac_1^{y53}, lac_1^{w112}, lac_2^{45}, lac_4^{w67}, lac_1^{w3229}, lac_1^{w3146}, and for 12 lac- derivatives of W1895 (1940-51). In addition, F- or F/ prototrophs were prepared for lac_3^{w108}, lac_5^{w145}, lac_7^{w133}, and lac^{w3128} (Table 1 and Fig. 1).

In the course of this work, three lac- stocks were isolated which differed in recombination and reversion patterns from the lac- parent. W3159 is a stable isolate from a cross of Y10 with the very highly mutable W1951, and fails to recombine with W1951 and all but one of the apparently single-step lac-1 mutants. W3229 is a spontaneous derivative of W3120 accidentally isolated in serial transfer. It is much more stable than its lac_1^{y87} ancestors and fails to recombine with any of the recognized lac-1 mutants. At present it is the means by which lac-1 is identified, since the lac-1 pseudoalleles have sufficiently high recombination rates to be indistinguishable from unlinked loci in streak tests. W3146 was isolated from a cross of W3129 by W112 in a attempt to introduce lac_1^{w112} into an Hfr stock; it recombines with W112 and all tested lac-1 mutants and is almost certainly

not a derivative of W112, since it remains S^r gal- V_6^r like W3129. (Of the stocks in table 1, the Hfr lac_1^{w112} is the only one not yet prepared.) The origin of the two-step mutants W3229 and W3159 raises questions about the nature and frequency of spontaneous changes in recombination pattern of lac- mutants.

Streak allelism tests

Cross-streaks of Hfr M- lac- and F- lac- prototrophs on M lac plates are convenient tests for allelism, but their interpretation, although clear in most cases, is in others made difficult by too frequent lac+ reversions, especially when they occur in the M- line, and by the relatively low fertility of 3H3, W3164, and W3140. Tests with highly fertile Hfr stocks have been unambiguous.

The lac- stocks tested fall into two groups. The majority fail to recombine with W3229, and are therefore designated lac-1 (Table 2). Of these Y87, Y53, W1950, and W1951 appear to be allelic, but may be separated by their reversion rates, which are in the order Y53 < Y87 < W1950 = W1951 when compared as prototrophs. The latter two stocks are exceptionally revertible and are probably identical, as they were isolated in the same experiment. Similarly, W1948 and W1949 have not been distinguished by recombination and revertibility tests. All other apparently single-step lac-1 mutants recombine with one another. Five lac- genes remain unclassified with respect to locus, since they recombine with lac_1^{w3229}, lac-2, 3,4,5, 7, and lac^{w3128}, as well as with each other. The two recently obtained lac- from W3236 have not been adequately tested. With chromosome mapping tests, some of these unclassified genes will probably be found to be pseudoallelic with known loci.

Intensive allelism tests

Quantitative recombination tests have been deferred until V_6^r P- stocks are available. A few intensive allelism tests were carried out on material at hand, without re-isolation of stocks, so that reversions

in the agar stabs over varying time intervals were confounded with unavoidable reversions in the Penassay broths in which the cultures were grown up and on the M lac plates on which they were tested. Colonies were counted at 24 hrs. to minimize reversions on the plates. Despite the crudeness of these tests, they are of interest in confirming the cross-streak tests and providing a rough measure of reversion rates (Table 3).

W3128 lac- Hist- F+

This stock was received from Borek as a questionable double mutant. Hist + reversions on D(0) remain lac-. Lac- prototrophs were obtained from a cross with W1895. Both hist- and hist + were isolated from lac+ reversions on B lac. All the evidence is consistent with independent origin of hist- and lac-, with hist+ reversions in some lac+ papillae.

Persistent diploids

From H1 lac_1^{y53} colonies were isolated which carried Het, as shown by lacv colonies in the cross with W1940. The lac- parents have been stabbed as N13-2 and the lacv diploids as N13-1.

An attempt was made to test allelism of the lac- segregants of H271, a diploid lac+ which segregates stable and mutable lac-. The original constitution of this stock was lac^{y53}/lac^{w112}, which was lac- in phenotype. Unfortunately, the y53 Hfr tester is of low fertility and the w112 tester has just been synthesized, so a conclusive analysis has not yet been made.

Interaction of lac_1 gal- and lac_1 gal+

E. M. Lederberg reported that cross-streaks of lac_1- gal+ and lac_1- gal- gave a bluish color after 48 hrs. on B lac, but that other lac- loci are negative or give a less intense color. This has been confirmed, the color reaction being much clearer on paper prints than on the agar plate. A gal_1- lac+ tester should be tried. Cells lysed by T6 on B lac agar give a blue reaction, but I was not able to differentiate lac-1 from other loci by this method. In fermentation tests on EMB agar, read at 24 hrs.,

all the lac- prototrophs in this study (with the exception of the mal-1 and gal-2 stocks) behaved as follows:

Locus	mal	mtl	gal	zylose
2 and 3240	slow	slow	≠	≠
3 and 5	0	0	very slow	≠
all others	≠	≠	≠	≠

<u>P1 transduction</u>

Attempts to grow high titer P1 in L broth were unsuccessful on a variety of lp^s stocks. The Swanstrom- Adams confluent lysis plate method is now being tried. As soon as good lysates are made, the transduction system will be explored.

Fig. 1 Pedigree of important stocks

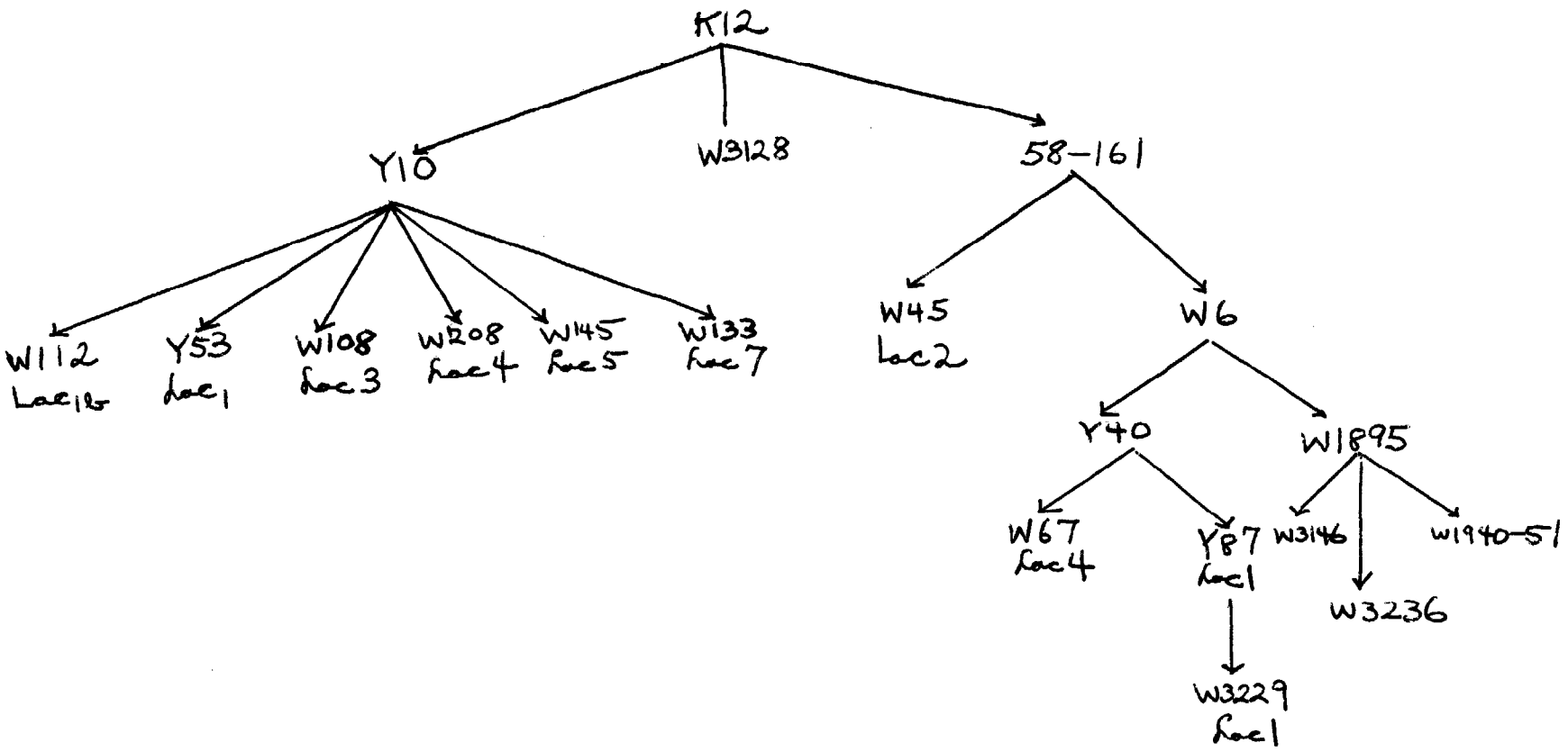


Table 1 Lac Stocks

Source	Locus	Hfr M-	F- prototroph	
y87	1	W3120	W3230	N23
y53	1	3H3 ind. Hfr (JL)	W3134	N2
w112	1	W3221 M+ N6	W3089 mal_1^-	
w1941	1	W1941	W3148	N9
w1945	1	W1945	W3152	"
w1946	1	W1946	W3153	"
w1948	1	W1948	W3174	"
w1949	1	W1949	W3156	"
w1950	1	W1950	W3157	"
w1951	1	W1951	W3158	"
w3146	1	W3146 gal_2^- V_6^r S^r N6	W3175 V_6^r	N6
w3159	1		W3159	N9
w3229	1	W3229	W3133	N1
w45	2	W3164 S^r N5	W3112	
w108	3		W2243	
w67	4	W3140 S^r N4	W2244 F'	
w208	4		W3127	
w145	5		W2245 F'	
w133	7		W3238	N23
w3128			W3239 F'	N7
w1940			W3147	N9
w1942			W3149	"
w1943			W3215	"
w1944			W3151	"
w1947			W3154	"
		Hfr M- P-		
w3237		W3237		
w3240		W3240		

Table 2. Lac_1 recombination pattern

Stocks recombine to give lac+ if the corresponding bars do not overlap.

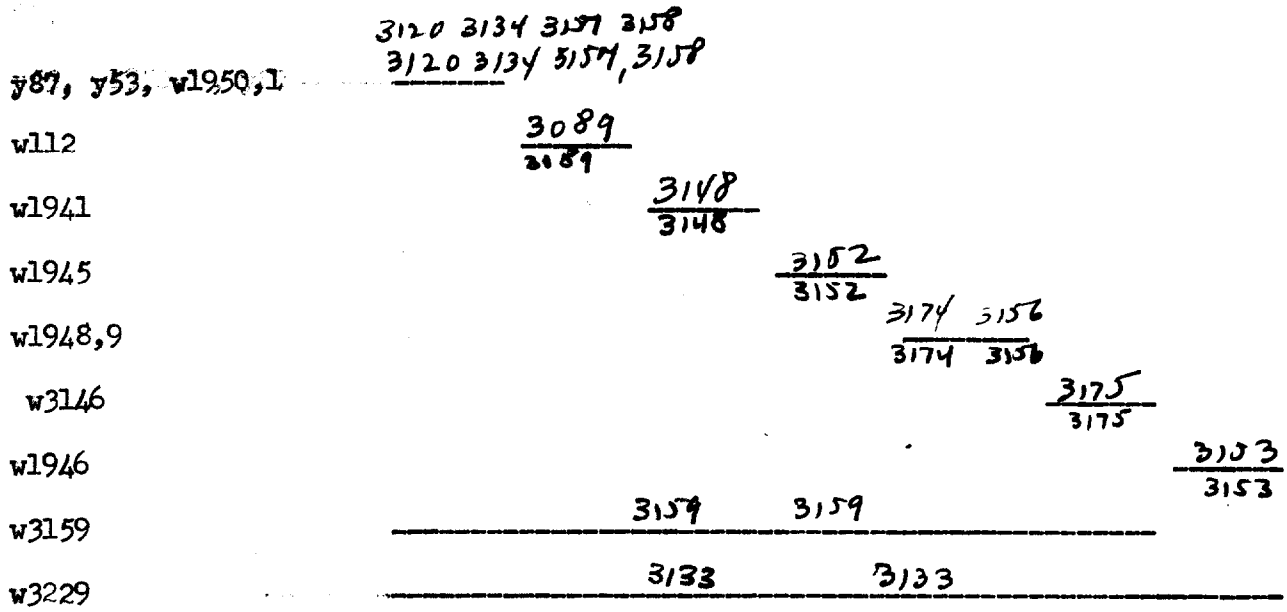


Table 3 Allelism tests

Exper. 1. 0.1 ml. F- and 0.1 ml. Hfr from overnight cultures into penassay. After 4 hrs. plate 0.1 of mix on M lac.

F-	Hfr M- W3229	3H3	W1941
W3133	0	0	0
W3134	22	23	>1000
W3148	1	13	0
W3089	0	—	>1000

Exper. 2. Mix centrifuged, washed with saline, concentrated in saline 1/10. 1.0 ml. of concentrate on M lac.

	W3229	W1941
W3133	3	2
W3089	0	—

Exper. 3. 0.1 ml. F- and 0.1 ml. Hfr in 10 ml. penassay. After 3 hrs. plate 0.1 ml. on M lac.

F-	no Hfr	allelic Hfr		Hfr = W3229
W3133	0	W3229	0	0
W3134	44	3H3	52	50
W3089	0	—	—	0
W3148	0	W1941	0	0
W3152	0	W1945	0	0
W3153	14	W1946	15	11
W3174	0	W1948	0	0
W3156	0	W1949	0	0
W3157	26400	W1950	14200	15400
W3158	29000	W1951	17000	23200
W3159	0	W1951	32	0
W3175	0	W3146	0	0

Table 3 (cont.)

Exper. 4. 0.1 ml. F- and 0.1 ml. Hfr in 10 ml. penassay. After 24 hrs. plate 0.1 ml. on M lac.

F-	no Hfr		allelic Hfr
W3127	32	W3140	153
W3112	0	W3164	0
W3151	0	W1944	0
W3154	1	W1947	1
W3147	3	W1940	2
W3149	1	W1942	0
W3150	1	"	0
W3155	0	"	0

N30

Prep. of high titer P1

DATE: 10/4/56 REF:

Previous attempt to prep. high titer stock in L broth failed; all stocks gave $< 10^8$/ml. (N20).
A subsequent attempt with complement lysis plates gave incomplete lysis, yields less than Lennox P1 (N25).
Plaques on L agar are pinpoint size only against W1485.

11:30 AM. W1895, W1366 into L broth. Rotate.
3 PM. Pour plate 1 drop Lennox P1.
8 PM. Complete lysis (too much phage). Add L ϕ broth.
10/5/56. Decant broth, chloroform, spin down, transfer to frig.
W1895 prep is contaminated (yeasty smell, milky broth. Discard.
W1366 is allright. Much more lysis with these Lp^{+} stocks than with Lp^{s}.
10/8/56. Streaks on Lϕ against W3236.
10/9/56 P1(1366) and P1(1895) give good lysis by comparison with P1 from Lennox. Contaminated with λ (?). P1(W1485) very wk.
Try D(M) + Ca^{++} as broth + 0.1 ml W3089. On rotator.

10/12/56. Lysis from D(M) broth not as clear as from Lϕ layer plates. P1(W1366) agar layer plate produces as much lysis as Lennox phage. B(0) + Ca^{++} plates are not as clean as Lϕ.

W3236(P1) prepared on Lϕ plate. resistant to P1 by comparison with W3236.

11/6/56. Also resistant to T1 (see S. Lederberg).

N31

loc v ? from N13-2 X other loc

DATE: 10/4/56 REF:

	1	2	3	4	5	6	7	8	9	10
W	3112	W2244	W3127		no loc v isolated.					
	31-1	2	3							
10										
20										
30										
40										
50										

Linkage test of W3236 XW945

DATE: 10/8/56. REF:

W3236 = M⁻ Hfr P⁻

W945 = gal₂⁻ lac₁⁻ (T<B₁)⁻

10 A.M. mixture in primary. Plate $X10^{-2}$ on D(0) + prol.

10/9/56. Streak on ¼ for single colony isolation.

Test on 8 gal, 8 lac, and 5 lac M. 60× colonies.

all gal —.

	lac⁻ Prol+	lac⁻ Prol−	lac+ Prol+	lac+ Prol−
plate 1	17	1	IIII	IIII
plate 2	23	11	III	##### ###
Total	40	3	6	14
%	63.5	4.8	9.5	22.2

gal lac P

lac+/lac⁻ 20/43

prol+/+ 17/43

$\frac{20+2(3)}{60}$

lac+ prol+ 6

prol⁻ lac+ 3

gal Prol lac

$\frac{17+3}{60}$ $\frac{3+9}{60}$

Indecisive with respect to proline location.

See next page.

N32A

DATE: REF:

H1

gal I Prod II V_6 III lac IV V_1

doubles I 22.2 + 4.8 = ~~27.0~~

II + III 9.5 + 4.8 = 14.3

I, II, III 22.2 + 9.5 + 2(4.8) = 41.3

gal Prod V_6 lac

20.6 14.2

27.0 14.3 41.3

II. $P^- V_6^+ + P^+ V^S$ = (2.8) + 5.7 + 7.8 + 4.3 = 20.6

III $V_6^+ lac^+ + V_6^S lac^-$ = 5.7 + (2.8) + 4.3 + 1.4 = 14.2

34.8

H2

gal V_6 lac Prod. V_1

I II III IV

III 9.5 + 4.8 = 14.3

I + II lac^+ 9.5 + 22.2 = 31.7

I + II + III 2(9.5) + 22.2 + 4.8 = 46.0

II $V_6^- lac^+ + V_6^S lac^-$ = 5.7 + 2.8 + 4.3 + 1.4 = 14.2

III $lac^- Prod^- + lac^+ Prod^+$ = 5.7 + 1.4 + 5.7 + 7.8 = 20.6

II + III 34.8

34.8

gal V_6 lac Prod

14.2 20.6

31.7 14.3

46.0

Under H1 doubles 3 singles 60

under H2 10 13

6 5

10 13

N33

DATE: 10/9/56 UV induced loc in W3236. REF:

1	2	3	4	5	6	7	8	9	10

~40 single colonies of W3236 picked from B loc into penicillin. 1 ml from 0 overnight cultures per plate B loc. UV 10 sec!

10/11.

	W3266	N33-1	loc-
10	W3267	N33-2	loc-
	~~W3268~~	~~N33-3~~	~~[redacted] dead~~

20

30

40

50

W344

N34

DATE: Transduction with P1 lac REF:

1	2	3	4	5	6	7	8	9	10

10/10/56 .1ml W3236 on S lac + M + 0.1 ml. P1(W1366).
control: 0.1ml W3236, 0.1ml P1(W1366).
10/12/56

W3236 blank
P1(W1366) blank ?
W3236 + P1(W1366) ca. 100 prototrophs gal lac.
Streak out 8 colonies and test remainder on
M lac + Y10 + + lac +
S lac ——
(most colonies reproduced at both) { S lac + meth + + lac +
S lac + M + T6 + + lac +
The colonies streaked out are lac+.

10/16/56. Repeat on B lac. add T6 after 2 hrs.
0.1ml W3236
0.1ml P1(W1366)
0.1ml W3236 + 0.1 ml P1(W1366)
Repeat on S lac + M
1 .05 ml W3236 ——
2 .05 ml P1(W1366) ——
3 .05 ml P1(W1366) + W3236 + (ca. 200 colonies)
} a spot test on S lac + M gives 10 lac+ from W3236 + P1(W1366).

10/17/56. Colonies from 34-3 streaked on B lac. all lac+. Purified
colonies from 10/12/56 stroked on Y10 on M lac for Hfr test. all Hfr.

1 5
2 6
3 7
4 8

P1(W3089) from DM + Ca++ broth on W3140 on M lac.
P1(1366) from agar layer plate on W3127 on M lac ⟸
use W3140 many revertants 1-1 .05ml 3127 2-1 .05ml 3127 many revertants
contaminated w. bacteria 1-2 (W3089) P1 .05ml. 2-2(W1366) P1 .05ml ⟸ O.K.
1-3 .05ml 3127 + P1 (.05ml) 2-3 .05ml W3127 + P1(W1366)
see N37. (over)

10/18/56.

Tests of Y70 = lac$^-$ (from Y53) TCB$_1^-$ F$^-$

DATE: REF:

10/14/56 1 cracked tube from lyophil into Penassay (2 left, O.K.)

10/15/56 No growth. 1 good tube from lyophil into Penassay (1 left).

10/16/56. Lyophil O.K. Isolates streaked against lac 1 Hfr for spots on EMB allelic lac$^-$ F$^-$ control and Y10 control

	+Y70		+allelic F$^-$	Y10
w3229	0	3133	0	++
w3120	0	3230	20	++
29-2	3	3089	0	++
29-6	4	3089	0	++
1941	3	3148	0	++
1945	5	3152	0	++
1948	2	3174	0	++
3146	6	3175	0	++
1946	1	3153	10	++
w3236	15	Y70	10	++

10/20 Repeat using new isolates of Hfr and ~~[illegible]~~ Y53 control.

10/21.

	Y53	Y70	Y10	
w3229	0	0	+++	
w3120	0	0	+++	
w3221	+	+	+++	35-1
w1941	++	+	+++	
w1945	++	+	+++	
w1948	+	+	+++	
w3146	+	+	+++	35-2
w1946	+	+	+++	
w3236	++	++	+++	
W3140	+	+	+++	
~~[illegible]~~				

∴ Recombination pattern of Y70 not different from Y53

N36

W3120, W1950 UV
look for lac stable in B lac.

DATE: 10/16/56 REF:

10 sec. UV (1 drop). 12 plates of W3120 (1), 12 of W1950 (2), 10 of W3236 (3)

10/18 lac-stable colonies of W1950, W3120 into Penassay for allelism test. Two doubtful lac streaked on B lac.

10/19. One lac from W3236 = N36-1 (W3268) streaked on B gal for stab.

Test of lac stable W3120, W1950 controls

		36-1A	1-B	1-C	36-2A	36-2B
36-1	W3089	O	+	O	O	O
2	W3148	O	+	O	O	O
3	W3152	O	+	O	O	O
4	W3156	O	+	O	O	O
5	W3175	O	+	O	O	O
6	W3153	O	+	O	O	O
7	W3133	O	+	O	O	O

10/21. Four colonies (2 from W3120) 2 from W1950) clean on all indicators. One other clean on W3175 only. Overnight broths into M lac.

	1A	1B	1C	2A	2B
Y10	++	++	++	++	++
W3133	O	+	O	O	O
W3175	O	++	O	O	O
W3153	O	++	O	O	O

Streaks of 1A - 2B onto fig.

10/23 Discard 1B. Stab others for single colonies on B-O.

~~1B 1A 2A 2B~~.

1A = 3269
1C = 3270
2A = 3271
2B = 3272

W1366 lac_1^{w112} $V_6^r V_1^r$ TLB_1^- × W3236 $M^- H f^r P^-$

DATE: 10/19/56 REF: N34

10/18/56. Mutation, phage sensitivity and resistance of single colonies OK.

10/19 1 drop of each on D0+ pool. Spread from this to 2 other plates.

10/20 in frig. 10/21 streak out single colonies on B-0.

10/22 Pick independent on D0+ pool. 10/23 in Frig.

10/25. Replicate on D0, B lac, Streak on B-0 + T1, B-0 + T6.

N37A

Tested 200 discrete colonies from N37. Clear replication of long streaks on B-O+T1, T6.

DATE: 10/29 REF:

plate no.	a V6	b Lac	c Prol	d V4	e	5	V6	Lac	Prol	V4	10
11											
13											

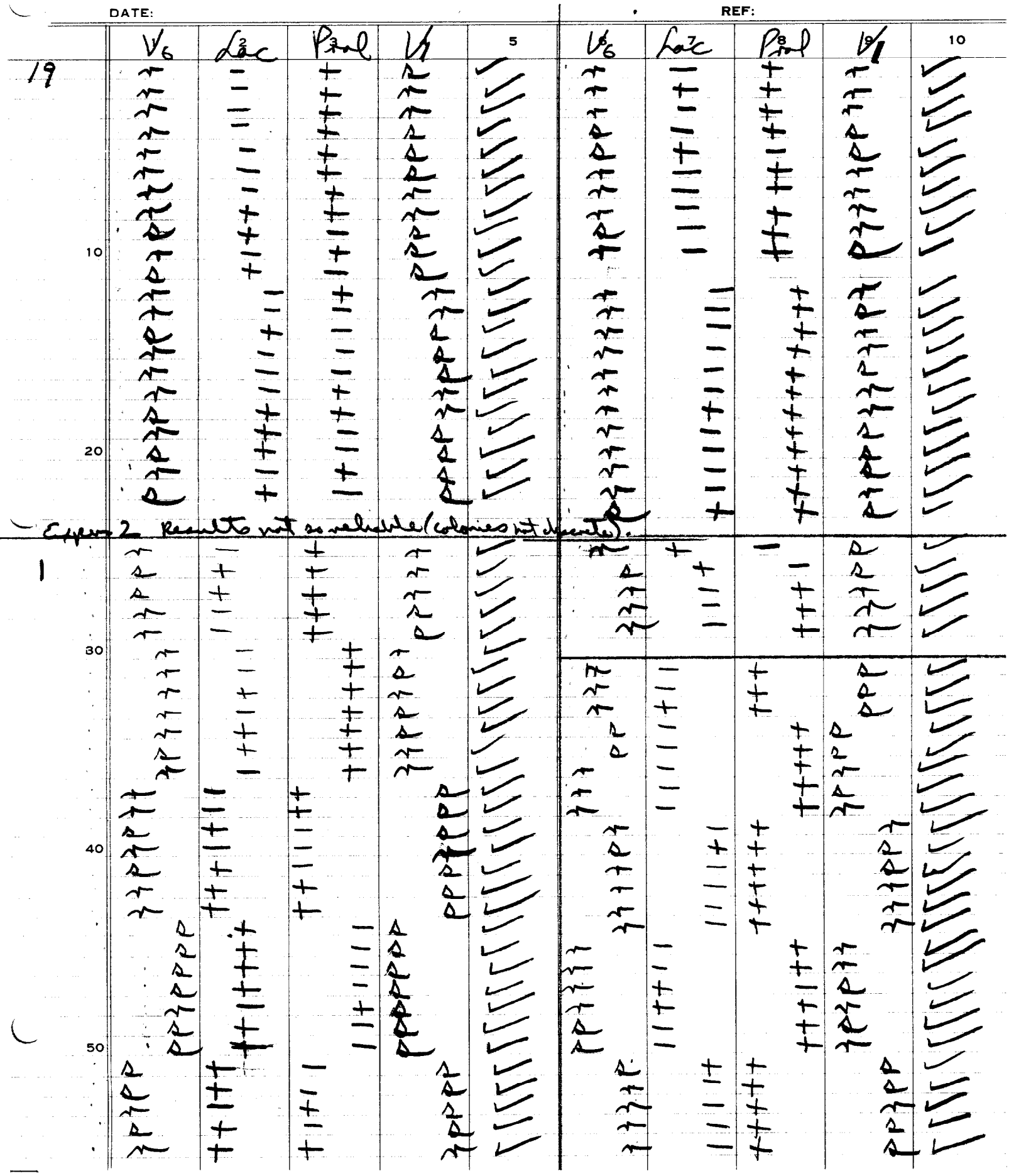
N37B

DATE: REF:

	V6	Lac	Pnl 3	V4	5	V6	Lac 7	Pnl 8	V1	10
15										
10										
20										
17										
30										
40										
50										

N37C

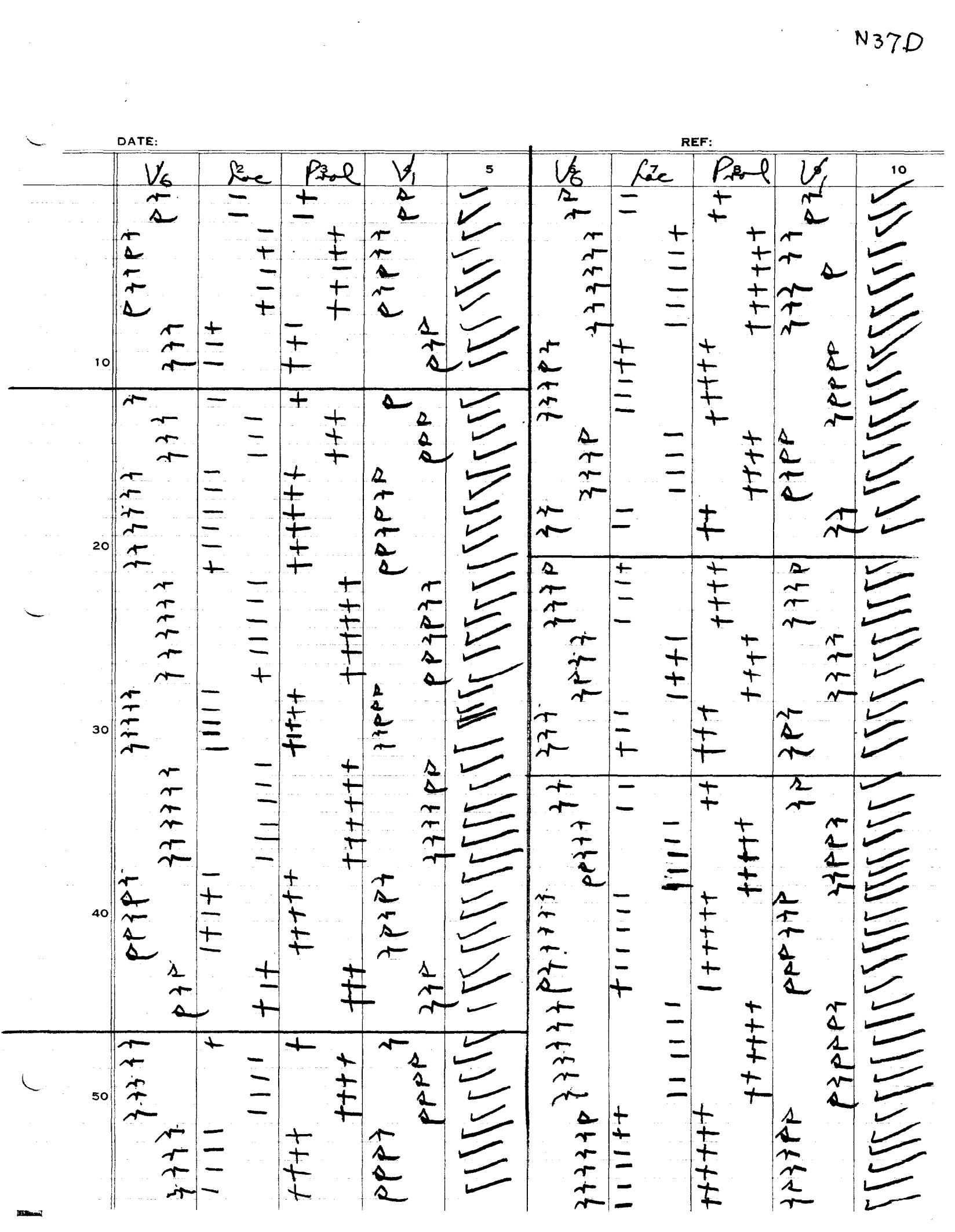
DATE: REF:



N37D

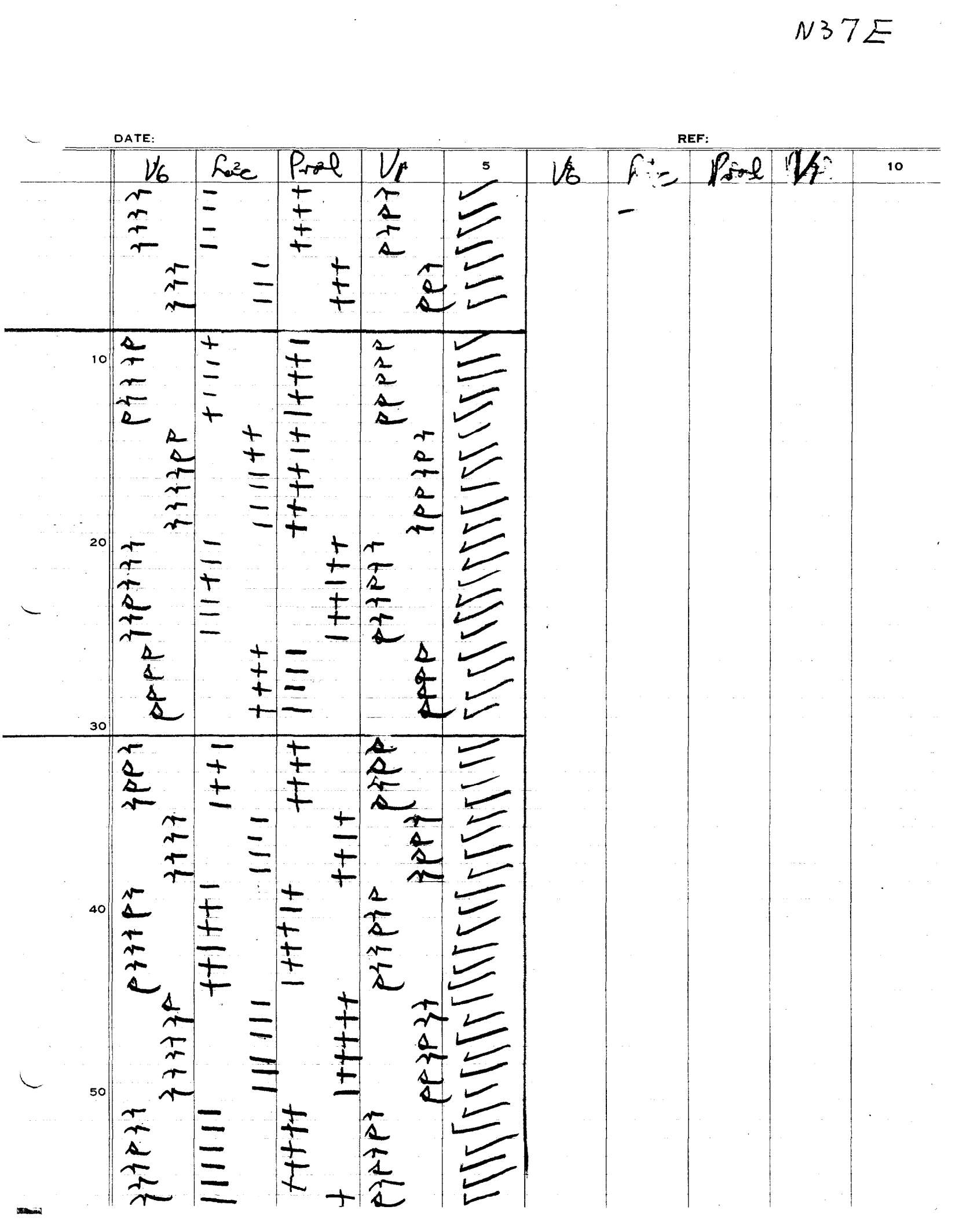
DATE:

REF:



N37E

DATE: REF:



Summary of experiments 21, 32, and 37

mapping Δ, V_6, lac_1, Prol, V_1, (TL).

DATE: 11/2/56 REF:

All experiments were carried out by plating the two parents on D(0) + proline. Except in experiment 37A, there was contact between some of the colonies. Under the denser growth of the other experiments there was apparently some selection for P+.

Exper.	P−	P+	Total	%	lac+	V_6^S	V_1^S	%
21	28	113	141	19.9	35	31	~~31~~	
32	17	46	63	27.0	20	—	—	
37A	84	134	218	38.5	79	76	136	
37B	32	192	224	14.3	61	52	128	

assuming independent selection for other factors [an obvious oversimplification because of linkage to Δ, P and (TL)], an unbiased estimate of the recombination fraction between two markers is gotten from the determinant γ of their 2 × 2 table, viz.

$$\gamma = \begin{vmatrix} a & b \\ c & d \end{vmatrix}$$

, where a and d are the recombinant classes and the recombination fraction is estimated as

$$\hat{\theta} = \frac{\sqrt{\gamma}}{1+\sqrt{\gamma}},$$

$$\sigma_{\hat{\theta}}^2 = \frac{\theta^2(1-\theta)^2}{4}\left\{\frac{1}{a}+\frac{1}{b}+\frac{1}{c}+\frac{1}{d}\right\}$$

$$I_{\hat{\theta}} = 1/\sigma_{\hat{\theta}}^2$$

Summary experiments 21, 32, & 37

37A weighted X2

DATE: REF:

Region	Exper.	a_2	b_2	c_4	d_6	$\frac{ad}{bc}$	$\sqrt{\frac{ad}{bc}}$	$\hat{\theta}^8$	$I\hat{\theta}$ product	$\hat{\theta}^{10}$ terminal
$\Delta - V_6$	21	31/141						~~.220~~		
	37A	76/218						.349 ✓		.349
	37B	52/224						~~.232~~		
$\Delta - loc_1$	21	35/141						~~.248~~		
10	32	20/63						~~.317~~		
	37A	79/218						.362 ✓		.362
	37B	61/224						~~.272~~		
$\Delta - P$	21	28/141						~~.199~~		
	32	17/63						~~.270~~		
20	37A	84/218						.385 ✓		.385
	37B	32/224						~~.143~~		
$\Delta - V_1$	37A	136/218						.624		.608
	37B	128/224						.571		
$V_6 - loc$	21		20/141 = .142			.042591	.20637	.171		
30	37A			33/218 = .151		.03569	.18892	.159	.165	.148
	37B			33/224 = .147		.04172	.2042	.170		
$V_6 - P$	21		29/141 = .206			.14296	.3781	.274		
	37A			57/218 = .239		.10913	.3303	.248	.245	.213
	37B			38/224 = .170		.06962	.2639	.209		
$V_6 - V_1$ 40	37A			88/218 = .404		.24573	.4957	.331	.360	.430
	37B			108/224 = .482		.5111	.7149	.417		
$loc - P$	21		29/141 = .206			.1226	.3501	.259		
	32		7/63 = .143			.03214	.1793	.152	.209	.193
	37A		47/218 = .216			.08331	.2886	.224		
50	37B		41/224 = .183			.05145	.2268	.185		
$loc - V_1$	37A		81/218 = .372			.31216	.5587	.358	.369	.398
	37B		101/224 = .451			.41082	.6409	.391		

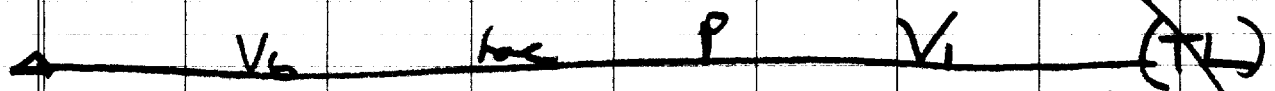
Summary of Experiments 21, 32, and 37

DATE: REF:

Region	Exper	a	b	c	d	$\frac{ad}{bc}$	$\sqrt{\frac{ad}{bc}}$	ϕ	I_ϕ	10
$P - V_1$	37A		$\frac{68}{218} = .312$			.08535	.29215	.226	.220	.357
	37B		$\frac{102}{224} = .446$			.0695	.26363	.209		
$V_6 - (T2)$	37A	$\frac{82}{218} = .376$								.376
	37B	$\frac{96}{224} = .429$								.394

Binomial

Estimates assuming $\theta = w - \frac{1}{2}w^2$



.349	.148	.193	.357	.394		$\Sigma = 1.441$
.362					.497 − .124 = .373	
.385					.690 − .238 = .452	
.608					1.047 − .548 = .499	
	.148					
	.213				.341 − .058 = .283	
	.430				.698 − .244 = .454	
	.651				1.092 − .596 = .496	
		.193				
		.398			.550 − .151 = .399	
		.638			.944 − .446 = .498	
			.357			
			.615		.757 − .282 = .475	
				.394		

$w - \frac{1}{2}w^2$

DATE: 10/19/56 REF:

1	2	3	4	5	6	7	8	9	10

M lac ± W3140 (+meth) ± P1(W1366) 1 drop ea.

38-1	M lac + W3140	0	38-4	~~…~~ P1(W3089)	0
38-2	+ P1(W1366)	0	38-5	W3140 + W3089	0
38-3	+ W3140 + P1(W1366)	ca. 300	~~38-6~~		

10/21 Streak from 38-3 on B lac.

10/22. Several possible heterozygotes streaked on B lac.

10/23 ~~After~~ Second streak (Some lac⁻ in first, probably parental).

10/24 No lac⁻ in second streak; some bull's eye colonies  .

10/25 Streak from periphery of bull's eye. ~~w6~~ all lac +.

11/6/56. M lac + Meth + W3140. 11/7/56 Streak on ~~B lac~~. Streaked on B-0 + T6 and B lac + T6. 1/73 colonies V_6^{r} (on B-0), This streaked on B-lac + T6, with several possible lac⁻ sectors from V_6^{s}.

11/15/56. The V_6^{r} colony lac⁺. No heterozygotes found. Lac⁺ and lac⁻ colonies streaked on B lac + T6.

11/16/56. all V_6^{s}.

N39

DATE: 10/19/56 W3236 UV on Blue REF:

2 drops, 12 sec., 17 plates. 10/21/56 No lac.

Repeat, 15 plates. 11/28/56.

11/29/56. Three lac. Streaked on B-O for stab.

39-1 mal- glucose-?

39-2

39-3

N40

allelism Tests

DATE: 10/21/56 REF:

O — M	loc	2	3	4	5	6	7	8	9	10
	w3237	w3240	w3266	w3267	w3268	w3236				
w3133	++	O	++	++	++	++				
w3112	+	O	+	++	++	++			40-1	
Y10	++	O(3)	++	++	++	++				
w2243	—	—	—	—	—	—	—	++		
~~w2244~~ w3127	++	O	++	++	++	++				
w2245	—	—	—	—	—	—	++		40-2	
w3238	++	O	++	++	++	++				
w3239	++	O	++	++	++	++				
w3147	++	O	++	++	//	//				
w3149	++	O	++	++	//	//			40-3	
w3215	++	O	++	++	//	//				
w3151	++	O	++	//	//	//				
w3154	++	O	++	//	//	//				

10/22. w2243 and w2245 are largely reverted to loc+.

galactosidase tests N#8

DATE: 11/2/56 REF:

Hereafter use YZ + ½% lac + ½% glycerol.

YZ + lactose broth. ^ Lac stocks streaked on B-O.

11/4/56 Single colony stabs from B-O into YZ + lac. Rotator overnight. Streak on Blac.

AS Spin down. ~~[illegible]~~ Discard supernate. ^ add 1ml H_2O + 1-2 drops benzene to ~~[illegible]~~ pellet, shake well. 0.1 ml of this mix + 2 ml H_2O + 0.1 ml ONPG (30 mg/20 ml.)

Read at (10 min.)

		ONPG	Blac			ONPG	Blac	
lac+	W3236	++	+		W1942	O	✓	
1	W1949	+	✓		W3240	O	✓	
	W3266	O	✓		W1940	O	✓	
1	W3159	+	✓	1	W1950	+	+ pinkish	
*	W3270	O	✓	lac cst.	W3239	+	✓	also + from glucose
1	W3221	+	✓		W1943	O (±)	✓	W3244 also + from glucose
*	W3269	O	✓		W3237	O	✓	streaks from Blac on lac.
1	3H3	+	✓	1	Y70	+	✓	
1	W1941	+	✓	1	W1951	+	✓	
	W1947	O	✓		W3272	O	✓	
1	W1948	+	✓		W3164	O (±)	✓	
*	W3267	+	all + stab no good / single lac+		W1945	+	✓	
1	W3120	+	✓					
*	W3268	+	all + stab o.k.(1)					
/	W3146	+	✓					
*	W3229	O	✓					
*	W3271	O	✓					
re-?	W3238	~~+~~ O	~~+ pinkish~~ ✓					
1	W1946	+	✓					
	W1944	O (±)	✓					

± = very faint yellow tinge.

1) Of 6 two-step lac-1 mutants, only one (W3159) is ONPG+. Also, Y70 is ONPG+.

2) all recognized ^single-step lac-1 mutants are ONPG+.

3) other ONPG+ are W3267 & W3268.

~~Also W3238 lac-1 is ONPG+.~~

Reisolate ^W2243, W2245, W3268, W3267, W3238, W3140, and Davis cst lac.

0.15 ~~+ stab~~ again o.k.

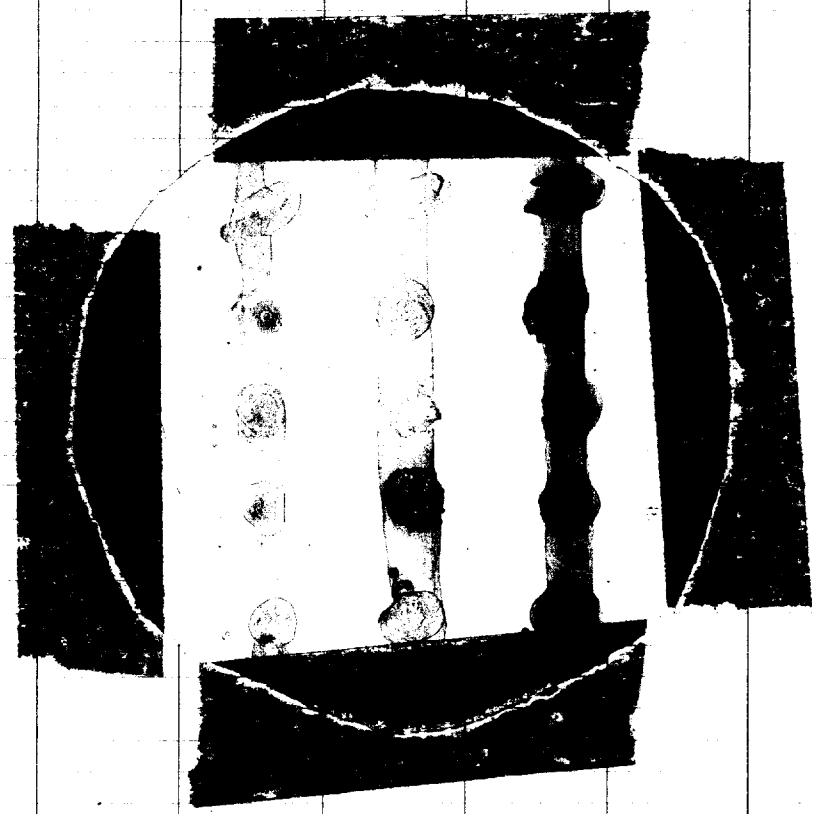
11/23/56. W3159, W3267, W3268 rechecked; all ONPG+.

N 42

allelism tests of w3269-72, w3240 against w3127

DATE: 11/3/56 REF:

1	2	3	4	5	6	7	8	9	10
On M lac			w3133	w3127	Y10				
	w3269		O	O	++				
	w3270		O	O	++				
	w3271		O	O	++				
	w3272		O	++	++				
	w3240		O	O	+ (ca. 50).				



N 43-A

DATE: 11/28/56 REF:

Hfr and F- grown 3 up overnight 5 from single colonies of B-D 9 10

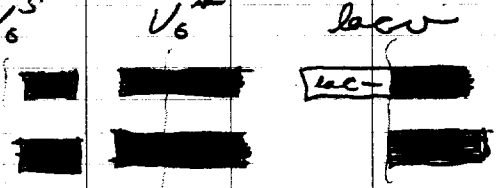
7.5 ml + 1 ml Hfr + 1 ml F-

1 W3229 V6 X W3133
2 W3229 V6 X W3127
3 W3229 V6 X W2244
4 W3140 V6 X W3133
5 W3140 V6 X W3127
6 W3140 V6 X W2244
7 W3133
8 W3127
9 W2244

Make matings 1-6 in duplicate (7.5 ml. parents + 0.1 ml Hfr + 0.1 ml F-. call duplicate X & Y. Put in rotator.

A after 1 hr. add 1 loop T6 to X. Put in rotator. after 4 hrs. dilute 10 x 10 x 10 .... and plate 0.1 ml from each dilution tube on M gal and M lac. count colonies at 24 hrs.

B Dilute X x 10 and x 100 and plate 0.1 ml on M lac. Streak colonies on M lac. Replicate on B lac + T6 and S lac + T6.



Streak out lacy colonies and isolate duplicates.

C. after 12 hrs. plate from X to give 100 colonies per plate of S lac. (i.e. V_6 P.C.a). Incubate for 48 hrs. or more and pick lac stable colonies. Test for allelism with parents and pick lac x lacy. Recombination should be suppressed only between x and y. lac-1 x and lac, y should give ONPG double mutants, lac 1 x and other lac + should give ONPG -.

4 for M^- (P-) V_6^- lac$^-$ X V_6^S lac$^-$ prototrophs in primary

2 nro. add T6

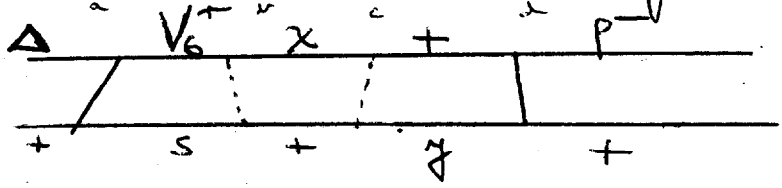
4 hrs. Plate serial dil. on M lac and M gal.

$\theta = \frac{\text{M lac count}}{\text{M gal count}}$ D-O

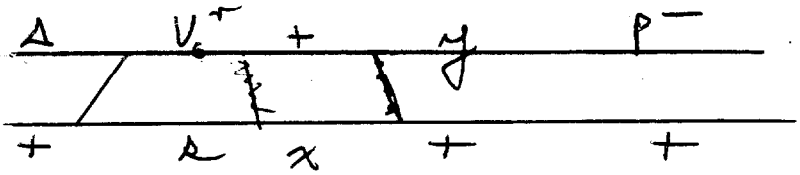
$\theta = \frac{a\,b\,c\,d}{n}$

Cross 1

- M lac D-O
- D-O + gal

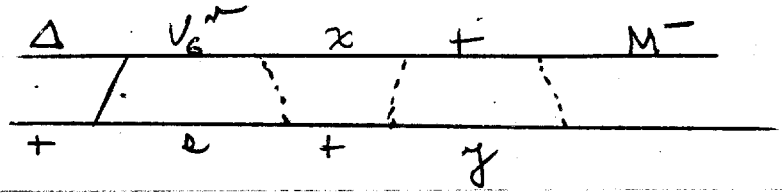


Reciprocal cross



$\theta = \frac{a\,c}{n}$

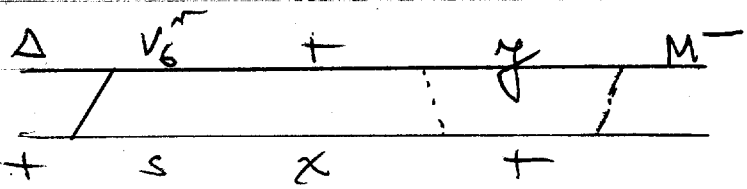
P+



on M lac D-O

$\frac{lac}{n}$

~~D-O + gal~~



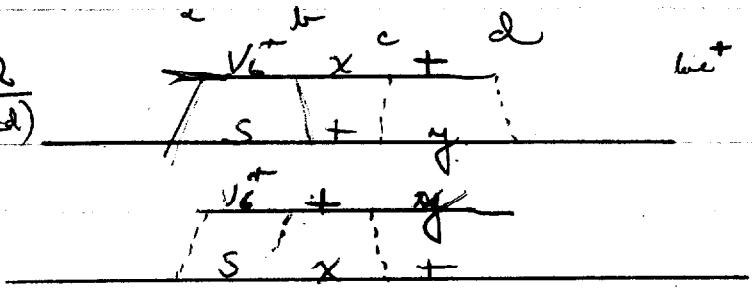
$\frac{a}{n}$

Transduction

+T6

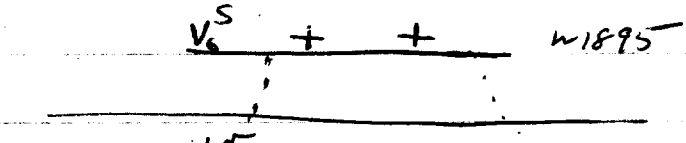
$\theta = \frac{\text{M lac count}}{\text{M gal count}} = \frac{a\,b\,c\,d}{n(b+c+d)}$

$\frac{ac}{a(b+c+d)}$



lac$^+$ using V_6^S

F^- x^+



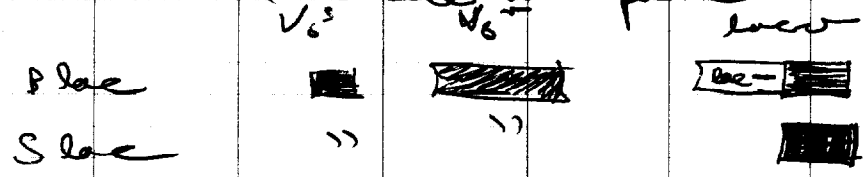
Streak from M lac

M lac into lac + T6

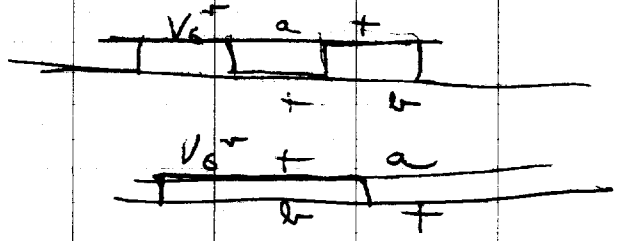
Streak V_6^S colonies onto B lac + T6 and replicate on B lac + T6.

DATE: REF:

D. Transduce lac^-_x V_6^s F^- prototroph with lysate from Hfr-M+P$^-$ $lacy^-$ V_6^r on M lac. Streak colonies on M lac and replicate on B lac + T6 and S lac + T6. Streak out lac+ colonies and isolate heterogenote.



E. Transduce lac^-_x V_6^s F^- prototroph with lysate from Hfr-M+P$^-$ $lacy^-$ V_6^r ~~~~ in broth after 1 hr add T6. after 4 hr. plates on M lac and M gal from serial dilution tube. Count colonies. M lac V_6^r transduction, lac$^+$ transduction, C.O.



F. Plate from E on B lac$_x$ to give 100 colonies per plate. incubate for 24 hrs. and pick lac$^+$. Test for allelism with parents and pick lac$_x$ lacy.

N43-C

DATE: 11/30/56 REF:

A incubate X and Y in refrigerator overnight. Plate 2 Y and 4 X on M mal and M lac.

1	0.1 ml	
2	X 1/11	
3		X 1/11

0.1 ml. A-1, 3, 5, 6, 7, 8, 9

B. Y 2,3 and X 4,5 on M lac.

12/3/56. Incomplete lysis of V_6^S in A, as shown by plaques in streaks on B lac.

B/

	count
1-1	0
1-3	0
2-1	TMC
2-3	ca. 200
3-1	TMC
3-3	ca. 200
4-1	ca. 300
5-1	ca. 68
6-1	0
7-1	0
8-1	70
9-1	0

growth of plated recombinants on M lac. Count at 24 hrs.

∴ w3127 allelic to w3140, but with greater mutation rate rel M.

Try UV on w3127 to obtain lac stable 2-step mutant. Can be used with w3150 to obtain 2-step mutant by recombination.

DATE: REF:

	1	2	3	4	5	6	7	8	9	10
	A	count M lac	M mal							
	1-1	0								
	2-1	TMC	TMC							
	2-2	ca. 2000	TMC							
10	2-3	ca. 500	TMC							
	2-4	55	14 (dry plate)							
	2-5	3	134							
	2-6	0	33							
	2-7	0	0							
20	3-1	TMC	—							
	4-1	ca. 300	TMC	← ca. 200 plaques.						
	4-2	42	TMC	"						
	4-3	4	TMC	"						
	4-4	0	TMC	"						
30	4-5	0	ca. 600							
	4-6	0	335							
	5-1	85	—							

∴ suggested order is

V_6^+ lac 4 + M^-

S + lac 1 +

B. Streak directly from B 2-3, B-3-3, and B-4-1 on M lac.

12/5. Poor for replication on B lac & M lac + T6.

Better to streak from recombinati plate onto B lac, isolate host, and pick single lac+ colonies to streak against T6 on B-0, B lac, or M lac (on plate of each). Pick single colonies into 1 ml. pearsay, streak for this against T6.

Several possible lac+/4 heterozygotes streaked out ~~against~~ on B lac from 3140 × 3133 (B4). Single colonies picked. host. detected O.K.

(over)

Two lac^{st}, two lac^{m} picked. Streaked on M mal & B lac.
all four colonies prototrophs. Streaked on B-O agar at T6.

	T6
lac^{st}	S
lac^{st}	S
lac^{m}	S
lac^{m}	S

all four colonies lac^{w3133}.

Cross streaked on M lac

	3229	3140	3271
lac^{st}	O	O	O
lac^{st}	O	O	O
lac^{m}	O	O	O
lac^{m}	O	O	O

N44

Search for heterogenotes with lac_1^{Y53}

DATE: 12/3/56 REF: N13

1	2	3	4	5	6	7	8	9	10

lac_1 (het?) TLB_1^- from $\frac{W3236}{W3286}$ × Hfr V_6^r lac^- M^- testers (W3267 is V_6^s). 0.1 ml. of each parent in DM + 1% lac. Streak on B lac.

1941	no lac+	
3221	"	
3143	"	
3229	"	
1949	"	
1946	"	
1947	"	Try again
2243	"	
3140	"	
3240		
3237		
3266	✓ het streak out	
1945	no lac+	
3146	"	
1942	"	
1943	"	
3239	"	
3164		streak out
2245	no lac+.	
3238	"	
3268	streak out	
3267	no lac+	
1940	"	
1944	" but see other plate. Streak out.	

DATE: 12/4/56 REF:

9 A.M. 2 ml. <Ø .6% agar + 0.1 ml. 4 hr V_6^r + 0.1 ml P_1 kc (10^7/ml).
Pour on <Ø plate (ca. 45 ml/plate).

4 PM. add 4 ml <Ø broth.

8 pm. decant. add chloroform. Put in ice-box.
W3229 showed discrete clear plaques.

12/5/56 C on <Ø.

	P_1	W3236	W3236P_1^+	▭	
46-1	1940	S	r	O	
	1941	S	r	⊕	
	3229	S	r	O	
	1485	S	r	O	
	3146	wh. lysis	r	+	add more chloroform.
46-2	1946	S	r	O	
	1147	S	r	O	
	3164	S	r	O	
	3271	S	r	O	
	3240	S	r	O	
46-3	3159	S	r	O	
	3143	S	r	O	
	3237	S	r	O	
	1943	S	r	O	
	1945	no lysis	r	O	

↑ from B-0 pour plate.

N47

Tests on V_6^r and F^-X^+ derivatives of new lac⁻ stocks

DATE: 12/5/56 REF:

1	2	3	4	5	6	7	8	9	10

47-1 Mutation. Spot on B gal plate for replication to B mal, B lac, B glucose.

3237	3270	3112
3240	3271	2243 wh. to +
3266	3272	2244
3267	39-1 wh.	2245 wk.
3268 wk.	39-2	3238
3269	39-3	3239

47-2 V_6^r stocks streaked against T6.

3164	r	39-1	(only a few colonies on streak)	3239	r	1941	r	1942 r	3266 r
2243	r	39-2	r	3120	r	1945	r	1943 r	3268 r
3140	r	393	r	3143	r	1948	r	1944 r	3269 r
2245	r	3272	r	1950	r	1949	r	1947 r	3270 r
3238	r	3236	r	1951	r	3146	r	3237 r	3271 r
3239	r	1940	r	3221	r	1946	r	3240 r	

	3229 V_6^r	3269 V_6^r	3270 V_6^r	3271 V_6^r	3272 V_6^r
3112	++	(4)	(1)	(3)	++
~~2243~~	~~+++~~	~~+++~~	~~+++~~	~~+++~~	~~+++~~ revertants
	++	0	0	0	++
~~2244~~	~~++~~	~~0~~	~~0~~	~~0~~	~~++~~
3147	++	0	0	0	++
3149	++	0	0	0	++
3215	++	0	0	0	++
3151	++	0	0	0	++
'54	++	0	0	0	++
3238	++	0	0	0	++
3239	~~~~				
~~2245~~	~~++~~	~~++~~	~~++~~	~~++~~	~~++~~

allelism tests of V_6^r x F⁻

1940	✓
1942	✓
1943	+++ } F⁻ revertants
1944	+++ }
1947	✓
3229	✓
3120	✓
3143	✓
1950	recombination
1951	✓
3164	✓
3269	✓

322 [no good]
1941 ✓
1945 ✓
1948 ✓
1949 ✓
2245 alt+
3146 + discard this culture
1946 ol.
323 ? ?

3270 ✓
3272 ✓

nutrition test

DATE: REF:

47-1 replicated on several sugars

	1	2	3	4	5	6	7	8	9	10
	B gal	B lac	B mal	B mtl	B glu.	B ara				
3237	+	−	+	+/	+	+				
3240	+	−	+	+/	+	+				
3266	+	−	+	+/	+	+				
3267	+	+	+	+	+	+				
3268	−	−	−	−		−	very little growth			
3269	+	−	+	+	+	+				
3270	+	−	+	+	+	+				
3271	+	−	+	+	+	+				
3272	+	−	+	+	+	+				
3[illegible]	−	v. wk.	−	−	−	−	like 2243 in pattern			
39-2	+	−	+	+/	+	+				
39-3	+	v. wk.	−	+/	+	+				
3112	+	−	+	+/	+	+				
~~2243~~	~~+~~	~~+~~	~~+~~	~~+~~	~~+~~	~~+~~	revisited			
2244	+	−	+	+	+	+				
2245	−	−	−	−	+	+				
3238	+	−	+	+	+	+				
3239	+	−	+	+	+	+	(only few colonies replicated)			

allelism test on m lac.

Hfr V6	Y10	allelic F−	w3133	~~allelic F−~~
3229	++	0	0	
3120	++	14	0	
3113	+	6	0	
950	++	++	++	discard, lac+ 1956 V6
1951	++	50	3	
3221	++	20	?	
1941	++	3	0	

Hfr V6 3159	Y10 7	allelic F 3229 7	w3133 +3229 7
1945	++	3	1
1948	++	0	0
1949	++	13	0
3146	++	3175 F−xF 0	0
1946	++	0	0
3269	++	0	0
3270	++	0	0
3271	++	0	0
3272	++	0	0

N47A

DATE: 10/12/56 REF:

	1	2	3	4	5	6	7	8	9	10
F⁻X⁺		+Y10	+allelic diff V6	allelic diff						
39-1	±	±	—	±			To prepare			
3272	—						39-2			
3271	—						39-3			
32700	—						3268			
3269	—						3267			
3237	—						3266			
	3269 V6	3270 V6	3271 V6	[illegible]			3240			
3133										
3112 20										
2244										
3147										
3149										
3215										
3154 30										
3238	10	10	10	10						
Y10	++ V6 S	++ V6 S	++ V6 S							
47-1 3133										
3112										
2244										
Y10										
3147										
50		39-1 V6	39-1	3236	—					
	39-1 FXT			+						
	Y10	++	++	++						

O slight growth

DATE: 12/13/56 REF:

0.1 ml 3120V6 + 0.1 ml. F⁻X⁺ in 7.5 ml. prossay. All from week-old broths. Cross + streak on M lac + T6.

	1	2	3	6 (S)	7 (r)	8
1	3230		24			
2	3089		20 (dry plate)			
3	3148		50	31	18	✓
4	3152		100	36	18	✓
5	~~3174~~	3153	300	42	18	✓
6	3156		100	3[illegible]	14	✓
~~7~~	~~3175~~		~~300~~			
8	3146 F⁻		300	45	15	? ✓
9	3244		30 (dry plate)	15	11	

12/14. 0.1 ml 1–9 in 10 ml. H_2O; plate 0.1 ml on M lac.

Plate 0.1 ml. from 3139, 3089, etc. To half-plate add 1 drop P1 (3143 0V6).

	1	+ P1	control
11	W3139	0	0
12	w3089		
13	w3148	0	0
14	3152	28	14
15	3156	30	10
16	3175	22	0
17	3146 F⁻X⁺	40	10
18	2244	on M mal	repeat
19	3153	on M mal	repeat

10. also 1 drop on B lac. (12)

~~W3229 w3269 w3270 w3271 w3272 w3229 w3269 w3270 w3271 w3272~~

w3112
w2244
w3229
3239
50
38 – 1 F⁻X⁺

12/14/56 Repeat with fresh 24 hr. broths from B-4 single colony. (see N48A).

W-236 V6
V6

DATE: 12/14/56 REF:

P 0.1 ml. W3120 V_6^r + 0.1 ml F⁻ lac⁻ in 10 ml. H_2O. Plate directly 0.1 ml. on M lac. 12/16/56. Streaked & cross-streaked on B lac, S lac, M lac + T6

		1	2	3 (B lac)	4	5 (M lac)	6	7 (S lac)	8	9	10
Y53	1	3134	17	r		Streaked only on S lac and [illegible] 2	14		S		
W112	2	3089	105			17	28	✓			
1941	3 (10)	3148	161			11	32	✓			
1945	4	3152	176			11	35	✓			
1949	5	3156	ca.200			17	32	✓			
~~3146 F⁻~~	~~6~~	~~3175~~	~~ca.300~~			~~43~~	~~0~~				
3146	7	3146F⁻	ca300			14	36	7	39	21 ✓	75
1946	8 (20)	3153	125	12	20	[redacted]		11	30		23/50 ✓
	9	2244	25			9	10				
	10	3147	ca.400	14	36	15	34	9	30		
	11	3149	ca.400			12	41	13	38		
	12	3215									
	13 (30)	3151									
	14	3154									
	15	3112									
	16	3238									

17 1 drop P1 (W3236 V_6) on W3112 on M lac. Cell spread after P1, each half plates separately.

12/15/56. no colonies yet

12/16/56. 37 colonies with P1, none on other half plate.
~~most colonies look restored lac +,~~. all V_6^S

12/17/. T6 test ~~much clearer with B lac than with M lac and S lac.~~

order V_6^r Y87 Y53 | other lac | lac 4 |

Try 2945 H for 4

DATE: 12/13/56 REF:

Hfr	1 W3269	2 3270	3 3271	F-X+ 4 3272	5 2244	6	V_6^T	49-89	3133	Y90
W3229	0	0	0	0	/		3237		++	++
W3221	(+	+ reversion?	+	+)	++		3240		0	++
① 1941	0	0	0	0	+		3266		+	++
1945	0	0	0	0	+		3267 V_6^S		++	++
1949 10	0	0	0	0	+		39-1		++	++
3146	0	0	0	0	++		39-2		++	++
1946	0	0	0	0	++		39-3		+	+
② 3164	3	/	4	5	/		3268		++	+
39-1	++	++	++	++	++					
39-2 20	0	0	0	+	0					
39-3	0	0	0	+	+					
1940	++	++	++	++	++	?				
③ 1942	0	0	0	+	0					
1943	0	0	0	+	0					
1944 30	0	0	0	+	0					
1947	0	0	0	+ reversion?	0					
3237	++	++	++	++	++					
④ 3240	0	0	0	0	0					
3266	0	0	0	± (4)	0					
3268 40	++	++	++	++	++					

	W3269	W3270	W3271	W3272	W3229 V_6	W3268 V_6 W3269	W3269 V_6	W3270 V_6	W3271 V_6	W3272 V_6
⑤ 3112	0	0	0	++	++	++	0	0	0	++
2244	0	0	0	+	+	++	0	0	0	+
3238	?	?	?	?	?	++	0	0	0	0
3239	0	0	0	0	0	++	0	0	0	0
39-1 F X+	++	++	++	++	++	++	++	++	++	++
Y10	++	++	++	++	++	++	0	0	0	++
7 [illegible] 0	0	0	0	++	++	++	0	0	0	++
3149	0	0	0	++	++	++	0	0	0	++
⑥ 3215	0	0	0	+	+	++	0	0	0	+
315-1	0	0	0	+	+	++	0	0	0	+
315-4	0	0	0	++	++	++	0	0	0	++
3237 F X+	0	0	0	++	++	++	0	0	0	++

N49-7, N49-8

N50

DATE: REF: Y10J (EML.)

	1	2 M lac 3	4	5	6	7	8	9	10
	3229								
W3133	O								
Y10J	O								
W3112	+								
Y10	++								

Y10J S streaked against lac, H+ for on M lac.

	Y10J	allelic F-	W2244		Y10J	allelic F	W2244			
W3229	O	O	W+	W1949	O	O	++			
W3120	O	4	++	W3146	O	O	++		?	
W3221	O	O	++	W1946	O	O	++		o	
W1941	O	O	++	W1940	O	O	++			
W1945	O	O	++	W1942	O	O	O			

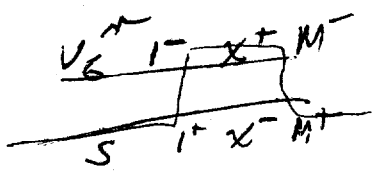
Spot test of lysate transduction and purity.

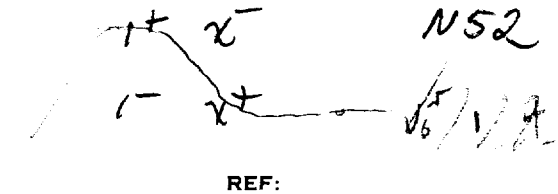
DATE: 12/16/56 REF:

Several chloroform "burns" cells should be applied after lysate has dried.
good lysis of W3112 on Blac.

lysate	B lac	M lac + W3112	Transduction
T6(Y10)	O	lysis	O
T6(Y10)		lysis	O
T6(Y10)		lysis	O
P1(W1162)			1
W1163			O
W3236V6			1
39-1V6		lysis.	++
39-2V6			O
39-3V6			O
2245V6		lysis.	O
W3224V6			4
3143V6			O
W3221V6			O
W3159V6			O
W1941V6			1
(Y) W1945V6			O
W1944V6			O
3146V6			O
1946V6			4
3269V6			O
3270V6			O
3271V6			1
3272V6			O
1945V6			O
(x) 3146V6			O
1943V6			6
1942V6			O

	Transduction
W1940V6	5
W3164V6	1
W3140V6	O
W1485	6
P1(1366)	1
P1 lac	O
W3268	10
W3240	1
W3237	O
W1947	O
1944	3
1895	O





N52

DATE: 12/18/56 REF:

	1	2	3	4	5 1942	6	7	8	9	10
				w3229 V6	[redacted]					
1	M loc		Y 105	0	0					
			w3133	0	++					
			3 [redacted]	++	0					
	0.1 ml. / 10 ml. H_2O. added 1 drop F^-x^+, Hfr to each plate M loc.									
10	Hfr	F^+x^+		5	r					
2	w3229 V_6^r	w3215	30	5 [redacted]	0	23	w3146	w2244	50	15
3		w3151	14	6	0	24		w3147	200	27
4		w3154	100	57	0	25	Pl (w3229 V_6)	w3147	5 (w3147), 4 (Pl)	
5		w3112	100	72	0	26		w2244	0 (w2244), 80 (Pl)	82
6 20		w3238	200	[40	8]?	27		w3089	2 (w3089), 24 (Pl)	18 0
	1946 V_6^r			[redacted]		[redacted]	[redacted]			
7		w3134	200	52	11	✓				
8		3089	150	40	9	✓				
9		3148	150	57	4	✓				
10 30		3152	70	50	8	✓				
11		3156	150	42	10	✓				
12		3146F-	200	-1	7	✓				
13		w3153	27	30 (light colonies, small)	6					
14		w2244	6	6	4					
15 40		w3147	200	38 [redacted]	17					
	w3146									
16		w3134	200	[redacted]	[redacted]	✓				
17		w3089	70	[redacted]	7	✓				
18		w3148	100	[redacted]	3	✓				
19 50		3152	20	[redacted]	[redacted]	✓				
20		3156	70	[redacted]	[redacted]	✓				
21		3146F-	0							
22		3153	30							

Second report on lac- study

N. Morton
Jan. 10, 1957

New stocks. Five new lac- stocks have been prepared by UV-induced mutation in W3236 and called W3267,8; N39-1,2,3. V_6^r stocks were derived from each of the Hfr lac- stocks by selection in broth. In future, a V_6^r stock will be used as source of new lac- mutants to assure identity of the V_6^r marker.

Lac- stable derivatives of the two allelic mutable stocks W3120 and W1950 were obtained after UV treatment (N36). Of 10 non-papillate colonies of each type tested, 2 of each stock failed to recombine with other lac-1 mutants. These colonies were labeled W3269-72.

Mapping of V6, lac, P. A cross was made between W1366 V_6^r lac_1^{w112} V_1^r $(TLB_1)^-$ F- and W3236 Hfr-1 P- M- on D-O+proline. The scoring of 218 recombinants gives as the most likely order

(Hfr) — V6 — 35 — lac — 15 — P — 22 — VI — 31 — (TLB) — 38

In several other experiments there was some selection for P+, but the same order and approximately the same distances are indicated.

ONPG tests

Single colony isolates from B-O were grown overnight on a rotator in YZ broth + 0.5% lactose +0.5% glycerol. These cultures were spun down, the pellet resuspended in 1 ml. water, and the cells autolysed by shaking with 1-2 drops of benzene. To 0.1 ml. of this mixture were added 2 ml. of an ONPG solution (30mg./400ml.) and the color read by eye after 10 minutes at 37° C. All of the single-step lac-1 mutants were ONPG+. Of the two-step lac-1 mutants, W3159 and Y70 were also +, but W3229 and W3269-72 were ONPG-. All of the remaining lac mutants were ONPG- except W3267, which contained reversions, W3268, which is a weak fermenter of several sugars, and W3239, a lac- stock received from Borek. Lac-3, lac-5, and N39-1,2,3 were not tested.

Aliquots of all stocks were tested for lac+ reversions before autolysis; only W1950 and W3267 contained a detectible number of reversions.

Fermentation tests

W3268, N39-1, and lac-3 ferment galactose, maltose, glucose, and arabinose weakly or not at all. N39-3 does not ferment maltose, but does ferment the other sugars. Lac-5 does not ferment galactose or maltose but does ferment glucose and arabinose. The remaining lac- mutants will ferment all the other sugars tested.

Crossover suppression in two-step mutants

Of the 2 ONPG + two-step lac-1 mutants, Y70 shows the same recombination pattern as Y53, and W3159 covers all of the recognized lac-1 region except W1946. W3229 and W3272 cover all of the lac-1 region and presumably extend beyond it, since they are ONPG-. However, they have not been shown to be allelic with any ONPG- single-step mutant. W3269-71 do not recombine with lac-2 and lac-4 or with the majority of lac- mutants of unknown location. Either the region of crossover suppression is large, or most of the lac- mutants are in the neighborhood of lac-1.

Transduction tests

If an M lac plate is spread with two drops of an F- lac- prototroph and half of the plate respread with 1 drop of a P1 lysate obtained from a non-allelic lac- Hfr V_6^r by the Lennox modification of the Adams layer plate technique, then a yield of from 20 to 300 colonies will be obtained with P1 at a time when the control half of the plate is blank or has at most a few colonies. Allelic lysates give zero yields. In all combinations so far tried, the proportion of V_6^r among lac+ colonies is less than 1%. No persistant heterogenotes have been obtained.

Recombination tests

More than 20 crosses have been made of Hfr V_6^r M- lac- x F- lac- prototroph

on M lac, the colonies being cross-streaked, without purification, against T6 on M lac. The Hfr parents have been W3229, W1946, W3146, and W3120. In every case, the proportion of V_6^r among the recombinants is less than 50%, with no evident reversal of ratios in reciprocal matings.

Discussion

So far attempts to map the lac-l region have been unsuccessful. This may be due to multiple adjacent crossovers in pairing regions, to proximity of the Hfr-l elimination region to V_6^r, or to heterogeneity in the location of V_6^r mutants. It is not clear what the most expeditious way to investigate these possibilities will be. Tentatively it is proposed to use W3133 as a universal receptor for all P1 lysates and as a universal F- for all Hfr-l stocks, excluding lac-l lysates and stocks. This procedure is suggested by the fact that W3229 V_6^r gave no V_6^r recombinants with W3215, W3151, W3154, and W3112, indicating perhaps that there is a V6 locus closely adjacent to the crossover-suppression region of W3229 and W3133. If at least one cross gives an excess of V_6^r, this will give some hope that the region can be mapped with existing stocks. If there is no such cross, the most hopeful possibility would seem to be that the Hfr-l locus is responsible for the mapping difficulty, and either transduction with F- donors and receptors, another fertility system, or the use of P as an unselected marker may be tried. To introduce P or another Hfr into existing P+ stocks would be difficult.

DATE: 11/10/58 REF:

1. 2 drops W3133 on M loc3 [illegible] half plate with P1 lysate, other half with 1 drop broth. Score loc⁺ recombinants on M loc against T6.

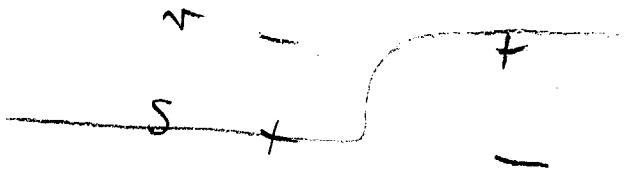
2. 0.1 ml W3133 in 10 ml. H_2O. 0.1 ml Hfr V6 in 10 ml. H_2O. Plate 1 drop of each on M loc. Cross-streak loc⁺ recombinants against T6 on M loc.

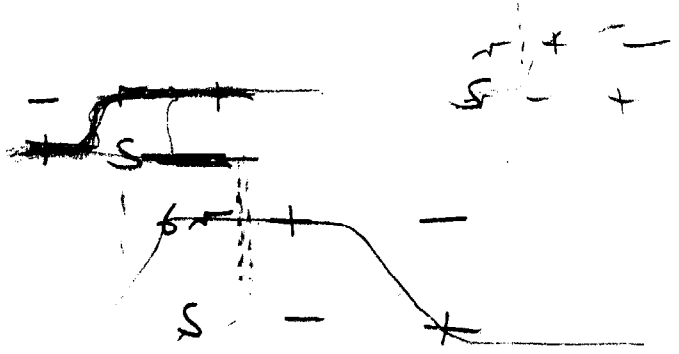
Hfr V6

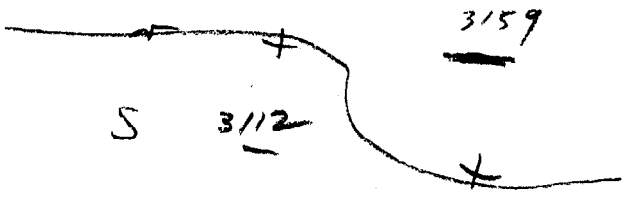
		colony count	V_6^s	V_6^r			+P1 colony count	V_6^s	V_6^r
W3164 (no dil.)	①		11	9		(21)	0		
W3140 (no dil.)	②		24	0		(21)	0		
W3245 (no dil)	③	[illegible]	all loc⁻ on streaking.			(22)	0		
W3238 (no dil)	④	−MC	all loc⁻ on streaking.			(23)	—		
W1940	⑤		12	2		(24)	200	24	0
W1942	⑥		5	5		(25)	200	23	1
W1945	⑦		14	9		(26)	200	24	0
W1944	⑧		15	1		(27)	±		
W1947	⑨		18	2		(28)	200	24	1
W3237	⑩		3	9		(29)	200	28	0
W3240 (no dil)	⑪	0	—	—		(30)	200	14	0
W3266	⑫		18	3		(31)	0	—	
W3268	⑬		3	26	✓	(32)	200	7	0
N39-1	⑭		6	15	✓	(33)	200	16	7
N39-2	⑮		19	2		(34)	0		
N39-3	⑯		6	15	✓	(35)	0		
W1946 (control)	⑰	0	—			(36)	0		
W3120 (control)	⑱	0	—			(37)	0		
W3236 (control)	⑲	not done				(38)	0		

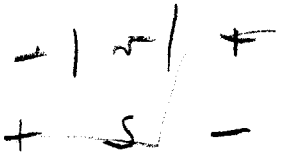
Recombination — Transduction

				Recombination	Transduction
W3159 V_6^r — ×	W3147		(39)	[illegible]	1
	W3149		(40)	1	[illegible]
	W3215		(41)	[illegible]	[illegible]
	W3151		(42)	0	0
	W3154		(43)	[illegible]	[illegible]






3159
3112



DATE: 1/10/57 REF:

	1	2	3	4	5	6	7	8	9	10
					Colony control	Colony count + PI	V_6^S	V_6^T		
	W3159 V_6^T	— x W3112		(44)	5	19	5	12	←	
		W3127		(45)						
		W3237F-X+		(46)						
		N39-1F-X+		(47)	0	0				
10		W2244		(48)	0	0				
		W2243		(49)						
		W2245		(50)						
		W3239		(51)						
		W3238		(52)						
	W1366 V_6^T	— x W3147		(53)	15	45	38	3		
		— x W3149		(54)	1	0	41	15		
		— x W3215		(55)			30	5		
		— x W3151		(56)			13	0		
		— x W3154		(57)						
30		— x W3112		(58)			49	6		
		— x W3127		(59)						
	H for V_6^T	Colony count	V_6^S	V_6^T						
	W3229 V_6^T (60) W3147	53	27	0	0					
	(61) W3149									
	(62) W3215									
40	(63) W3151									
	(64) W3154									
	(65) W3112									
	(66) W3127									
	(67) W3238									
50	(68) N39-1F-X+									
	(69) W2244									

V6ʳ and lac unselected

DATE: 1/23/57 on D-O, M-lac

	1	2	3	4	5	6	7	8	9	10
1.	W3236 V6ʳ		Δ	r	lac+	P−		predict V6ʳ excess.		
	x Y10			S	−	±		V6ʳ lac+ P⁻		
								S		
2. ✓	W3236		Δ	S	+	−				
✓	W1366 V6			r	−	±				
3.	W3229 V6		Δ	r	−	+	M−			
✓	x Y10			S	+	+	TL−			
4.	W3229		Δ	S	−	+				
W2035	x F− V6 TL			r	+	+				

~~P28 mating made on D-O. 0.1 ml of 1/100 dil. 2 ... 1×10^{-4}~~
~~single colonies streaked on B-O~~
~~selected ... single colonies from~~
~~B-O onto phage assay.~~

10^{-3} ml ... 1/10 ml.

✓5. On M lac W3236 x W3159 V6

✓6. " W3236 V6ʳ x W3133

7. P1 (W3236 V6) 1 drop X W3133 on M lac. 10^{-1} ml, 10^{-2} ml, 10^{-3}

8. P1 (W1366) x W3236 on D-O.

A28 No growth.

matings 1-4 on M lac. 10^{-4} ml.

P29. Good growth of 1-4. Colonies streaked on M lac.

Purified colonies on M lac from 5 to ... 1 ml phage assay.

Repeat #7.

N54A

DATE: 1/31/57 REF:

counts for	V6^{r}	M lac	3	4	V6S	6	7	8	9	10
1	—				—					
2	8				17					
3	0				33					
4	28				2					
5	3	7	5		10	23	24			
6	10	21			9	5				

P2 streak from D-O to B-O. P3. Pick from B-O onto plenary.

P4 Cross-streak against T6 on S lac

	lac^{+} r	+S	−r	−S
1	0	0	0	6
	0	0	0	23
	0	0	0	35
2	0	1	26	0
	6	0	15	0
	0	0	29	1
3	0	19	5	0
	0	20	9	0
	0	4	2	1
4	28	1	0	7

mapping lac, region

DATE: REF:

1	2	3	4	5	6	7	8	9	10

Controls: Pick single colonies from Mb into peroxcay. ~~[illegible]~~

~~[illegible] spread and drop [illegible] plate~~

next day spread, drop (0.1ml) F⁻ prototroph on M lac, Then respread 3/4 plate with H fr auxotroph (0.1ml). This gives control on reversions in F⁻ prototroph. I include W3133 control in all matings, to determine reversions in H fr auxotrophs, and Melibi control.

✓ 1. W1946 × W3159 V_6^r control 0, ca. 1000 colonies 13, 10, 14, 28, 17, 17 (S ... R)

✓ 2. W1946 V_6^r × W3159 control 0, ca. 1000 colonies 23, 26 15/12

control 3. W1946 × W3133 0

4. W 1946 V_6^r × W 3133 0

5. W1946, W1946 V_6^r × W3153 V_6^r. ca. 50 on all W3153, no recombination

Streak colonies twice on B lac, test on S lac + T6.

Test ca. 40 colonies per cross. — 1 purification — 2 purifications

A15

				1 purification	2 purifications
6	W1946 × Y70 V_6^r	200 (0 on control)		21r, 8S	51r, 20S
7	W1946 V_6 × Y70	200 (0 on control)		12r, 18S	17r, 42S
✓ 8	W3120 × W3153 V_6^r	2000 (20 col. on control)		20r, 10S	49r, 29S
✓ 9	W3120 V_6^r × W3153	2000 (20 col. on control)		10r, 20S	20r, 52S
10	test W1946	on W3133	10		
11	test W 1946 V_6	"	5		
12	test W3120	"	3		
13	test W 3120 V_6	"	12		

P18

14	W1946 × W3089 V_6^r	2000, 0	40r, 1S		
✓ 15	W1946 V_6 × W3089	2000, 0	30, 51S		
✓ 16	W 3221 × W3153 V_6^r	2000, 0	17r, 30S		
17	W3221 V_6^r × W3153	2000, 17	7r, 4S	20r, 13S	
18	W1946	} 3133	0		
19	" V_6^r		1		
20	3221		28 } very small		
21	3221 V_6^r		38 }		

1946 / 3089 = 3089

27/17

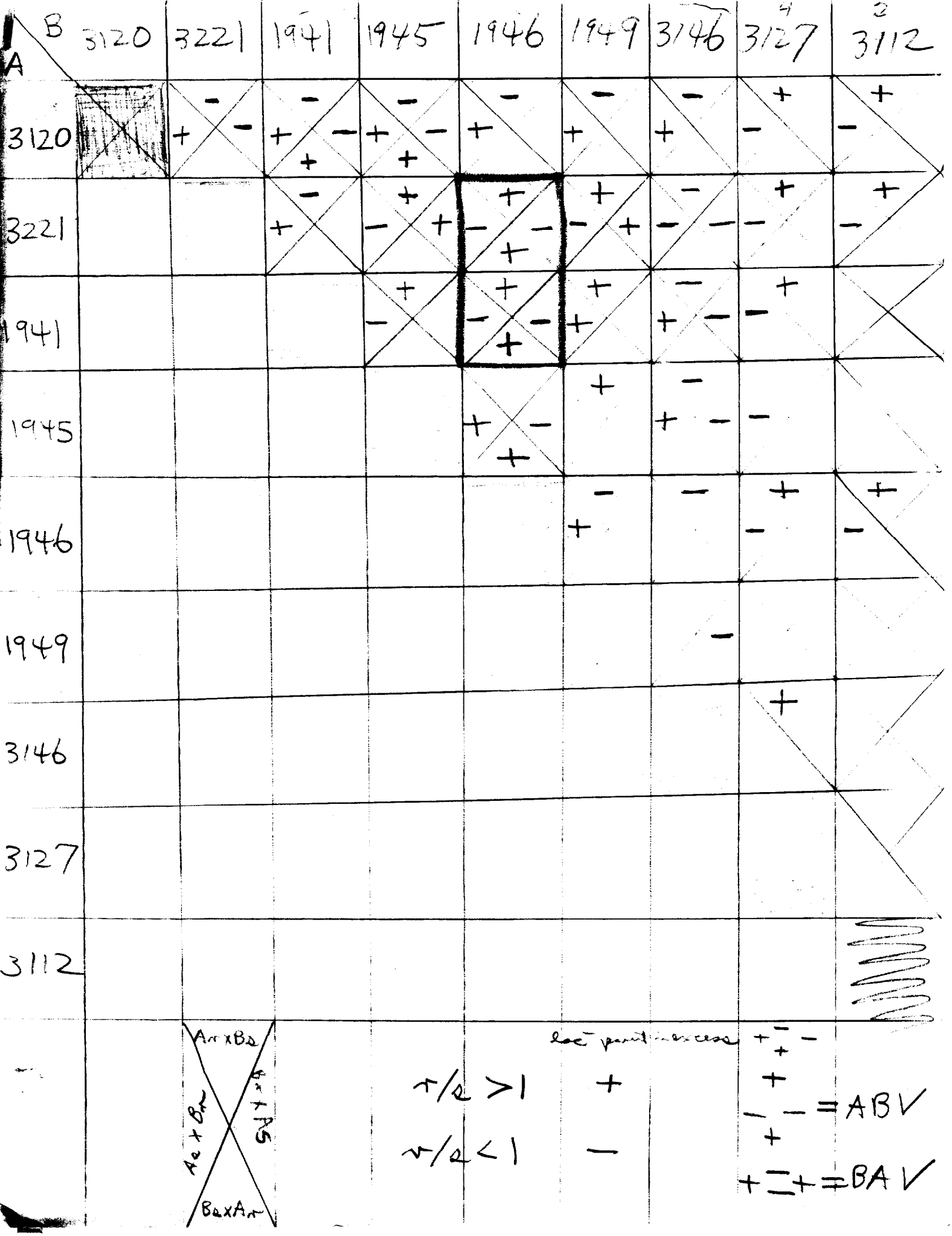
N55A

DATE: REF:

	1	2	3	4	5	6	7	8	9	10
22	w1946 X w3148 V6				1500, 2		23 r, 11 d			✓
23	w1946 V6 X w3148				9 r, 24 d				✓	
24	w1941 X w3153 V6									
25	w1941 V6 X w3153									
26	w1946 X w3152 V6				36 r, 12 d				✓	
27	w1946 V6 X w3152				10 r, 40 d				✓	
28	w1945 X w3153 V6									
29	w1945 V6 X w3153									
30	w1946 X w3156 V6				26 r, 13 d				✓	
31	w1946 V6 X w3156				18 r, 26 d				✓	
32	w1949 X w3153 V6									
33	w1949 V6 X w3153									
34	w1946 X w3175									
35	w1946 V6 X w3146 F V6 S									
~~36~~	~~w1946 X w3153 V6~~									
36	w1946 X w3127 V6				17 r, 31 d				✓	
37	w1946 V6 X w3127				29, 19 d				✓	
38	w1946 X w3112 V6				14 r, 36 d				✓	
39	w1946 V6 X w3112				33 r, 17 d				✓	
40	w1946, V6 X w3133									

X maleless Y ~~XHf r V6~~ A B C D Order

( ) ( ) V6


A
B
3120
3221
1941
1945
1946
1949
3146
3127
3112
3120
3221
1941
1945
1946
1949
3146
3127
3112
$A_r \times B_a$
$A_a \times B_r$
$A_r \times B_r$
$B_a \times A_r$
last point increase
$r/a > 1$ +
$r/a < 1$ −
+ − − = ABV
− − + = BAV

DATE: 3/12/57 .05ml each parent on M lac REF:

Hfr r	F⁻	2	F⁻ control	F⁻ Hfr r recom.	r (5)	s (6)	7	8	9	10
W3221	3112V6	1	0	2000	24	19	repeat	✓		
W3221V6	3112	2	0	2000	31	16		✓		
W3221	3127V6	3	4	2000	15	30		✓		
W3221V6	3127	4	16	2000	25	11		✓		
W3221	3156V6	5	3	1500	21	29	} repeat			
W3221V6	3156	6	5	1500	22	28	}			
—	3152V6	7	4	2000	20	31	}			
— V6	3152	8	2	1500	26	22	}			
—	3148V6	9	0	2000	25	23				
— V6	3148	10	0	1500	47	0				
—	3153V6	11	8	2000	13	36	✓			
— V6	3153	12	17	2000	30	17	✓			
—	3133	} 13	0,0 ——							
V6	3133	}								

3/23. allelism test of lac⁻ stocks.

	F⁻V6^r		F⁻V6^s		w3133	
	Hfr V6^r	Hfr V6^s	Hfr r	Hfr S		
3120	15	15	15	15	15	a few lac+ from w3134.
3221	50	50	50	50	50	many small revertants from w3089
1941	10	10	10	10	10	
1945	10	10	10	10	10	
~~1946~~						
1949	20	20	20	20	3	many (ca 30) lac+ revertants = F⁻
3146	1000 (small)	——	1000 (small)	——	1000 (small)	**reinoculate 3146.**
1940	0	0	0	0	2000	

DATE: 3/26/57 REF:

	1	2	3	control	5	6	7	8	9	10
W3120	V6	W3089	1	0	1500					
		" V6	2	0	1500	49	2			
	V6	3148	3	12	1000	25	26	✓		
		" V6	4	0	1000	30	19			
	V6	3152	5	2	1500	22	28	✓		
		" V6	6	5	1500	32	18			
10	V6	3153	7	14	2000	17	33	✓		
		" V6	8	12	1500	30	19			
	V6	3156	9	9	2000	20	30	✓		
		" V6	10	7	1500	26	24			
	V6	3146 V6 S	11	0	2000	18	33	✓		
		3175	12	0	2000	36	14			
20	V6	3127	13	15	2000	34	14	✓		
		" V6	14	3	1500	24	26			
	V6	3112	15	0	1500	39	10	✓		
		" V6	16	0	2000	12	38			
	—, V6	3133	17	3,5	—					
	V6	3147	18	0	2000	9	41			
		# V6	19	0	2000					
30									(24)	
		—	V6 ✓	Tests	—	on Blac.				I6
20			V6 ✓	22		V6 ✓			W3134	S
	3120	—	—		S	—	new pref 4/11/57	O.K. do over	W3134 V6 ✓	S (P)
	3221	S	✓	F-	S	✓				
	1941	S	✓		S	✓		do over	W3147 V6 ✓	S
	1945	S	✓		S	✓			W3270F-# V6 ✓	R
40	1946				S	✓		do over	W3269F-# V6 ✓	S (P)
	1949	S	✓		S	✓				
21	3146	—	✓	23	S	✓				
	3127	—			S	✓				
50	3112	—			S	✓				
	1940	S	✓		S	(S)	N.B.			

DATE: REF:

	1	2	3	4 control	5 recording	6	7	8	9	10
W3221	V6	3148	20	0	700	22	27 ✓			
	" V6		21 ✓	2	2000	29	20			
	V6	3152	22 ✓	3	1000	30	19	✓		
	" V6		23 ✓	3	2000	19	31			
	V6	3153	24 ✓	1	2000	29	21 ✓			
	" V6		25 ✓	6	2000	16	32			
	V6	3156	26 ✓	2	1000	30	20	✓		
10	" V6		27 ✓	0	300	19	30			
	V6	3146 V6	28 ✓	0	2000	22	28	?		
		3175	29 ✓	0	200	12	35			
	V6	3127	30 ✓	12	2000	36	14			
	" V6		31 ✓	3	2000	15	33	✓		
	V6	3112	32 ✓	0	2000	25	21]	?		
	" V6		33 ✓	0	2000	22	23]	?		
20	V6 }	3133	34	0,0						
	V6	3147	35	0	2000	23	25			
	V6	3134	36	10	2000	17	28			
	4/2/57 into DO.				↑	↓				
30				control						
W1941	V6	3089	2	2000, 0	18	22	(not complete lyrics)			Repeat
	" V6		3	0, 1000	14	14	" "			
	V6	3152	4	1500, 3	22	17	✓			
	" V6		5	2, 1500	17	23	✓			
	V6	3153	6	2000, 10	26	14				
	" V6		7	2000, 6	15	24				
40	V6	3146 V6	8	2000, 0	15	21	(not complete lyrics)			
	3125		9	2000, 3	28	21				
	V6	3127	10	2000, 12	23	10				
	" V6		11	2000, 0	15	31				
	V6	3112	12							
	" V6		13							
50	V6	3133	14	0, 0						
	V6	3147	15							
	V6	3134	16	2000, 1	14	26				✓
	" V6		17	2000, 20	31	19				

DATE: REF:

	1	2	3	4	5 (r)	6 (D)	7	8	9	10
1941	V6	3156 11V6	18 19	2000,3 2000,0	23, 13,7	17 27,13	✓ ✓			
	Tried streaking from M lac plate (after transfer from original M lac recombination plate) — very difficult to read. no good.									
						H	D			
1945	V6	3230 3230V6	1 2			15 40	36 10			
	V6	3089 V6	3 4	——— repeat		28	19	?		
	V6	3148 V6	5 6							
	V6	3152 V6	7 8	——— repeat						
	V6	3153 V6	9 10			34	24		✓	
	V6	3156 V6	11 12			24	20	?.	✓	
	V6	3146V6 S F X+ 3175	13 14			23 30	27 17	?	✓ ✓	
	V6	3127 V6	15 16			22	28			
	V6	3147 V6	17 18			20 31	26 18			
W3221	V6	3230 — V6	19 20							
1941	V6	3230 — V6	21 22							
	1945 V6 on 3133		23							

map lac_1, lac_2, V6.

DATE: 4/29/57 REF:

	1	2	3	4	5	6	7	8	9	10
	Streak W3164, W1366, W3164 $V6^T$, and W112 on B-O for single colony isolation.									
5/1	Single colonies to parasay									
5/2	1 ml W3164 + 1 ml W1366 into D-D								①	
	1 ml W3164 $V6^T$ + 1 ml W112 into D-O								②	
10	Spread on D-O. Pick single colony & streak on B-O. Purified colony into 1 ml. D-O. Spot on M lac.						contact ~~test~~ on B-O against T6.			
		—	W3229	W1940	W1946		V6			
	~~lac2~~	O	+	+	+					
	lac1	O	O	+	+					
20	~~lac1,2~~	O	O	+	O?					
	~~lac+~~	+	+	+	+					
	Two pair cells	O	O	O	O					
②	A 1	—	O	O	+		s			
	2	—	O	O	+		s			
	3	—	O	O	+		s			
	4	—	O	O	+		s			
	5	—	O	O	+		s			
	B 1	—	O	O	+		s			
	2	—	O	O	+		s			
	3	—	O	O	+		s			
	4	—	O	O	+		s			
	5	—	O	O	+		s			
	6	—	O	O	+		s			
	C 1	—	O	O	+		s			
	2	—	O	O	+		s			
	3	—	O	O	+		s			
40	4	—	O	O	+		s			
	5	—	O	O	+		s			
	6	+	+	+	+		s			
	D 1		O	+	+		s			
	2		O	+	+		s			
	3		O	+	+		s			
	4		O	+	+		s			
	5		O	+	+		s			
	E 1		O	+	+		s			
50	2		O	+	+		s			
	3		O	+	+		s			
	4		O	+	+		s			
	5		O	+	+		s			
	F 1		O	+	+		s			
	2		O	O	+		s			

DATE: REF:

	1	2	w3229	w1940	w1946	V6	7	8	9	10
F 3		0	0	+	+	s				
4		+	+	+	+	?				
5		0	0	+	+	s				

① G 1–5, F 2–5, H 2–5, J 2–7, K 2–7, L 2–7, M 2–7 [handwritten +/0 entries in columns 2, w3229, w1940, w1946 and V6, not transcribed]

Too few cells for allelism test

Other colonies tested only for lac; —

	+	—
①	9	36
②	12	42

Preparation of lysate titer T6 on bacto-tryptone (TSB)

DATE: 5/31/57 REF:

24 hr. broth strain B — Bacto-Tryptone ✓

10:45 .05 ml B to 25 ml. broth. Rotate. ✓

12:45 add .05 ml. T6. Rotate & inspect every hr. ✓

4:45 clearing. add CHCl3

TSB 10 gm Bacto-tryptone
5 gm NaCl
1000 ml H_2O

6/11 Dil. 100 X 100 X 100 X 100

add .1 ml. to .05 ml. TSB, plate .2 ml. = 10 7 dil.

on B-0 pre-spread with .1 ml W3230. ca. 700 colonies = 7 X 10 9 ✓

W3230 dil 100 X 100 X 100

(1) .1 ml into .1 ml. TSB. plate on B-0 } incubate 20 min. at 37°.

(2) .1 ml into .1 ml. T6 plate on B-0. }

at .50 ml. TSB and plate on B-0.

6/12 (1) clear ✓

(2) ca. 500 colonies

Selection of lac-stable colonies from W1951V6-

DATE: 6/27/57 REF:

24 hrs broths of 5 colonies, picked from B-O.
Spread 1 drops/plate on B-lac. 10 sec. UV ✓

7/1/57 lac-st picked & streaked on B lac.

7-3/57 streak on B gal

7-5 pick into TSB

M lac

3

	3133	Y10	3089	3148	3156	3146F-Y+	3153	3127
① 60-4	O	+	+	+	++	++	++	++
60-3A	O	+	+	+	++	++	++	++
60-3B	O	+	+	+	++	++	++	++
60-3C	O	+	+	+	++	++	++	++
60-1A	O	+	+	+	++	++	++	++
60-1B	O	+	+	+	++	++	++	++
② 60-1C	O	++	+	++	④ ++	++	++	++
(63A)	O	++	++	++	++	O	+	++
63B	++	++	++	++	++	++	++	++
63C	++	++	++	++	++	++	++	++
63D	++	++	++	++	++	++	++	++

allelism tests of w3269-72, w3229

DATE: ✓ ✓ ✓   REF:

(1)

	w3229	w3269	w3270	w3271	w3272	
w3133	0	0	0	0	0	
w3127	+	0	0	0	+	papillae on streak
w3112	++	0	0	0	++	papillae on streak
w2243	↔ (arrow across row, +)					+ on streak
w2245	↔ (arrow across row)					+ on streak

(2)

	w3229	w3269	w3270	w3271	w3272	
Y10V6r	++	++	++	++	++	
w3238	+	0	0	0	++	many papillae on streak
39-1F-X+	↔ (arrow across row)					a great many papillae on streak
3147	++	0	0	0	++	
3149	++	0	0	0	++	

(3)

	w3229	w3269	w3270	w3271	w3272
3215	+	0	0	0	+
3151	+	0	0	0	+
3154	++	0	0	0	++
3237F-X+	++	0	0	0	++

(4) w1282 test

	w1282	3133	Y10
w3229	0	0	↕ + on streak
w3120	±	±	
w3221	+	0	
w1941	+	0	
w1945	+	0	

	w1282	3133	Y10
w1949	+	±	↕ w streak +
w1946	+	0	
w1940	++	++	
w3146	++	0	
w3268	++	++	

w1282 same as Y70, Y53, Y87 in recombination pattern

(5)

w3267 X w3133	++
w3269F-X+	++
w3270F-X+	++
w3271F-X+	++
w3272F-X+	++
Y10	++

(6)

w3268 X w3133	++
3269F-Y+	++
3270F-X+	++
3271F-X+	++
3272F-Y+	++
Y10	++

Lac- strain from W3267

DATE: 6/29/57 REF:

	1	2	3	4	5	6	7	8	9	10
	1 drop W3267	2 hr. hrs. on			B lac-	10 sec 21V.				
7/2/57	Colonies all lac+.				Lac- (?) sectors streaked on B lac.					

10

20

30

40

50

DATE: 7/2/57 REF:

	1	2	3	4	5	6	7	8	9	10
	reversion test of lac- st									
	W3159V6T, W3133V6T, W3269-72 F-K+V6T									
	streaked on B gal.									
	# papillae / .05 ml.									
3159	50									
3133	0									
3269	0									
3270	0									
3271	0									
3272	0									

Linkage of V6, P, and W3229, W3269-72 65

DATE: 7/3/57 REF:

1. W3236 V6 X W3133 etc, on M loc. If heavy growth, no linkage with P. Dilute and plate on M loc. Pick and count single colonies against V6.

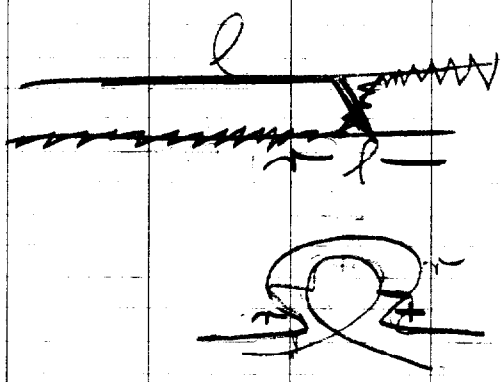
2. W3236 X W3133 V6, etc. .05 ml of each point on D loc on M loc

1-1	W3236 R X	W3159	2-1	W3236 × W3159 V6 T
1-2	"	3133	2-2	
1-3	"	3269 F-x+	2-3	
1-4	"	3270 F-x+	2-4	
1-5	"	3271 F-x+	2-5	
1-6	"	3272 F-x+	2-6	

7/5/57 dense recombination ∴ P and loc not accurately linked. Scrape ½ plate and attach on M loc.

	T	A
1-1	0	20
1-2	0	20
1-3	1	18
1-4	0	36
1-5	7+14	16+13
1-6	6	11
2-1	18	0
2-2		
2-3	2	11
2-4		
2-5	17	0
2-6		

!



linkage map, loc 1, 2, 4

D-O recombination, Stoc cross.

DATE: REF:

	1	2	3	4	5	6	7	8	9	10
7/5	W3164 X W208									
		+	−							
		8	60							
		30	92							
10		38	152							
20										
30										
40										
50										

on M lac — reversion test

DATE: +3269F-X+ REF:

	1	2	3	4	5	6	7	8	9	10
+3159				+3159						
60-4	O			60-1C	+	1951	+			
60-3A	O			63A	O					
60-3B	3			63B	+++					
60-3C	5			63C	+++					
60-1A	++			63D	O					
60-1B	++			3267	+++					
	63B					63B				
	++		w3133			O				
20	++		w3146F-X+			O				
	+++		3127			O				

30 Purify lac+ colonies on B-O. Spot on B-O. Replicate on W3133, W3159 (lac-), W3269-72F-X+ on M lac.

60-4	gal-	60-1A	63B
60-3A		60-1B	63C
40 60-3B		60-1C	63D
60-3C	glu- gal-	63A	

50

W1B lac V6 V1 T6B1 X W3 M P

Summary of N37

assuming

selection for Prol+

Type	V6	lac	Prol	V1	5	Exper A	B	8	assuming Prol V6 lac V1	10
ace	s	+	+	r		8	8		bde	
acd	s	+	+	s		5	10		b	
a	s	+	−	s		46	21		a	
ade	s	+	−	r		2	1		ade	
abe	s	−	+	r		3	7		bce	
abd	s	−	+	s		6	4		bcd	
abc	s	−	−	s		5	1		acd	
abcde	s	−	−	r		1	0		ace	
bce	r	+	+	r		6	8		cde	
bcd	r	+	+	s		2	9		c	
b	r	+	−	s		9	4		abc	
bde	r	+	−	r		1	0		abcde	
e	r	−	+	r		57	71		e	
d	r	−	+	s		47	75		d	
c	r	−	−	s		16	4		abd	
cde	r	−	−	r		4	1		abe	
						218	224			

Exper I.

8 a	4 ab (15)	4 bd (10)	V6-S	
8 b	4 ac (19)	4 be (12)	V6-R	
8 c	4 ad (14)	4 cd (12)	lac+	
8 d	4 ae (14)	4 ce (19)	lac−	
8 e	4 bc (14)	4 de (8)	Prol+	134 (.61) / 192 .86
			Prol−	84 / 32
			V_1-S	
			V_1-R	

Exper A

(46) $a = \frac{76}{218} = .349$

(9.5) $b = \frac{33}{218} = .151$

(16) $c = \frac{47}{218} = .216$

(47) $d = \frac{68}{218} = .312$

(57) $e = \frac{82}{218} = .376$

175

Summary of experiments 21,32,037

DATE: REF:

1	2	3	4	5	6	7	8	9	10

Product formula

V_6 Δ_{sec} P_{mk} V_1 (TL)

(.349) .165 .209 .220 (.394) $\Sigma = 1.337$

10

(.349)

(.362)

(.385)

(.608)

.165

20

.245

$E = .374 - .070 = .304$

.360 $.594 - .176 = .418$

(.651)

.209

30

.369 $.429 - .092 = .337$

(.638)

.220

(.615)

40

.394

50

91

133

16

12

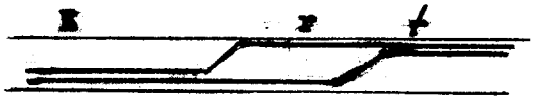
MAPPING THE LAC LOCI WITH W6 AS THE UNSELECTED MARKER

Prior information

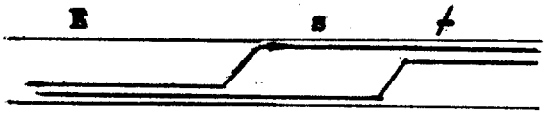
(J.L.) E gal Lp lac7 lac5 lac3 lac2 lac4 Y87 W112
? W6

(W1366 x W3236) E W6 lacI P
35 15 22

I. lac + x lac−; W3236 Hfr W6($^{r}_{s}$) lac+ x F− W6($^{s}_{r}$) lac− on M lac or D lac.

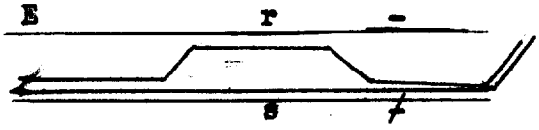
r+ x s− 

r/s ~35/15

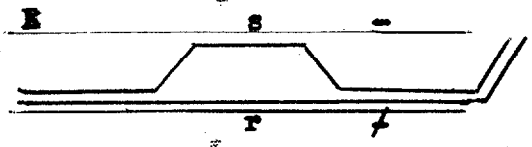
s+ x r− 

r/s ~ 15/35

II. lac − x lac +; Hfr W6($^{r}_{s}$) lac− x Y10 F− W6($^{s}_{r}$) TLR^{r} on M lac or D lac.

r− x s+ 

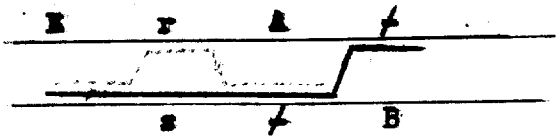
r/s ~ $\frac{(15 \times 35)}{85 \times 65}$ = 5/55

s− x r+ 

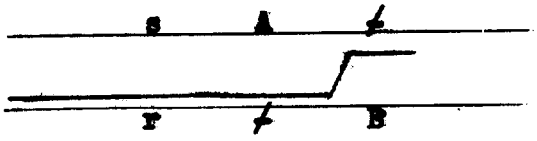
r/s ~ 55/5

III. lac A x lac B; Hfr W6($^{r}_{s}$) lacA x F− W6($^{s}_{r}$) lacB

Case 1. — E W6 A B

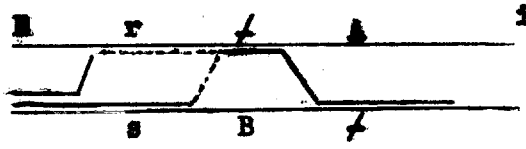
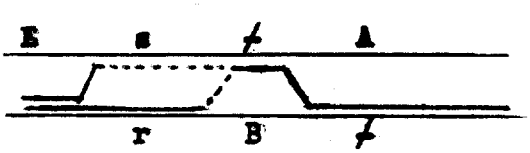
rA x sB 

r/s < 1

sA x rB 

r/s > 1

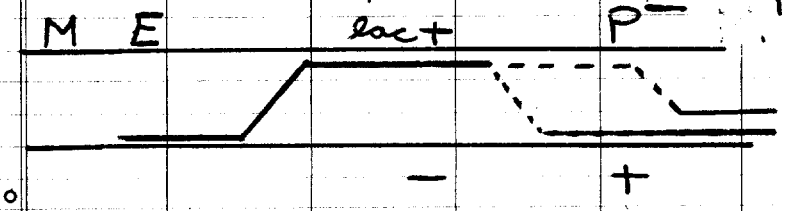
Case 2. — E W6 B A

		negative interference	positive interference
rB x sA		r/s < 1	r/s > 1
sB x rA		r/s > 1	r/s < 1

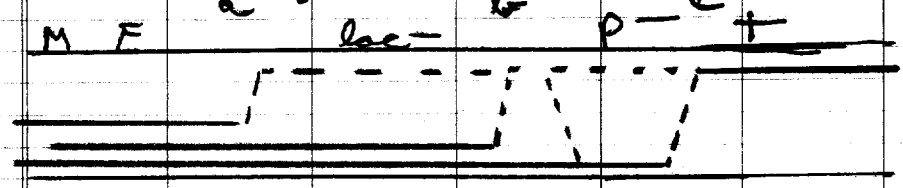
mapping the lac loci with P^- as the unselected marker

DATE: REF:

I. lac+ x lac−; W3236 Hfr V6($\frac{+}{2}$) lac+ P^- x F− V6($\frac{2}{+}$) lac−
on D lac + proline. Score for P+, −.



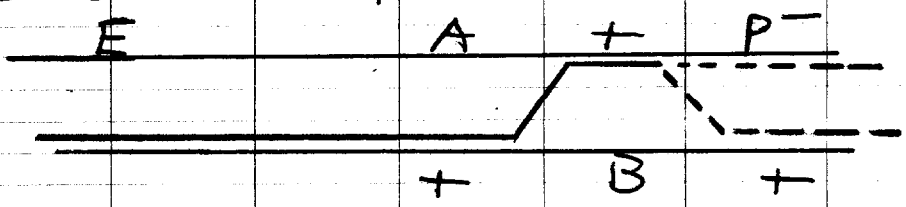
II. lac^-_a x lac^+_c; Hfr V6($\frac{+}{2}$) lac^-_c P^- x Y10 F− V6($\frac{2}{+}$) TLB$_1$−
on D − O + proline.
Score for P+, −; lac +/−.



lac+ P− b
lac− P+ abc
lac− P− a
lac+ P+ c

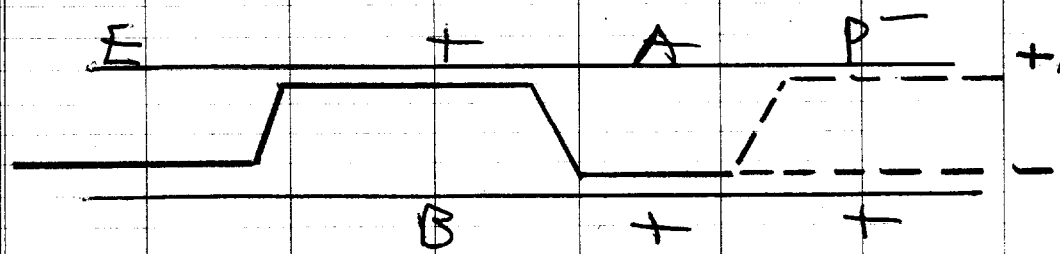
III. lac A x lac B: Hfr V6($\frac{+}{2}$) lac A P^- x F− V6($\frac{2}{+}$) lac B
on D lac+ proline

Case I. E A B P



neg. interference pos. interference
+/− >1 +/− <1

Case II. E B A P



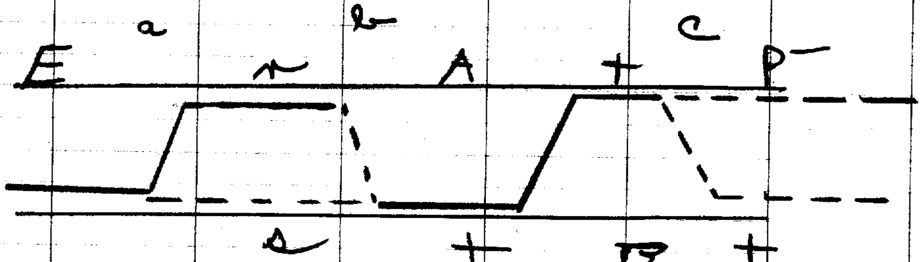
+/− <1 +/− >1

mapping the lac loci with V6 and P as unselected markers

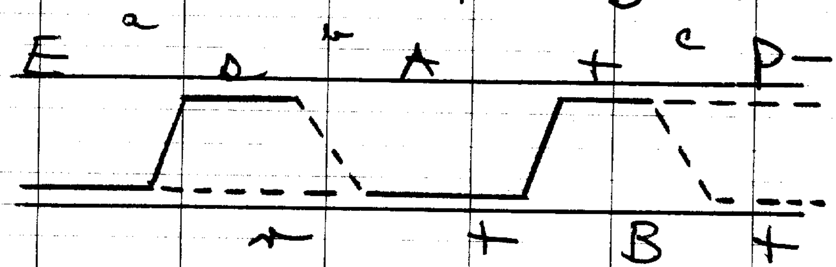
III. ~~lac~~ A x lac B ; Hfr V6(r/s) lac A P⁻ x F⁻ V6(s/r) lac B P⁺ on D lac⁺ + proline.

Case I. E V6 A B P

r A x s B



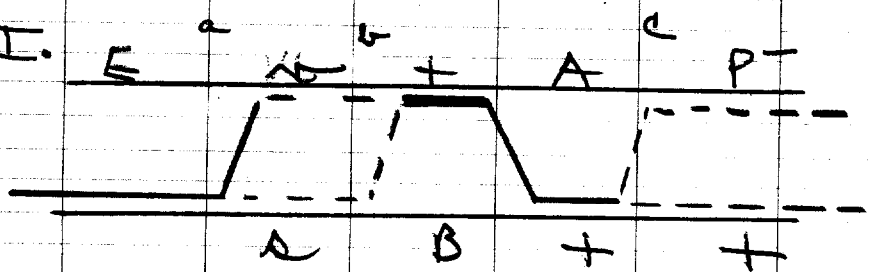
s A x r B



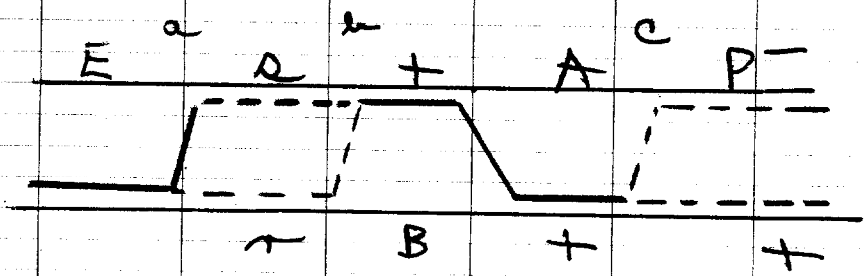
V6	P	
r	−	ab
s	+	c
r	+	abc
s	−	—
r	−	—
s	+	abc
r	+	c
s	−	ab

Case II. E V6 B A P

r A x s B



s A x r B



V6	P	
r	−	ac
s	+	b
r	+	a
s	−	c
r	−	c
s	+	a
r	+	b
s	−	ac

Mapping Lac-1 Region

r/s

DATE: REF:

A	B	2 Ar x Bs 3	4 As x Br 5	6 Br x As 7	8 Bs x Ar 9	Order
W3120	W3221	17/28 ?	49/2 ?	7/33 ? 17/28		
	W1941	11/32 ? 18/31 ? 35/26	30/19	W3134 14/26	W3134 31/19	?
	W1945	11/35 18/36 32/28	32/18	15/36	40/10	?
10	W1946	30/72 23/50 13/42 17/33	69/39 30/19			
	W1949	17/32 ✓ 18/37 20/30	26/24			
	W3146	21/75 15/45 18/33	36/14			
20	W3127	34/14	24/26	—	—	ABV
	W3112	39/10	12/38			
W3221	W1941	22/27	29/20			
	W1945	30/19	19/31	28/19		?
30	W1946	27/17 30/17 29/21	12/30 13/36 ✓ 16/32	30/51 7/40	40/1	ABV
	W1949	30/20	19/30	24/20		
	W3146	22/28 ?	12/35	7/53		
40	W3127	25/11 36/14	15/30 15/33	—	—	ABV
	W3112	31/16 25/21	20/22 22/23			
W1941	W1945	22/17	17/23			
50	W1946	26/14	15/24	9/24	23/11	ABV
	W1949	23/17	40/20			

Mapping Lac-1 Region

r/s

DATE: REF:

A	B	2 Ar x Bs 3	4 As x Br 5	6 Br x As 7	8 Bs x Ar 9	Order
W1941(cont.)	W3146	15/21	28/21	3/56 ?		
	W3127	23/10	15/31	—	—	ABV
	W3112					
10 W1945	W1946		34/24	10/40 ✓ 8/50	36/12	
	W1949	24/20				
	W3146	33/27	30/17	19/35 ?		
20	W3127		22/28			
	W3112					
W1946	W1949	18/26 ✓ 10/42	26/13			
	W3146	9/41				
30						
	W3127	29/19 ✓	17/31 ✓	—	—	ABV
	W3112	33/17 ✓	14/36 ✓			
	W3159	27/53 ✓	62/37 ✓	—	—	BAV
40 W1949	W3146			10/47 ?		
	W3127					
	W3112					
W3146 50	W3127	27/15 ✓				
	W3112					