DATE: 12/15. REF: 1101

	1	2	3	4	5	6	7	8	9	10
	Fresh cross 1:1:10 12:30 - 3:30. dilute in H_2O.									
A. 3:27 -	1. ++ incl 2 v. large (1 pend. zygs)	23 no paris no v. large.	21 do.	15. v dirt do.	8+ ? pairs	3+ dirt	3	1. 0	1+2+	3
7:55:	1-2 ✓	0	0	0	1?	0	0	0	0	0
3:55 -	23 (2 fairly long)	++ no pairs	++ one ↑ rather large	++ one v. large	+.. 1 large (paired?)	+.. 1 v. large	++ 1? v. l.	+T 1 pr 1 large but rather sm.	+ some chromp but no very large	+ do.
B.	∴ If v. large cells are zygotes, expect following to have more zygs: →							↑	1 large. 1 not	
4:20	2 pr. ic.	0 ✓	1 ✓	1	1	1	?	0	0	0
	7-10 dmd? esp. 10.									
C. (Cust. ED.)	×	×	×	×	×	×	0		×	×
D.	2	×	×	0	0					
4 40 -	Streak out dilution suspension also →									

	P1	P2	RIcomplex
	13	131	3

- = P1 + = P2

plot

12/16: A 1 1RI/P1, 13P1, 22P2
2 14P2
3 12 P1 4P2 dic
4 4 P1 7P2
5 2P1 1P2
6 2P2 + ? c.g.
7 2- 1+
8 0
9 - ? mgloos →
10 1- 1+

B 1 + + (> 23!)
2 4P2
3 23P2 1RI/P1 + c.g.?
4 61P2 6P1
5 37P2 1P1
6 49P2 2P1
7 6P2 + cg ?
8 3P2 : 2P1 : 1RI/P1...?
9 3P2 : 14P1
10 17P2 : 8P1

DATE: XII. 16 REF: 1102

		1	2	3	4	5	6	7	8	9	10
		~~6 minute~~ 0 + ~~5~~			V.S.!						
		fresh cows.									
		12[15] – 2:40		dil in H2O.							
2:50	A.	++1+[illegible]+	cg/2 dil	5	+	2,·	0 8	0	1.	1	0
		"	changed	"	"	"	"	"	"	"	"
3:10	B.	X		~~2 + 5~~			0.	∅	1+d	3 d	0 but change of pigments
			all 0	X	2	H d.			X		X
			add fluid		"	"					
			lost in deep hold to!		Ch 1? H 1?						
			hold to								
			4:40								

XII. 17

		Cells	Colonies	
A.	1	3	3 –	
	2	12	~~4 – 1 +~~ 12?	Observation not precise enough
	3	5	2 –	
	30		0	
	4	1	2 – 1? (e.g.)	
	5	2	1 –	
	6	2	0	
	7	1	0	} order? probably correct but suspicious
	8	1	1 +	
	9	1	1 –	
	10	0		
B	2	0 2		
	4	0 2		
	5	0 1		

DATE: XII.17.53 REF:

	1	2	3	4	5	6	7	8	9	10
	Fresh cross: 8¾ to ca 10 AM					.5 : 5 : 10				
A.	8	0	0 + d?	d?	0	0	0	0	0	0

add moderate fluid and incubate. Cell probably re-formed in 1, but these drops are too large for proper counting. Left at R.T 10³⁰–4²⁰ PM.

B

10 — moderate large thin

	0	0	0	0	———

Plating:

A	1 1...	4+ ! (1 fission)
	2 0	0
	3 0	0
	4 1	0
B	1 2 (…)	0
	2 ++	2– V. low RECOVERY!
D	1 6+ (2)	4+ 1– Incomplete recovery unfortunately.
	3 1	1+
	4 1	1+
	7 ?	0

20

Faster?

still counted in replicating dep.

yields v. low!!

30

Fresh cross 3:30 – 4:40 1:1:10.

C

5⁰⁰	4	x	x	0	0	0	0	0	0	0

(cap too small or resp too dilute ?).

40

D.

	includes (1)	0	1	0	0	1	1	6 + pair ?		
	0	0	0	0	0	0	0	1?	x	0	0

50

DATE: XII.18.53 REF: 1104

	1	2	3	4	5	6	7	8	9	10
A.	Freshcross 1:1:10 12:15 – 4.				∞					
B.	+ X	0	0 1. + 0	0	0	5, rare / unusual	0	0	0	0
C	5, +	2 + 1? (sup. drops)	0	0	2 + ?	0	0	0	0	X

Plating:

A 5

B 3 0

6 3+, 1 –, 1 z. ∴ 1 type cell = z. → R1, P1 only do not same.

7 1 –

C 1 2+ 6 –

2 0

5 0

30 3 types have been seen:

		normal site (presumably)
0	0	0
↓	↓	↓
R1 P1	R1	R1
P2	P1	P1

(→ mutant ↓ R1)

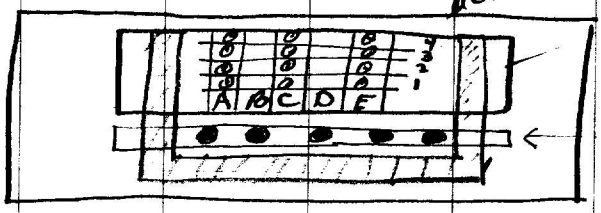
Spend next few days setting up & to determine

① Microlope (Schouten): fiddly method but not very convenient for microisolation.

② Capillary technique: needs practice in chambers. Some pul. experiments to isolate yeast (W413)

DATE: 1/6/54. REF:

Developed micromanipulation methods during past few days.
Set up oil chambers:



- Isolation capillaries
- Reservoirs

after isolation to small droplets, add extra broth and incubate overnight.

<2PM 1/6. I 7 8PM II.

A C1 ● C3 ╱ (P2) E1 ○ G3 ●⬭ detached in smearing
C2 ⬭⬭ large C4 ⬭ E2 ○ v. large
↑ somewhere

B. ~~A~~
A1 ○ large others × stops plating
A2 ○ formerly in pair, broken apart ✓ appearance of pair not accurate
A4 ○ ✓ stops ?
C2 ⬭ headdirt? ×
D2 ● ? ×
D1 ⬭⬭ stops
E2 ~~✱~~ ⬭ no change to 8PM ×
E3 ⬭ ·
E#4 ⬭⬭ tangled rope stops

One other pair not succ. isolated ? ~~0~~

~~Plate P7.~~

~~C ✱ ✱ ✱ ✱~~ probably stops
E1 stops?
E2 ×
G3 ✓ stops?

I II

(over)

		plating	appearance
A.	E1	P1	P1
	G3	P1	P1
B	A1	P1	P1
	A2	P2	P1! ~~(had been 2 in pair).~~ Reverts other mutants.
	A4	P1	?P1
	D1	P1	P1
	E3	— ng.	
:	E4	P1 + P2 (+ R1)	(from ∞)

BA2: note: Although visl colonies were all P2 r lac, a few lac++ papillae noted in brush. Streak, replicate to EMB lac sm.

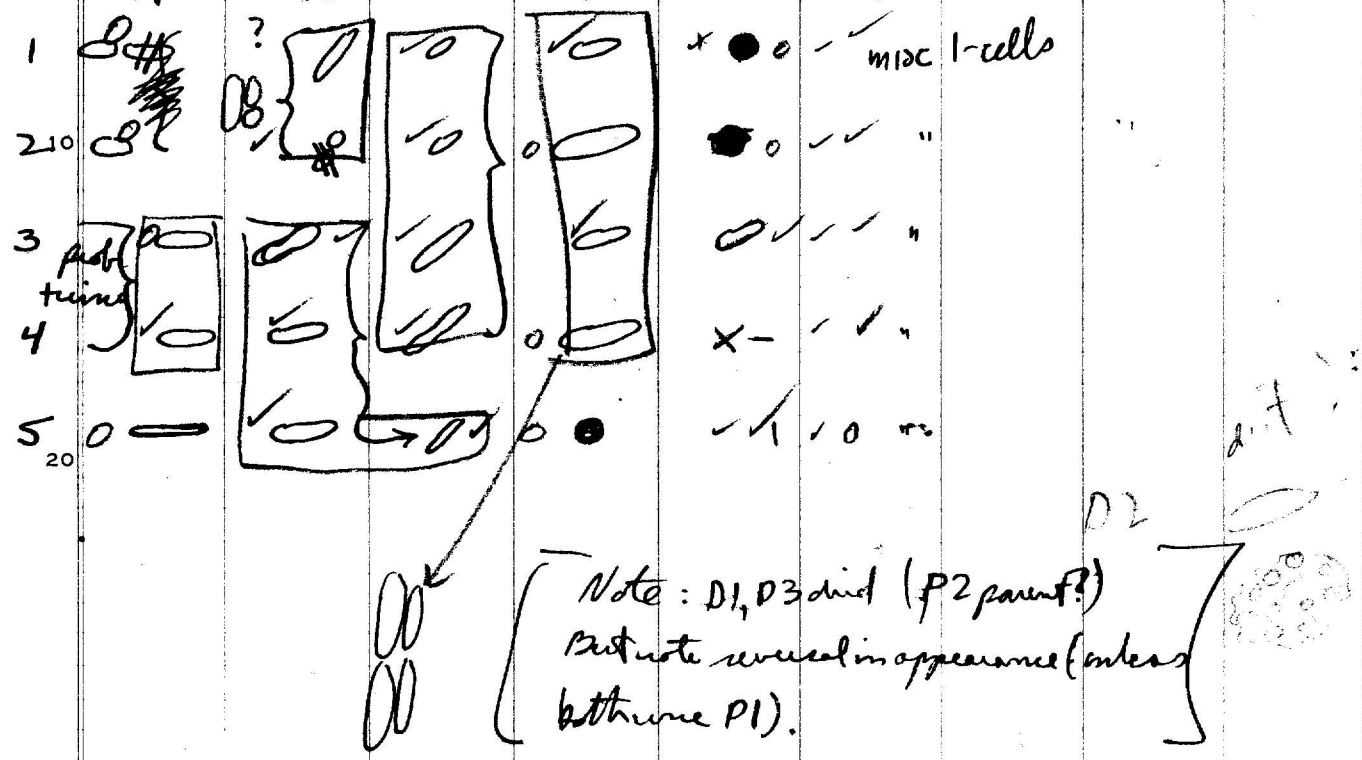
↓

A2 Original plating. One colony was lac- (previously overlooked). ✓ replica: This is also ~~lac~~ Gal+ S^R ∴ P1.

∴ This is a P1+P2 type from single cell. Originally in pair

DATE: January 8, 1954. REF:

P1 x P2. ~~overnight~~ 48hrs. motil. 1:1:10 12 30 - 2 30 PM. Dilute a 1:200 pre manipulation in Penassay. 2 30 - 4 45



B2' lysed. A1' lost. E, F mere 1-cell sol. No passis seen then.

B3-4-5-C5 from 1 clump of 4 cells.

C1-4 from 1 clump may include one tho small cell not clearly seen in the original clump.

1/9/53. Growth in diplets ✓ or 0. Series A probably dried out
Diplets not apparent at time of fluid addition
Fate of D2, D4? Not necessarily accident. Keep in mind poss. segregation of λ? (what rel'p? of P2).
B1 - probably oil globule only
Transfer to 1 ml broth A9.

1/10/54

Platings (streak out from Imelkoth tiemsfers, P9) on EMB Lac.
P1 = Lac - P2 = Lac +

1106.

A4.	P1
B1	0
B2	P1
B3	P1
B4	P1
B5	P1
C5	P2
C1	P2
C2	P2
C3	P1
C4	P1
E3	P1
E5	P2
F1	P1 + few + papillae / No + colonies
F2	P2
F3	P2
F4	P2
D1	P1
D3	P1

No apparent recombinants in this series

Save B2; B3-4-5-C5; D1, D3; C1-4.
mass stab D2, D4 steel empty P10.

Jan. 9, 1953.

W2338 P1 × P2 (old cultures) ca 1:5 at 10^{30} Remoc ca 1/50 12 N — 3PM
W2344

3 - Y 30.

6+, 6- tested:

Bact test, 2/2/54, B5 → Lac+ V_1 R A 2
These are all ✓ as Gal+, Lac+, -. Lac+ V_1 S B 4
Possibility of recurrent recombination here? Or Lac- V_1 R C 1
is this an illustration of a "twin" set. Lac- V_1 S D 5
Re ✓ OK of 111753.

see over.

10 P1 Plate. Most had 10^2-10^3 in clumped. VG in B1-3-4 D4-5
P10: Litto

1 clump
A 1 0 P1
2 0 P2
3 0 P1+R1
4 0 P2
5 0 P1 (1 col.). Replate from tube (original drop.).

B 2 0 P2
5 0 P1+R1

D 1 P1
2 P1
3 P1

∴ exclude zygotes either result from or tend to form clumps ∴ (P1, P2

Nutritional ~~test~~: 110705 (mess):

O	M	H	M+H	
-	-	-	++	(12h.)

∴ Set up crossing test for A, B, C, D and mess assuming each is M- H- .

Test 1, 2, 3, 4, mess, and W2338
× Y10 (F-) ; W1918 (F+).

all cultures × Y10 [on D(-)] - Sterile
× W1918 → [Ery S Lac] ++ prototrophs

∴ all F⁻ .

3[30] Start in with dress resp. Make necessary steps + tests for pairs.

① Clients [illegible] essential? Remove [illegible] first transfer to resp. Respond to [illegible]

A1 ○ A2 ~~○~~ ○ A3 ○

A4 ○ A5 ○ B5 ○ B4 ~~B3~~ B2 ○

○ Done doubtless typed. — 4[30] PM.

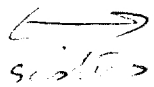
4[03] —  in reverse →  D1 →

Apparently [illegible]! [illegible]
despite [illegible] system [illegible]
separated. Altered [illegible]

after appropriate injection of

420 was able to separate. Then all were widely separated

D2 O D3 O D4 —— D5 O



y 25

D1 O

Add fluid Y: 30

ABD

Compulsory mating + —— ug.

1108

DATE: 1/10/54. REF:

P1, P2 fresh overnight. 12N – 5:45 in Permassay (sep) 1:100
5:55 – Separate midgps, then coalesce.

Repeat, allowing grown in reservoirs. Cells continued to divide in transfer droplets.

4 Drops coalesced. Manipulate A, B further. In B, P1 → ghost.
In A, cells conjoined 00 10^{40} but well separated 10^{45}. Could not force conjunction. Expt. abandoned 10^{45} PM.

objectives of single cell study:

① Genotypes issuing from primary hybrids < Total clonal separations
② Correlation of zygotes with associations with P2 parent
③ Cytol. appearance of the early hybrid
④ Early stages of the hybrid.
Should do ① first.

1/11. Started a few droplets, but milk broke. Spent day largely on review for Ephrussi &c.

1/12. Class; Hanson apptment. Almost no work.

1/14/54.

P1×P2 1:1:10 5:15 – 7:45 pm then diluted ca 1:100. Difficulties in capillaries. Isolations began 9:30 PM. Pick large cells or clumps (v

A1 ○ 9:34.

A2-3 ∞ separated 9:48

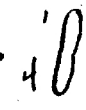
A4 ○ v. motile 9:30. B4 ○

B1-B2 / → ○ , ○

B3 remains apparently ○ but → only ○

C. ∞ in reservoir, → ○ + ∞ in first drop.
9:38 C4 9:43
C2 C3 9:46

D4 v. large 9:58 ○ (later lost.)

E  in reservoir →  spent. →  + ○ → E3

E2 ←  10:12 ←

↙ 10:13

E1 E4.

Later could not find D4, E1 drops. (evap?

A 1 ✓ 2 ○ 3 ✓ 4 ✓ all other P1 S^R (Loc-Gal+ V1^S) P2: Loc Gal-V1^R S^S

B 1 ✓ 2 ✓ 3 ○ 4 ✓

C 2 ✓ 3 ✓ 4 ○

E 1 2 3 4

& control drops n.g. Growth ✓ n.g. ○ and D1-4 ○

DATE: 1/15/54. REF:

C, Dn·1·E4. E1-3,5

P1 x P2 1:1:10 $12\frac{30}{-}$ $2\frac{40}{-}$. Then dilute ca 1:50. (Phages prob. to dup).

3:15 80 → C1, C2 — 3:50 and D2, D3 in same pipette.

D1

D2 D3 → D4.

But all inviable. (dried ?? - tried to add fluid!)

E. // → stayed together → // → 0 + 0 → E3. (seem maximated!)

4:08

E1 E3 separated 4:26 - 4:28

E 0 v. large → E4 → E5.

SURVIVORS. Plate 1/16.

	EMBlac	Gal	SM	TS
E1	−	+	R	S
E2	−	+	R	S
E3	−	+	R	S
E5	±, - very prop.	+ (-)	R (s)	S (K)

∴ has Lac+ Gal- V_1^K (S^s) and Lac- Gal+ V_1^S S^K.

= P1+P2

R if present not numerous. Plate crowded: restricts EMBlac. → very rare, pres. secondary.

rare Lac+ S^R also presumably secondary

(Tiny bad E4 was visible!).

Unless specified, all cultures saved are in peptone broth when mixed or type is present, saline as well as separate

Background for compulsory F-induction. 1111

DATE: 1/19/54. REF:

Fresh cultures (ca 24h. 1:5) W1655, W2338.
Mix under oil, room temperature, 2 hours in their own broth
+
A —
B = vol. fresh broth
C ca. 3 x vol. fresh broth.

and plate out.

A20. Grid to EML.
She found ca 1/10 - 1/5 F+ in A; 0/10 in B, C.

DATE: 1/16/54. REF:

P1 x P2 ... 10^{30} – 12^{30} Refr. to 2^{15}. Dilute 1:25, flat legs. Incubate 2:50 isolate random, empty. large cells.

			N17 (EMB growth)	Gal	Replicate S	Replicate V1	Diagnosis
B5	O		✓	–	+	R	S	P1
B4	O		✓	–, ++	+	R, R.	S	P1, R1
30	O	3:20	✓	–	+	R	S	P1
C4	O	X lost sight	O					
C5	O		✓	–	+	R	S	P1
D5	O		✓	±	–	R	R	P2
D4	O	X lost sight.	O					
20		but add both fairly promptly.						
A5	~~No~~	~~Prophylactic storage~~	✓	–	+	S	S	P1
A4	O		O					
Control + reservoirs (11)			O					
30								

(Note. all self-agglutinated except D5.)

∴ P1R1 may be paired, insignificantly, with P1. These expts. give little information or significance of pairing.

40 Also some time on yeast esci (W46) but could not break walls successfully. Suspensions too thick and numerous cells stick to surfaces of rather large capillaries needed. (Use dessicated now?)

✓D4. No bact. about 5 P1 : 1 R1

50

DATE: 1/17/54 (Sunday) PM. Pipettes for Lager REF: cells.

A4 ○ 2/18
A3. Record? ○ ✓

Probably mixup. Notes also refer to B4 pasture → A4, lost?

B 1 ○
2 ○
3 ○
4 ○

"A4 lost poss. in A4."

C 5/5 marked X. Probably several cells ≥3 ✓ others ○

D1 ○ 1 pipette too small! Carried at tip. Damaged? (was well shaped but too narrow for same cells? Passed others. Use only pipettes for filtrates??)
o.l. ○

D2 } 1/ → ○
D3 }

D4 1/ → ○ other damaged?

5/5

E2 } ○ ("sisters – may have scratched?")
E3 }
E4 ○

Initiate techniques of replacing fluid depths for replenishment and placement. Best arrangement:

	A	B	C	D	E	F
1	○	○	○			
2	○	○	○			
3						
4						
5						

Genus:

	Lac	Gal	S	T1
A3	− →	+	R	S
B1	−	+	R	S
2	−	+	R	S
3	−	+	R	S
4	−, ±	±	R	S R
C3	−	+	R	S
D2	±	−	S	R
3	"	−	S	R
E2	−	+	R	S
3	−	+	R	S

P1, P2 ?

extra fluid at A 0 etc.

Instructions to here

As control check all squares and note extraordinary growths if ever

O/N ✓

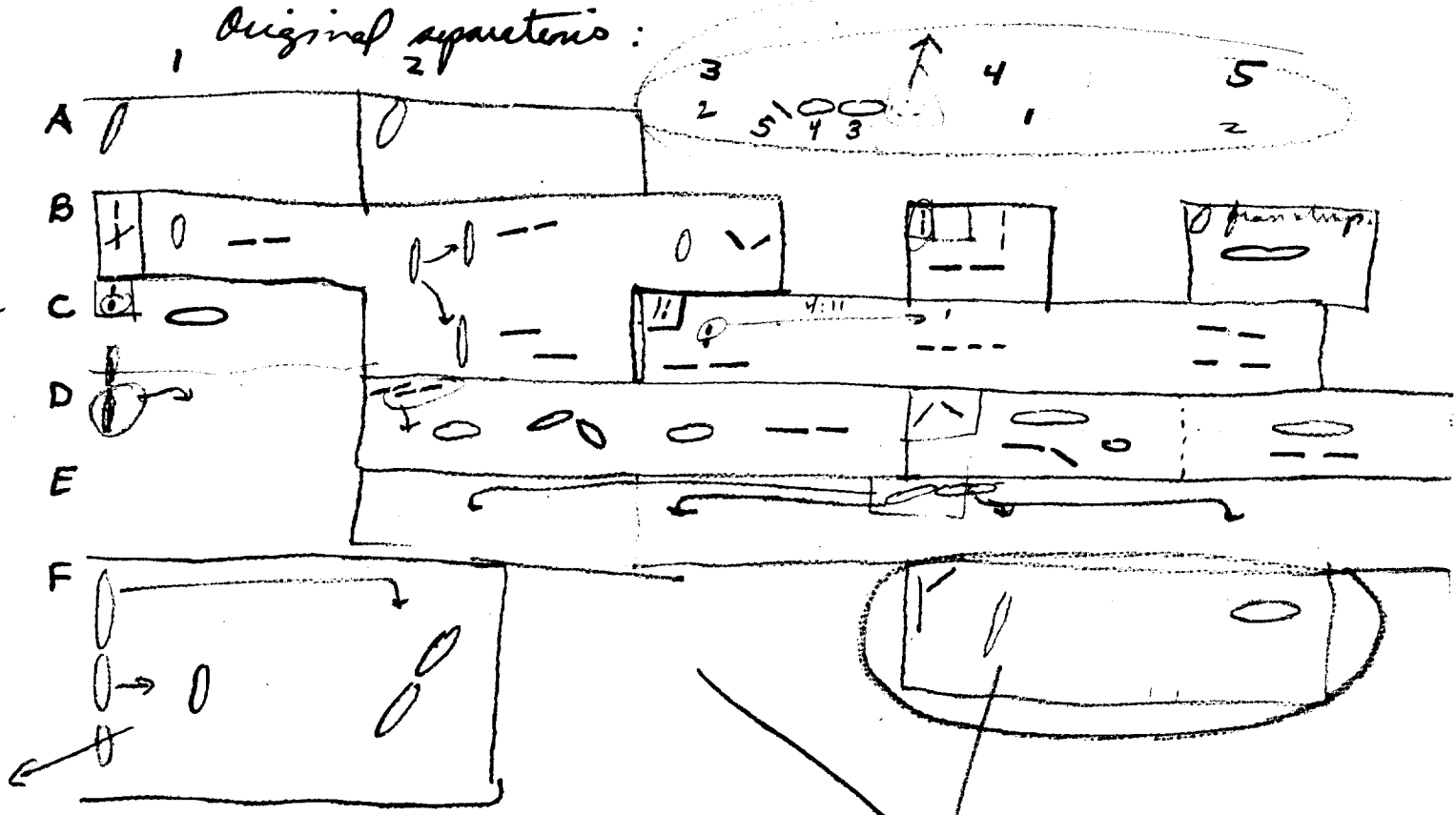
1/18/54.

10:1
P2:P1
1:.1:5 in Pancreas. Admin 1:25 1:20 PM.

$10^{20} - 1^{20}$

30 clones separated from about 2:30 - 5:30 PM (period time setting up dishes, chambers, etc.) Noted that many cells had given 2-4 cells. Refrigerate at 5:30 - 8:30 PM to permit further separation of selected cells.

Original separations:



Do all clones. wait for F4-5 if necessary

7 clones were transplanted, 9:15 - 11:30 PM to separate cover-glass squares, leaving one cell behind in situ. These were plated directly in EMB lactose.

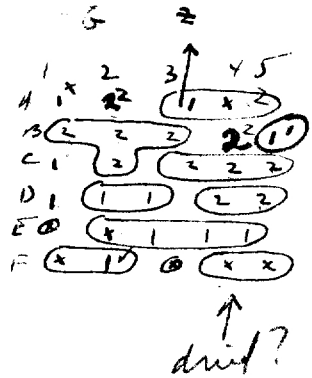
10^{43} exp Tg.

Growth Type

Singles:

A1
A2
B4
B5 (franchimp)
C1
D1

G2: P2
G1: —
G5 P2
G4 P1



G = plate on glass slide

Groups

A 3-4-5

Z 1-5 later. died (dipping)

B 1-2-3-C2

B1 | D3 C2 ?

C 3-4-5

5 1/3 1/4

D2-D3

E 3-4-5 large pair

4 5 / 2 3

D4-5 not stuck

B1 blank
F3 "

F 1-2 / /

F 3-4 /

Deaths may have resulted from dry.... Were anticipated by 10 PM in failure of division of A4, F1, F4, F5. E1, E3 were blanks. E2 should have grown.

W1113 stock susp. was also plated on EMB Lac & Sor.
Counts difficult, possibly ~4 R1 : 32 P1 : ca 200-300 P2. on EMB Lac
An Lac Sor, P1 : P1+R1 = 330 : 34 (very few R1 & P1)

P19. of P2, W1655, W2206 x P1. 10:1 ratio, Inoculate & housed Plate EMB Lac Sor.
W1655, W2206 No SR+ (>300-) each. P2 x gives 42 SR+ / 271 total (P1+P1R1)
(over) = 15.5%

Therefore the incidence of RI/PI isolations is now no better than chance!

Platings

DATE: 1/19/54. REF:

A) Direct platings of clones to EMB Lac agar.

		#cells recorded.	Lac: Found. 1/19 2:35PM.
possible leakage from glass!	1 (A2)	3	3 ±
	2 (A1)	3	0
	3 A5	3	6 ± (1 under glass)
	4 B5	2 (— —)	2 − Both Gal+
	5 B4	11 (one doubled?)	13 ± (1 under glass)
	6 B3	3 doubles	5 ±
	10 F2	3 (fuzz)	3 − (

to 11:10PM 1/18. Excellent recovery.

B) on cover glass. E1, E2, F1, 3, 4, 5 and A4 n.g. (as reported at 10^43 also! as empty sample cell smear)

all others grew, including 2 (extra from A3) 1-5.

	Lac	Gal	T1	SM.	Diagnosis	✓G↑:
2	1	−	+	S	R	P1
	2	−	+	S	R	P1
	3	−	+	S	R	P1
	4	−	+	S	R	P1
	5	−	+	S	R	P1
A	3	−	+	S	R	P1
A	5	±	−	R	S	P2
	A1	±	−	R	S	P2
	A2	±	−	R	S	P2
	B1	±	−	R	S	P2
	2	±	−	R	S	P2
	3	±	−	R	S	P2
	C2	±	−	R	S	P2
	C1	−	+	S	R	P1
	C3	±	−	R	S	P2
	4	±	−	R	S	P2
40	5	±	−	R	S	P2
	D1	−	+	S	R	P1
	D2	−	+	S	R	P1
	3	−	+	S	R	P1
	D4	±	−	R	S	P2
	5	±	−	R	S	P2
50	E	−	+	S	R	P1
	4	−	+	S	R	P1
	5	−	+	S	R	P1
	F2	−	+	S	R	P1

	Lac	Gal	T1	SM.
B4	±	−	R	S
B5	−,+	+	S	R

Lac− Inc Lac+ Pap.

B5 ✓ ∴

∴ all parentals except B5!

High ratio of P2:P1 not necessarily efficient.

Perfect concordance with drop platings.

∴ of original cells, Three were 7 groups of P1, 5 groups of P2, 1 cell → P1 → P1+R1 and 1 group P1+P2 → 2 P2 ✓

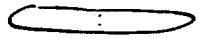
Cells: 11 P1 13 P2 1 P1+R1

✓ 2 blanks and 5 dead

B5.

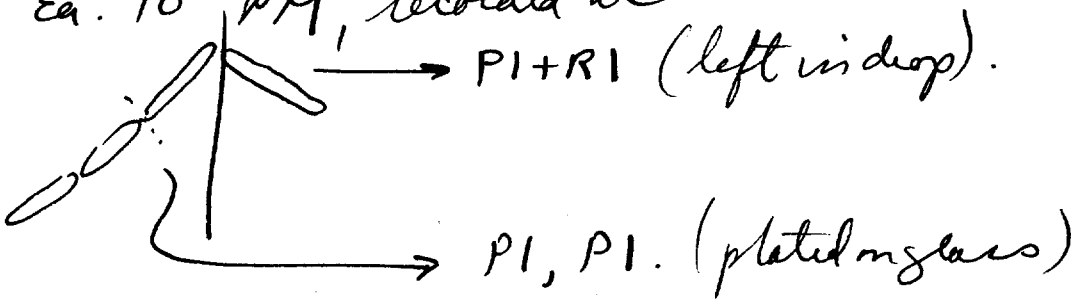
"Isolated as a single O cell from a clump."

at 5:12 PM recorded only as



"Long"

At ca. 10^{30} PM, recorded as



see remarks over next page.

DATE: 1/21/54. REF:

11 – 1[30] P1×P2 1:1:8 Permassay.

Bil. (Inten. visitors ...)

at 5[10-30] 8[30]

X 1. large cells 4[10]

. 2. 4[14]

same at 5[10] 8+smaller in one chain

A 3 } 4 } stayed together to 4:16 → → 5[10]

12+2

* same

5 } 6 } → 5 as unit 6 +2

4+4

0

separated

20 2 [not absolutely cutesi but no other similar cells in neighborhood I.

X 6PM – 8[20]

At 9PM → A1

30

Returns to B:

40 A1 → (B1).

A2 makes → B2

A3

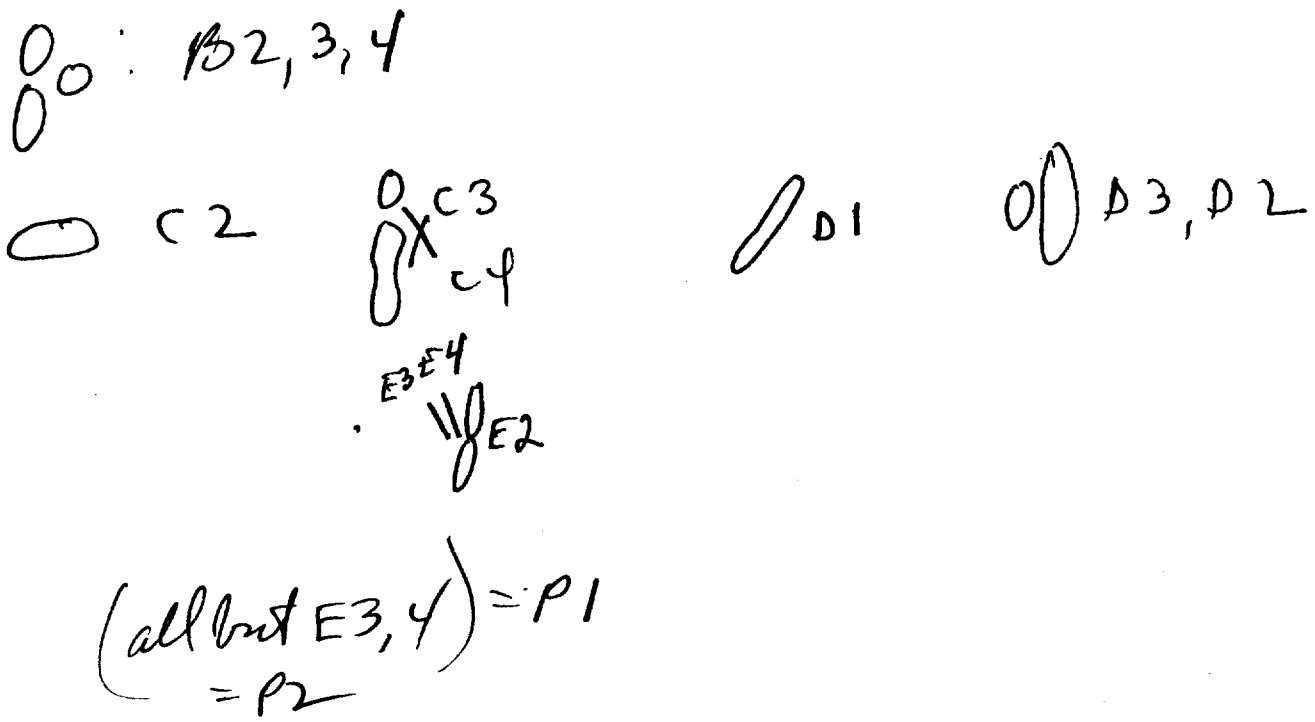
A4 → B3

A5 → A4

9:00PM A3 has:

(over)

1114C.



(all but E3, 4) = P1
= P2

1114
Series

EMBlac

A. 1 → B1 NG

2 ⇒ B2 ⇒ 1 P1 ← R1. Mal V_1 S.R S → P1+R1

3 4 P1 P1

4 → B3 NG; ↗ 0

5 → 10 P1 P1

6 6 P1 P1

B. 2 Lac+ S^R Gal+ V_1^S Pure Lac+ ! % A 2

C. B 2 Lac- S^R Gal+ V_1^S P1

3 " " P1

4 NG: " " P1

C 2 " " P1

3 " ✓ " P1

4 " " P1

D 1 " ✓ " P1

2 " ✓ " P1

3 " ✓ " P1

4 " ✓ " P1

E 2 " " P1

3 ± S^S Gal - V_1^R ✓ P2

4 ± S^S ✓ Gal - V_1^R P2

2 P2: 14 P1 :

A1 B1.  → A1, B1.

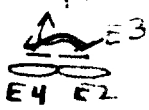
A3 B2 B3 B4 C1 C2 C3

B2 B4

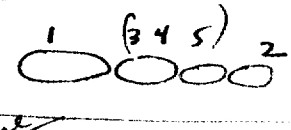
(B3, A3)

D1-2-3 D4-D5

E1 ?lost

E. 

E4 E2

F. 

all pure

	Growth	Lac (A25)	S	Gal	V1	
A1	✓	−	R	+	S	P1
B1	✓	−	R	+	S	P1
A3	✓	−	R	+	S	P1
B2	✓	+	−	−	R	P2
B3	✓	+	R	+	S	P1
B4	✓	+	S	−	R	P2

SN → grew out A25 (P1 morph) − (P2 morph) ✓ — λ^S ✓ — sent Wold for recheck

A25 entire OK.

C1	✓	−	R	+	S	P1
C2	✓	−	R	+	S	P1
C3	✓	−	R	+	S	P1
D1	✱ ×	−	R	+	S	P1
2	✓	−	R	+	S	P1
3	✓	−	R	+	S	P1
D4	×					
5	×					
E1	×					
2	✓	−	R	+	S	P1
3	✓	−	R	+	S	P1
4	×					
F1	✓	−	R	+	S	P1
2	✓	−	R	+	S	P1
3	✓	−	R	+	S	P1
4	✓	−	R	+	S	P1
5	✓	−	R	+	S	P1
G1, G2		− −	RR	++	SS	

(P1 2 P2 —

(+ destruct appearance on EMB bac and prob? Gal possibly a minor mutant) P1 P1

first P2's { P1 No Ey; P1 Ey }

{ P1

1/23/54

1:1:3

4:25 - 3:05 at 37°

3:05 - 3:15 at RT to Lab. for yeast, - 5:05. Plating at 4:05 PM:

EMBlac sm. 3 P1·R1 per 167 (2%).

EMBlac: probable zygotes = 5/130 P2 / 113 P1.

In many 3/4 of 3 plates, colony appearance suggests following distribution: R1 P1 (P1P2 R1P1P2). Simple mutant colonies P1+P2 not counted here.

111 ≠ HH 111

This may of course be maccinate.

G1,2 = pur. D1, D2. " controls OK.

Grew P23 - P24 at R.T.

A1,3, B1,2 C1,2,3 D— E2,3 (6?) F1,2,4,5,6.

∴ growth failed in (B3); D1,4,5; E1, E4;

controls OK. Some may have dried out.

Morphology ~ Lug 28A except: B2 (motile, bacillary)

B3 has self limited surface development. Hold!

large, wide elements

B4 grew in a small droplet. bacillary but not motile; - , - -

D2 small drops. D3 small drops D4 — D5 drop (2 colonies?)

E1— E4 — F3 small drop. D6 drop

Induction.

1/25/54

W1655, W2338.

exp. cultures in Rivasseau, 1:1:2 $3\frac{20}{}$ at 37°.

$3\frac{40}{}$ dilute 1:50. Try to find F^+/F^- pairs. But dosages are too dilute. However.

D4-5
E4-5 from poss. pairs, but both are morph. W2338 next day.

D1-2-3 from poss. { 10-2. D3 neg.
10-1. D1,2- to EML.

controls OK. A-E 1-5.

D3.

		EML F test (659)	
D1	-		
D2	-		
D3	-	0	0/4
D4	+		8
D5	-		
E4	+		8
E5	-		

1.0.

1/26/54.

P1 x P2 (old) 1:1:8 12[15] – 2[45].

(1) Plate on EMB Lac+ to see V segregation (cf. V_1^S character of all SR+ so far!).
(B) 20 hrs ca 3PM.

(2) Dilute 1:50 for single cell...
High ratio of minicells. This experiment resolved rather few "glom cells" still possibly in stationary phase?
Drops empty overnight still stuck at N28.

Crew:

			EMB Lac	Gal	T1	lac	
1	B	3	–	+	S	–	S R
2		4	–				
3		6	–				
4	C	4	–				
5		5	–				
6	D	3	–				
7		4	–				
8		2	–				
9	E	4	–				
10		2	–				
11		3	–				
12		4	–				
13		6	–				

Rev ++

[no lac I seen; T1: S, R.]

S – R P1

P1

off-stuck lac, gal almost all P1

23 P1
1 P1, R1
3 P2
compare with high incidence in δ!

Plate

P1, R1 < lac + gal + S V_1
lac – gal + SR V_1

Cols. stuck
any Lac+ V_1^S?

NG (drop growth also peculiar)

14	F	1		+	S		
15		2	–				
16		3	–	+	S		
17		4	–	+	S		
		5	–	+	S		P1
18	G	1	–	+	S		
19		3	–	+	S		
20		5	±	–	R ±	S	P2
21		6	–	+	S		
22	H	1	–	+	S		P1
23		5	±	–	R ±	S	P2
24	I	1	–	±	S –	R	P1
25		2	–	±	S –	R	P1
26		3	±	–	R ±	S	P2
27		6	–	+	S –	R	P1

L 4 was murky. 2s fading sys! 4 #5 n.g. But this was 150 minutes more.

1117B. Pick Lac+ from EMB Lac to classify progenitors of type and to characterize Lac-V_1 segregators.

Colonies 1, 2 6, 7 Brush 6 isolates on EMB Gal/TI (unpur.) and indicate TI/Gal types Gal+ V_1^S/Gal+ V_1^R respectively type each & save dried

Colonies 3, 4, 8, 9 Lac types on streak must be used to be repicked

---

1117B: Plated (TC N.) 2×10^{-7} ml per plate.

EMB Lac			Lac am	
$\bar{X}$:	Lac+	40.6		Lac − 48.8
	Lac−	55.2		
	+/−	3.5	(SR+)	+/− 3.7
		99.3		

∴ SR+ = Lac+/−. Plate all showed Poisson of each type.

$$\% \text{ zygotes} = \frac{3.5}{99.3} = 3.5\%$$

$$= \frac{3.5}{58.7} \text{ of Pl}_{\text{Type}} = 5.96\%$$

...inputted ca ~~1.43~~ 1.43 zygotes from 24 Pl- isolations.

Found 1 (see new): not different from chance!

Total χ^2 testing Poisson variance: $\chi^2 = 56.0$ d.f. = 50

Plate Galactose Newton Motor
2 - 9
3 - 8

$\bar{x} = 10.583 - 9.950 = .633$ $p = .6-.5$

1117

DATE: EMB Lac REF: EMB Lac Sm

	1 Lac +	2 Lac −	3 Het	4	5	6	7 Total	8 Het	9 − Mut	10
1	48	51	4				55	4	51	
2	44	48	4				56	1	55	
3	31	67	3				53	7	46	
4	52	61	3				62	9	53	
5	28	49	4				42	3	39	
			3					0		
Σx	203	276	4					3	$\Sigma x = 244$	
Σx^2	8089	15516	4					7	$\Sigma x^2 = 12072$	
$\bar{x}$	40.6	$\bar{x} = 55.2$	2					4		
Σd^2	447	281	1					3	$\bar{x} = 48.8$	
χ^2_4	11.0	5.1	8					3	$\Sigma d^2 = 165$	
p	.05-.02	.3-.2	3					2	$\chi^2 = 3.4$	
			3					4		
			2					2		
			2					5		
			3					2		
			2					4		
			4					4		
			5							
			3							
			5							
			6							

Lac + vs. Lac −
$\chi^2 = \frac{73^2}{479} = 11.1$
$p < .01$

Lac − vs. Lac
$\chi^2 = \frac{32^2}{520} = 1.97$
$p = .2-.1$

$\Sigma x = 78$
$\Sigma x^2 = 326$
$n = 22$
$\bar{x} = 3.545$
$\Sigma d^2 = 326 - 276.545 = 49.455$
$\chi^2 = \frac{49.455}{3.545} = 14$ $p = .4$

$\Sigma x = 67$
$\Sigma x^2 = 333$
$n = 18$
$\bar{x} = 3.722$
$\Sigma d^2 = 333 - 249.389 = 83.611$
$\chi^2 = 22.5$ $p = .2$

$F_{17,21} = \frac{4.918}{2.355} = 2.09$ $p > .05$

$s^2 = \frac{133.066}{38} = 3.502$

$s^2 = \frac{145}{40} = 3.625$

$\chi^2_{[1]} = \frac{9.9[.177]^2}{3.625} = .0856$ $p = .75$

$\chi^2_{[1]} = \frac{(2232)^2 n}{\pi} = \frac{2.8 \times 10^9}{1.72 \times 10^9} = 1.6$ $p > .2$

∴ Het variance Poisson, means homogeneous.

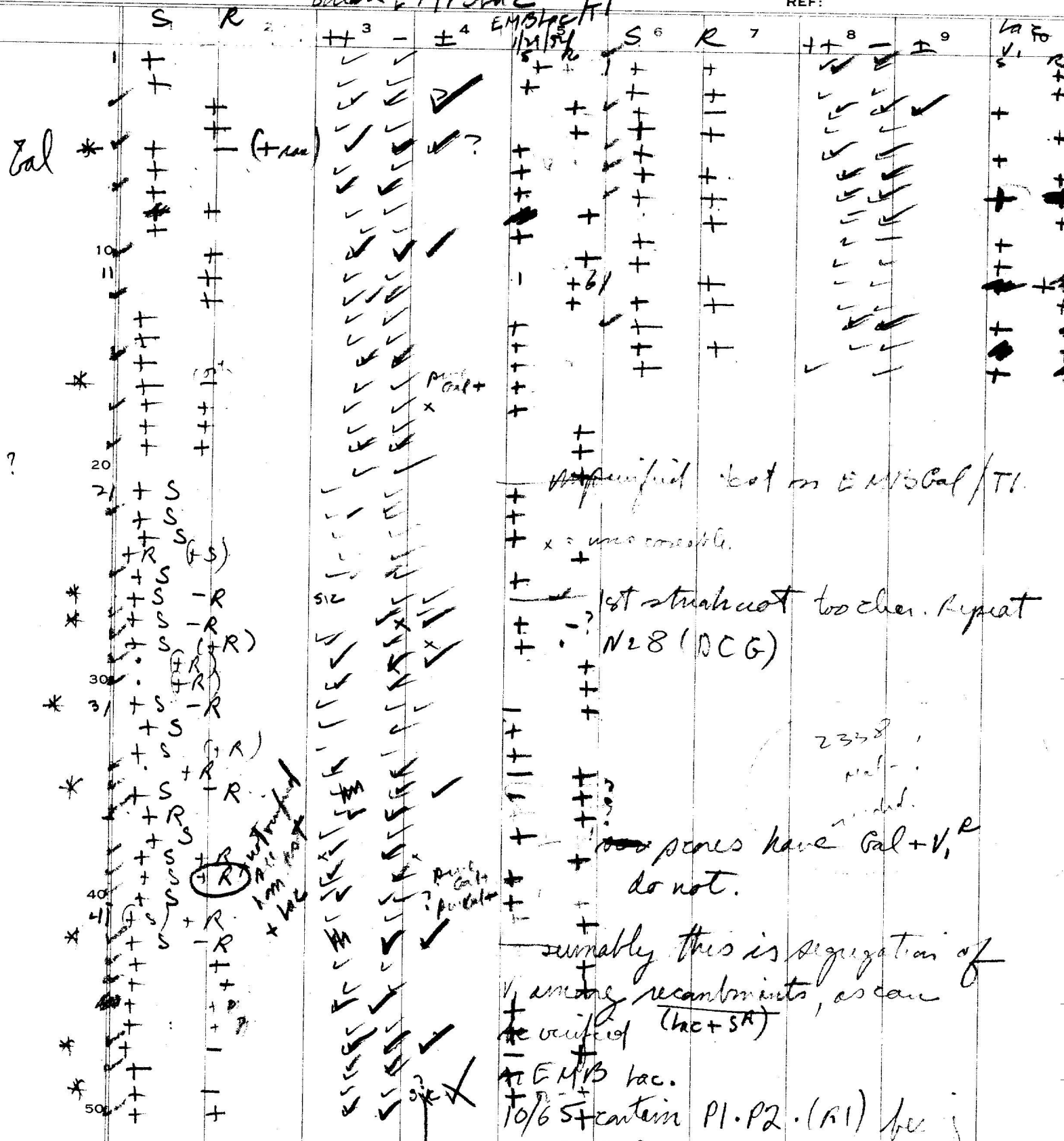
	Col. 2,3	Col. 8, 7-8	
+	276	244	520
−	18	+ 24	42
	294	268	562

∴ Frequency of het. homogeneous in the two tests het : lac.

11173
(1)

EMBGal 1/27/54

DATE: V1 Strain EMBLac REF:



? 20

? 30

? 40

? 50

Repeated on EMBGal/T1.

x = uncertain.

1st streak not too clear. Repeat

N28 (DCG)

2338 rel- ind.

Gal+ V_1^R ones have

do not.

Presumably this is segregation of V_1 among recombinants, as can be verified (Lac+ S^R)

EMB Lac.

10/6 S+ contain P1·P2·(R1) but rechecked. Some recorded as Gal+ V_1^R probably also have V_1^S but certainly deleted.

? Re-isolate on EMBGal (for Gal−) P1+P2

of 64 byte columns (1119B) scanned,

31 backed $Val + V_1^R$ ($\pm Val - V_1^S$), but found $Val + V_1^S$

32 had $Val + V_1^R$, at least 21 also $Val + V_1^S$ (pseudoparental) and recombinants

Thus an origin for 1:1 ratio S/R ~~if~~ the latter pure. Test for homogeneity of recombinants

---

① For this purpose, ignore pure + S strains

② Check ① Loc – four Loc + four others for homogeneity

~~Cut between Loc – V_1^S and pp pur + R~~ (use diff with 1119).

Reason Loc/S segs generally with Val

Nought not to be kept definite

Ref ~ 27,57 es

Have) Loc – V_1^R recomb.

These should have been detected if present,

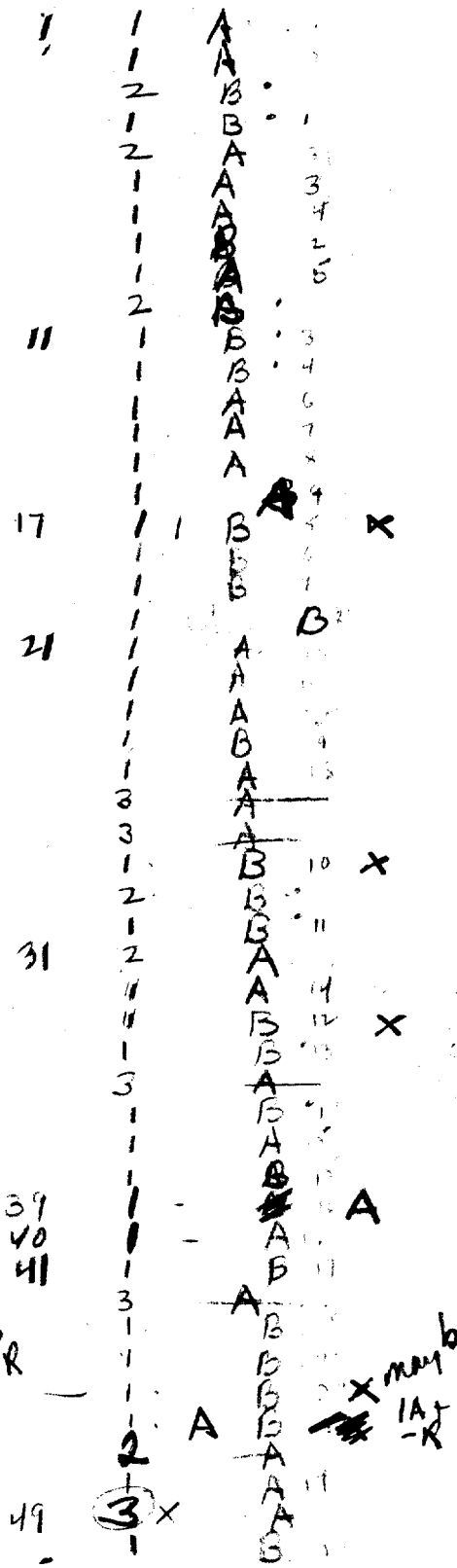
in view of all cases with Loc exp.

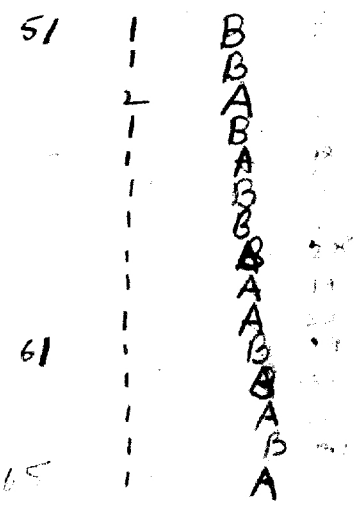
was V_1^S.

Reread scores

Zygote chart 1 R1
2 P1+R1+P2
3 P1+P2

V_1 character (incl. 3) 11/28/13
A pre-V_1 Gal(+)
B V_1 Gal+ present (1)
• ~~—~~ V_1 + notable





also Loc - V_1 R A+ -

Final
~~2A~~ 25A
53-1 < ~~5B~~ 4A 26B
7-2 < 3B
5-3 <

+350 type 1
B type 2
+1 4 type 3

at 2½ hours

4 to be verified

Va typing of V_1 R might be useful in separating type 2 and 3

2B = got the overcast stage (normal, confused c 3).

R1+P2 S

~~Totals 1,2 A 2B~~
~~1,2 B 3A~~

Notable discrepancies in Gal/Loc for #17, 28, 33, 45, 46, 57. were classified as 1B after confirmation, with 22 1A : 25 1B.

46 + S Hea - R

x maybe 1A+ -R

In Class 2, which were
detn. tot. as Lac+V_1^S Lac−V_1^R ?

3 Bur
5 Ayes
10 Bur
7 ; B
3 A yes
3 A yes

17 A

∴ Totals:

1 < 25 A Lac+V_1^S
    26 B Lac−V_1^R −2

2 < 4 A
    3 B.

$$\therefore \frac{\text{Lac+V}_1^S}{\text{Lac+V}_1^R} = \frac{25+2+4}{26+3} = \frac{31}{29}.$$

∴ no discrep. here see 1117C

If retain as 1-cell isolation:

Rexamine Lac/T1 Repeat Lac/T1

17 Clear plaques +
28 " " +
33 " " +
45 " " ++
46 " " +++
57 " " ++

probably Lac+V_1^S
mixed: Lac+V_1^S/Lac−V_1^R Lac+V_1^R
Lac+V_1^S/Lac±V_1^R (P2)
~~Lac+V_1^S/Lac+V_1^R~~
clearly Lac+V_1^S
Lac−V_1^R

and score as 1 each.
or remove from now.

accidental lac− only.

+ prob
++ certain
Lac+ V_1^R

17 28 33 may have −S,+R
45, 57 have −R+S
46? may have Cal−V_1^R

1 A
28
33
45
46

Homogeneity tests.

1117 G

1/29/54 (1117 G 4) II ~~Gal+ Lac+~~ of V_1^R Lac+ Gal+

Pick Lac+ (pref. not ± or −) colonies and test on Gal/TT.

1/29	10:	Gal+	5R	1 S
1 type	26 3:		6	+ R
2, 3.	10 25 29		6	+ R

(v Lac+) ~~Repeat test~~ Restrict to R, S

Lac+ components (Lac− presumed V_1^S)

~~Clearance~~ homogeneous, &c.

1B	50	6		1 R
1B	51			+ R
1B	34			+ R
1B	3		6	+ S ! :: 1A like
	43		6	1 R
	44		6	+ R

~~Pick~~ (Lac+ colonies picked)

Could this be a Lac− Gal+ by mistake? (= P1)

When streaked, S and R compare both pure Lac+ (R some but weaker!)

Some for new ½ crosses, in 2B types

There is likelihood of recurrent recombination where there was a pair or not! This is probable explanation! Restrict on EMB Lac 1 soon for further analysis.

Conclusions:

① Most zygotes are homogeneous in recent unless P2 still present. V_1 heterogeneity might be useful to discriminate primary + secondary recombination.

② V_1 S zygotes 1:1 of R/S

2/3 46: 10 Lac− are V_1^S 57: 9 Lac− : 2 V_1^S 7 V_1^R

1117B

		V_1		
57:	4 Lac+	S	4:	4 Lac− S
			11:	4 Lac− S
			27:	4 Lac− S
			30:	4 Lac− S
58:	2−	R		
	2−	S	assoc. with Gal+ (Lac+?) V_1^R ~~and~~ any Lac+ V_1^S?	
36	4−	S		

note, e.g. 58 Lac− also mixed R/S so

~~that there is~~ presumably Lac+R, Lac−R, Lac−S

Structure of V_1^R from 45: Gal+, mostly Lac− (some fuc+ papillae)

∴ 45 contains Gal+ { Lac+ V_1^S ~~Lac+ V_1^R~~ ~~Lac− V_1^S (PI)~~ Lac− V_1^R

Lac+ V_1^R? (new recomb.? cf. 1107B5!)

46: Gal+, mostly Lac+ ~~(some −?)~~

∴ Lac− V_1^S presumed. (PI)

Lac+ V_1^R

Re ~ exceptions

1117B

2/2/54 stzeq

① Restreak on EMB Lac, Gal:

17	pure Gal+ ;	lac+ seems weak.,	lac+/−
33	"	lac+	"
45	"	"	"
46	"	"	
57	"	"	

Types definitely seen.

1117 B.

Gal+ unless qualified

. = tested for. by present test

□ = present

⊙ = " burnt by present test.

	(Gal−)	Pl. lac−V_1^S	− R	+ S	+ R	tested
2A	3 ✓	□			✓	6
	4	□✓	.		□	4
3B	10 ✓	□		✓	✓	6
	11	□✓	.		□	4
	27	□ −	.			4
2B	29 ✓	□			□ −	6
	30	□✓	.			4
	34	□		.	□✓	
	36	□✓	.		□	
	39	□		✓	✓ □	
	43	□		.	✓	
	44	□		.	✓	
	50	□		.	✓	
	51	□.		.	✓	
	58	□ −	✓		□	4

from original tab. as 1A!

but not all

∴ many (~~all~~) 1B types are plus V_1^R in lac+. Lac− not tested [defer full analysis to multimarker test] (and lac− V_1^S for "most" but cf. 58).

	17	✓	✓R	✓	✓
	28	□	✓	□	□
	33	✓	✓	✓	✓
	45	✓	✓R	✓	✓R
	46	✓	✓ (not R)	✓	□ + [illegible] R
	57	✓	□	✓	⊙

cf. 1107/35

∴ a fair proportion of colonies may have, in addition to −S (= P1) 1, 2, or 3 addl. components. How many is not clear, as not many examples

R: weak + only.

If patient lac+ V_1^R was examined further. But note 39 above. V_1^R is therefore perhaps slightly overestimated

- Further colony analysis 2/3-4/54

Lac - 10.

17C1 4R 4S

2 → 4R

3 3R

4 8S 0R.

(probably before 1 ?)

Counted Stats 2/4/54.

type 1A (presumed simple!) 25 (pure $V_1^{s\,<\,+}_{-}$) − 25 + 5C

1B (genetics simple but possibly containing +S also) 24 + 5C +1

C. Containing $^{+}V_1^{R}$ and $^{+}V_1^{S}$: (17, 28, 33, 45?, 46) 5.

D. (77) Containing +R, −S, −R. (1)

∴ Detected +R (parental) = 30

Screened +S (+R not eloquent) = 30.

= 1:1

? Possibility of a detected recombinant to bit, ?

(1) Loc± S^S recombinant, Loc+ S^R (Gal+).

2. Gal+ V_1^S recombinant Gal+ V_1^R (Loc−)

i.e. pure V_1^S or derived V_1^R

S^S " " S^R.

Left arc: Loc− S^S among Loc− S^R (probably best handled with additional genetic markers)

and ~~+ − V_1^S S^S among Gal−~~

SR+ colonies picked & purification to EMBlac.
40 + 17. 1/31 Replica to EMBlac ± T1.

C1: Each photo on EMBlac was lac+/lac−. With T1:

Unaltered recipients (i.e. lac+ V_1^R (lac− not verified)	: 19	6	25
pure sensitive lac+ V_1^S/lac− V_1^S	: 17	9	26
← lac+ V_1^S/lac− V_1^R	: 3	2 (1117C 1-3)	5
~~Obvious lac+ V_1^S/lac− V_1^R~~	: 1 (1117C)		1
		plate 1	plate 2.

not verified

Or in sum: lac+ are 31 S : 26 R / 57 total.

Include at least 6 lac− V_1^R recombinants, possibly more.

1117 B: 31 S : 29 R. per

	recomb. (S)	(R)
B	31	26
C	31	29
	1	1

Totals:

Thier.

1082	26	25
1119	15	18

agreement is obvious
Seelso 1082. (25R : 26S)

Still queries on incidence of lac− V_1^R recombinants and association with lac+ V_1^R (S^R). Are the lac− components of these pure?

Misc cells.

DATE: 1/27/54 REF:

1235 Overnight P1, P2 1:5 310 1:1.5 X. Obs at

(1235 cross W2377 + W2341.) − 3:55

Cross with P ♀. EMB lac strains Gal, V_1, S (Lac)

A 1 ✓ — all nic Gal+ V_1^S S^R Lac− S =

2 ✓ —

3 tiny dots 0 — except C2 = Gal− V_1^R S^R Lac

4 ✓ — = P2

5 ✓ — and E5 = P1, P2 (SR+

B 1 ✓ — V_1^S V_1^R papill

2 0

3 0 ✓ —

4 ✓ —

5

20

C 1 ✓ —

2 ✓ ± and 2 = ? colonies (= contamination? (also Gal− V_1^R Spath, not in orig. tube)

3 ✓ —

4 ✓ —

5 ✓ —

D 1

2

3 ✓ —

30 ✓ —

5 ✓ —

E 3 ✓ —

4 ✓ —

5 ✓ —

40

F 4 ✓ ± and − unequal proportions P1, P2 (Successive plating of 1st is needed.) { N_0 SR+ (exc. at intersection). Lac− = SR on replica = Gal+ Lac+ = S^S = Gal−

5 ✓ —

50

entles OK

A3 OK

B2, 3 n.g.

18 P1

1 P2

1 P1+P2

DATE: Jan 27, 1954 REF:

W2377 x W2341 12:35 – 5PM.

A EMBlac

B EMBlac sm

C EMB lac T1. (for S-ratio)

D 10⁻¹ C ca 10X

C: ~~colonies~~ Lac–, Lac+ carp. (6 have only small papillae? unlike anything noted with streptomycin.)

19/165 tot.

11/122 } (patch-mated)

5/170 }

35/357 +-/-. A: + ca = . Not acc. scorable for +Vs ±, –.

B: +-/tot = 12/171

6/149

15/236

14/231

47/787

Sect virtually all of these colonies in B are in C are Lac+/Lac– sectored, no recombinants are here missed that are not otherwise picked as segregating subclones +/–.

Why discrepancy in rel. comit? 10% in C
ca 6% in B

Extract (apparently) in B: 2+1?
C: 0

a) Contains some 'pigillate" V_R Lac+
b) on other hand, some S^R lac+ are V_S!

1/28. Pick B, C, D to EMB lac + slant for later study.

This was done for rec. yield also.

alteration of phage on secondary crossing?

1119
B – C – D

DATE: 1/31/54. REF:

(B) Plates stored from 1/28 in frig.

Replica B to Lac, T1 to score the V_1 segr. ratio (cf. 1117B). all colonies have Lac – V_1^R (P1 residue); score strokes for presence of Lac+ s/r T1.

2/1/54

Lacchen:	– T1	+ T1	Score type
1	+	(–) +	1
	+	+	1
	+	+	1
	+	+	1
	+ –	– +	1
	+	–	2
	+	–	2
	+	+	1
	+	+	1
10	+	–	2
	+	–	2
	+	–	2
	+	+ –	1
	+	–	2
	+	–	2
	+	–	2
	+	+	1
	+	+	1
	+	– +	2
20	+	+ –	1
	+	–	2
	+	+	1
	+	+	1
	+ –	– (+)	2
	+	+	1
	+	+	1
	+ –	– +	2
	+	–	2
	+ –	+ –	1
30	+ –	+ –	1
	+	– +	2
	+	+	1
	+	+	1

∴ 18 type 1 15 type 2.

Lac+ V_1^R (par) V_1^S (recomb.)

Totals now are ~~98~~ parental ~~99~~ 99

Lac / V_1 : ~~105~~ recomb. 10^2

– among Lac+ S^R recombinants, ~~1/55~~ < 10 + all V_1^R

$X_1^2 = \frac{12.5}{101.5}$

$= .1$

$p = 0.8$

including V_1 reversal.

Lac+ Lac– < 10 all V_1^R. ∴ type 1.

Residuals 1119 B ~~[illegible]~~ B 20 for components:

actual numbers therefore type 1 : 19

2 : 14.

$\chi_1^2 = 0.7$

But p_{total} almost too high!

Scores: 1 : T1 no effect = +R/–.

2 Lac+ V_1^S/–R

[3 Lac+ V_1^S (+ V_1^R ?)/–R] probably = 1, phage inadequate mixture possible but ignore here. They would not influence ratios in some marked 3. in any event.

DATE: REF:

Replaces lac+V,R/−R to EMS lac + Sm.

2/1/54.

C1. 30. Lac+ equally prominent in EMS lac sm.

C2. ← C 10 / D 20. " " " " " "

D1. 23.

D2. 29.

Undetermined

112 all orthotype S^R among lac+V_1^R recombinants. Still possible that some S^S are present but obscured in mixture but no evidence for it. . . S behaves very differently from Lac, V_1. Other data show extreme rarity of Gal/S recombinants but this should be reviewed in multipoint tests.

Recheck sin 1119 only to verify 13, 20.

2/5 See also 1076, just noted!!

Note also, in 928-94) W1895 × W1177 among lac+S^R's, reported 4/S; 7 4R. (poss. of selection through linkage to TL?). Needs to be repeated.

Casual 1-cell cool. 1120

DATE: Jan 28, 1954. REF:

	1	2	3	4	5	6	7	8	9	10
Cross: old mouse P1×P2 1:1:8, 2:10PM – 3:30										
Setup 3:45. Morphol: (not [illegible]) Blue dyes 24 hours AFRT To										
Grow N29: N30 Gel Tube S A 30.										
A 1	∞	✓	not mot. (macro-)		+ S	–	R	P1		
2	X								15 P1	
3	X	✓	1						5 P2	
4	∞	✓	rec knotting?		+ S	–	R	P1		
5	o	✓	1		+ S	–	R	P1	1 P1, R1	
B 1	∞	✓	1		+ S	–	R	P1	[redacted]	
B 2	o	✓	0							
4	o	✓	1		+ S	–	R	P1		
3	∞	✓	1		(+S) (+R)	–, +	R	P1	R1 (V, R)	
5	o	✓	0							
C 4	o	✓	0 rec knot?		[redacted]	–	[redacted]	[redacted]	NG (with also unspiral?)	
20										
C 1	∞	✓	1		+ S	–	R	P1		
2	∞	✓	1		+ S	–	R	P1		
3	∞	o	0							
4	v. Sup.	✓	2 (+1?)							
5	o	✓	2		(+S)	(–R)		P1		
D 3	∞	✓	1		+ S	–	R	P1		
2	∞	✓	1		+ S	–	R	P1		
D 3	D3	✓	1		+ S	–	R	P1		
E 2	∞	✓	1		+ S	–	R	P1		
E 3	2 3	✓	1		+ S	–	R	P1		
D 4	∞	o	0							
D 5	o	✓	–		– R	± S		P2		
40										
E 1	∞	o	0							
E 4	∞	✓	1		+ S	–	R	P1		
F 1	∞ not dump.	✓	1		+ S	–	R	P1		
F 3	(hard)	✓	2		– R	± S		P2		
4		✓	2		– R	± S		P2		
5		✓	2		– R	± S		P2		
dump G 3		✓	2		– R	± S		P2		

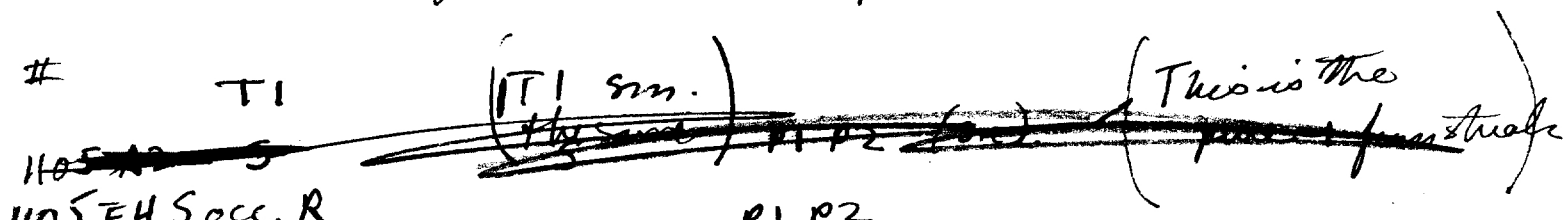
Previous feeding waste stock not exhaustive as well as cross back. Now omit the stock except where specific.

F3 cultures O.K. Selection for [illegible] per [illegible]

O4 O5 O6 O3 cells!

Review V, segregation in 1-cell recomb.

Cross bench against T1, T1 sm / Lac



1105 E4 S, occ. R — P1 P2

1107 -A3 S	P1 R1
-B5 S, R	P1 R1
1110 E5 R, S	P1 P2 ✓
1112 B4 R, S	P1 P2 ✓
1113 B5 S	P1 R1 ✓
1118 E5 S, R	P1 P2 ✓
1120 B3 R, S (+ −)	P1 R1 ✓

now heavily mixed with R1 also.

New information:

1107 A3 R1 is S

1107 B5 ~~R1 is probably R~~ (+ R, + S, − R, − S)

1105 E4 } stated as P1, P2 needs check.

1107 ~~A3~~ }

misread label ("B52").

DATE: 2/1/54. REF:

P1, P2 Mix fresh cells ~~1 : 2~~ 2 : 2 : 2 3^00 (− 9:30 PM.)
Look for clumps, pairs. Most isolations ca 4^30 .
— Note on motility. W2344 seemed much less motile in dilute growing cultures c̄ 2338 than otherwise. Compare effect of growth phase, mixture.
See protocols A1, E1 ng. Controls OK except (1 very large drop = Lac± Gal−) (Checking of possible accident).

From clumps.

D
C inhibits
as Lac ssn.

Loc	S
Gal	V.

	A	B	C	D	E	F	D	
1	x [] large	− 1 R	± S	2 cells!! R	x	± 2 S	± S	2
		+ S	− 2 R	− 2 R		− R	− R	
2	− 1 R	− 1 R	± S	+ S	− 1 R	± 2 S	± S	2
	+ S	+ S	− 2 R	− 2 R	+ S	− R	− R	
3	− 1 R	± 2 S	± S	− 1 R	± 2 S	± 2 S	− R	1
	+ S	− R	− 2 R	+ S	− R	− R	+ S	
4	[illegible] 2 S	− 1 R	± S	± 2 S	− 1 R	± 2 S	± S	2
	− R	[illegible] S	− 2 R	− R	+ S	− R	− R	
5	± 2 S	− 1 R	± 2 S	± 1 R	− 1 R	± 2 S	− R	1
	− R	+ S	± 2 R	[illegible]	+ S	− R	+ S	

∴ No zygotes from these clumps.

P1 : 11

P2 : 17.

Note: clumps AB 1, 2
C all 2
D one 1, 2
E 1, 2
FD all 2.

Good agreement c̄ morphol. prediction except for A4 which is drawn as smallish.

P3 [illegible] c̄ P2 [illegible]

Use W2384 as P1 from here on. To date P1 has referred specifically to W2338

Misc. segr. 1122

DATE: 2/1/54. Screen for carbons REF:

A. W2381-6 × W2344

2:45 – 9:30 PM Plate 10^{-6}, 10^{-7} on EMB lac ±sm, EMB Mal sm.

[Usefulness of new Mal⁻ markers, esp. in assessing residual P2 carps J.

	EMB lac sm	Mal sm	Mal sm	EMB lac
	✓= SR+		1 pinfl = 3/1000	
1	✓ –	0		
2	– ✓	0	+/–	
2/2 (3)	✓ ✓	0	0	
4	– ✓	0	0	
5				
6	– ✓	0	1	

Best these Mal⁻ evidently all/almost (nearly totally) the S^R segregators; and maybe equivalent. Use 2384 for future studies.

whose A5?

B. 3.20 W2057 × P1
2:45 – 9:30 PM

Plate 10^{-6} EMB lac sm. Streak lac+ S^R to confirm orthotypy for M+l, Mal among these recombinants in lines 1×28A.

EMB lac sm:

✓✓ (Some pure lac+). Pooled and restreak → ca 2% M+l – But analysis 16 all lac+. from EMB lac.

2/2 Mal sm: pure Mal+. (Some colonies mottled very likely lac+/–.)

30 EMB lac: some lac+/–, often too crowded. ~~Pick as likely to EMB lac sm. Pooled restreak~~

From EMB lac sm 1 ● lac ✓: 9 pure V_1^S 1 V_1^R (all M+l+) (different from line 1×28A in V_1 segregation!)
(V_1 orthotypy).

40 C. P1-P2 (W2338 × W2344) plate EMB Gal T1 } to test for lac +/– ratio among $V_1^R S^R$ recombinants (cf 1082.)
(plate as from 1121. 9:30 PM) EMB lac sm T1 }
did not exist (over)

50

Forebulk, pick EMB lac lac+ (A) and lac +/– (B) for Mal test.

10 · Mal⁻ SR+ 3 Mal +/– Spath, SR+ (over)

Control:

Plaque: heavy colonial background and partial survivors. Phage titer evidently inadequate.

Crosses:

EMB Lac sus T1: Lac+ probably >> Lac− but many of former also nibbled! By more dilute platings of cross (same T1) Lac → Lac+!

EMB Lac T1 Fewer TIR and: not reliable!
Repeat & better phage stock!

Conclusions:

A. Mal− has all effectively linked to S, shows orthotypy

B. W2057 has same elimination patterns for Mal, S, M R as W1895, but note that the V, ratio is quite different!! (look for V_1^S in × W2377?) Have these V_1^R been tested with T5? This is same V_1^R as W1177 (= W−1 V_1^R); may be different loci.
Also earlier data with W1895 × W1177 where V, Ratio was strongly orthotypic!

C. No information: need fresh T1 for selections.

2/7/52

See 1125

Cont. of W2393 (2390 - 2393) = Mtℓ − W2384.
Test for switchability in mating line. All showed ♂ for Mtℓ + SR compared to SR lac+, but W2393 showed ♂ showing highest (in × P2) (abt 10%).

2/7: W2393 × W2344 on EMB Mtℓ seq, lac sm. [Cross all day.]

(1) Ratio of ~~lac~~ Mtℓ+ to lac+: Pick SR+ from lac sm to EMB Mtℓ.
2/9. Found: 2 Mtℓ+ : 67 Mtℓ−.

(2). $V_1^R/^S$ ratio in lac+ SR of W1895 × W1177. Test colonies smith on lac T1. Classify as +S (A.) −R; +S .... (C); and +R .... (B).

1	A			31	A
2		A ⅓ B (lac+ w. $<^S_R$; $-^?_R$		32	B
3	B				C
4	A				A
5	B				C
6	A				B
7	B				C
8	A				C
9	A				A
10	C			40	C
11	B				B
12	C				B
13	C				A
14	B				B
15	B				B
16	B				A
17	B				C
18	A				B
19	A				?
20	C			50	C
21	A			51	A
22	B			52	A
23	A			53	C.
24	C				
25	A	17A + 1 A:B	} 32 V_1^S		
26	C	14C			
27	B				
28	B				
29	—				
30	B	19B + 1A·B	20 V_1^R.		

But note: incidence of B suggests that most zygotes had already segregated, thus showing bias.

2/3/54.

(A) 8:45

2384 × P2 1:1:2 3PM— Dilute ca 4PM,
~~chloramphenicol~~ Aerostain (G.D.) Also Plate 4:20:
EMB lac + Strs. i.c., ca 80 minute cross (dense mix.)

Gummy contaminant present (probably from W2384 broth and slant!) but ignore.

SR+ ca. 1%.

Pick Lac V (A: simple sector B: others ± lac++) to EMB's Mal.
(Most had overt Gal+ lac+ type ~~colonies~~) → of ~~7A, 2B~~ 7B, 2A, all but 1 had Mal+ (as suspected from colony appearance).

(B) W1177 × W2344. ca ~~8~~ hours in broth. Plate EMB lac+ Strs.
Pick SR+ directly vs/T1 on EMB lac. Each of 29 lac+: V, R.
Review of V, R = orthotypy, cross should perhaps be reversed in re the V, R used. (Cf. 110755.... carrying $V_{1(2344)}^{R}$ F^-.

(C). Note W2384 stock culture contaminated with gummy ~~phage~~ lac+, possibly also phage. Recheck single colonies for agency.

(D). Of W2384 MM−, 1-7, all but #1 appear λ^S. #2 = λ^R?
Variable intensity of response to λ.

1-cells : W2393 × W2344.

1124

2/7/54.

see 1125A for cross. Dilute at 1 hour.
Test isolates for MH, Mal, Lac, TI, S, Gal. Streptomycin Gal.

		Mal	MH	Gal	T/n.g.	Lac	S	Gal Strep.
B	1	+	+	−	+	S	−	
	2	+	+	−	+	S	−	
	3	+	+	−	+	S	−	
D	1	−	−	+	−	R	+	
	2	−	−	+	−	R	+	
	3	+	+	−	+	S	−	
	4	+	+	−	+	S	−	
E	1	−	−	+	−	R	+	
	2	+	+	−	+	S	−	
F	1	+	+	−	+	S	−	
	2	+	+	·	(+)	?	−	

all parentals!

8P2
3P1

V_1 segregation in Hfr crosses.

2/6/54.

Recap. 1. In Hfr W2344 $lac^+ S^S V_1^R$ × line 28 $lac^- S^R$..., almost precisely 50% of the $lac^+ S^R$ recombinants are V_1^R. (1117B).

2. In line 1, W1895 × W1177, data are rather heterogeneous, but sum is 41 S : 74 R. Should be recapitulated!

3. In W2057 × W2338 (line 1 Hfr $Lac^+ V_1^R S^S$...), in one experiment only, found 1 S : 9 R.

4. 1123B ~~W2057 × W~~ W1177 × W2344 : No V_1^S recombinants. [see 1122]

2/7. A. W2393 × W2344 11:35 AM — PM. Plate on EMB lac sm and EMB Mtl sm. Test SR+ for lac (Mtl) and V_1 segregation.
1:1:5

[B. W1895 × W1177 ". see 1122.] .

A). Mtl+ S^R picked at 24 hours. Cf. isolated Mtl+ and original mass

	lac		Isol	Mass	
			~~Mass~~	Isol	V_1
1	−	S	✓	✓	includes lac+ papillae
2	−	S	✓	✓	" " "
3	+	S	✓	✓	" lac− V_1^R
4	−	S	✓	✓	" lac+ V_1^S papillae
5	−	S	✓	✓	pure − S
6	+	R	✓	✓	includes, lac? S
7	−	R	✓	✓	pure: same background of S.

lac+ (4 colonies each) is:

1	~~lac~~ Mtl − V_1^S
2	Mtl − V_1^R
4	Mtl − V_1^S
10	Mtl − V_1^R

(over)

	lac	V_1	
∴	−	S	8
	−	R	3
	+	S	1
	+	R	3
			15

Expos. that some of these are Mtl+ recessions should be studied?

Also isolate the lac+ components: are they also Mtl+?

see 1129

8	−	S	✓
9	−	S	✓
10	−	R	no − S sm; lac+ pap.
11	−	S	✓
12	+	R	✓
13	+	S	✓
14	−	R	✓
15	−	R	✓

∴ $V_1^R = 6/15$

$Lac^+ = 4/15$

Exp. $+R = \frac{24}{225} \doteq 1.5/15$

1 ? (over) ~~but~~ but actually +R > +S!

AA: Ross. [Mal- lact+]

- S: 3

+ R 1

(+S-R) 1

- R 1

+S+S 1

1? S 1

Withhold conclusions re ratios.

Mal+lac⁻ S^R and Mal⁺ lac⁻ S^R

may occur in same colony!

Xyl – Lac segregations 1125CD

C. W2394 x P2 D. W2397 x P2 in SR+. See 1129.

D showed many more Xyl+ SR / lac+ SR than did C.

a: Test lac+ SR by direct streak on Xyl.

b. Do. Xyl/lac.

have lac+

b) C : 1/1 lac+ D 4/8 Lac+

a) C: 3/28 Xyl+ D 1/25 Xyl+. Not conclusive as to difference.

25E 2/12/54. Cross 2394, [redacted] x P2 on EMB lac, Xyl sm.

No SR+ formed on Xyl. ~~See 1129 for repeat.~~

Numerous on lac. . (= 1126 cross)