DATE: Apr 27, 1954 REF:

Control platings 2 and 3 parents.

young cultures
1 ml each per 10 ml, 2:10 PM.
Plate at 4:50 PM. EMB lac

A. W2344 + W1177

B. " + " + W1394
$Lac^+ Gal^- S^S$ Mal^+ Hfr; $Lac^- Mal^- S^R$; $Lac^+ Mal^+ S^R F^-$

(better to use 1895; 2344 being Gal −!)

C. W2344 + W2401 (for $Mot^+ S^R$ selections) — Inoc motility ± sm.

4/28/54. Young cells. 1:30 PM —

D W1895 × W1177

E. " × " × W1394. X too smeared.

A28: (B) Pick sectored colonies. Test Mal/S. $Mal^+ S^R$ indicates F^-/F^-

14/11 plates × 150 colonies scoreable. $Mal^+ S^S$ " Hfr/F^-

But $Gal^- Lp^s$ character of Hfr parent may prejudice results. However at this time, $Mal^- S^R$ = F^- parent.

the Hfr was probably not overtly distinct from W1394. (judging from plate + sm)

Est 3-4% recomb in A/sm

A29: 5 only had Mal^+. 6 were pure Mal^-. 3 clearly $Mal^+ S^S/Mal^- S^R$.
of remaining 2, one may be pure $Mal^+ S^R$, other is $Mal^+ S^R/Mal^- S^R$.
(#) (B)

Restreak to characterize components.

1152 B1-2: checked by DGG next page

1152

DATE: REF:

	1	2	3	4	5	6	7	8	9	10

B1 and B2 streaked out from spots on B mal.
B1 → mal + and mal − ; six of each picked
B2 → 2 types mal +, one dark, one light

	ed. picked as	mal	gal	lac	mtl	S all mated	V5
10	B1 + 1	+	+	+	+		S
	2	+	+	+	+		S
	3	+	+	+	+		S
	4	+	+	+	+		S
	5	+	+	+	+		S
	6	+	+	+	+		S
	B1 − 1	−	−	−	−		R
	2	−	−	−	−		R
	3	−	sl	+	−		R
	4	−	sl	+	−		R
20	5	−	−	−	−		R
	6	−	sl	+	−		R
	B2+ (dark) 1	+	+	+	+		S
	2	+	+	+	+		S
	3	(+)	(+)	+	+		R
	4	+	+	+	+		S
	5	+	+	+	+		S
	6	+	+	+	+		R
30	B2+ (light) 1	(+)	(+)	+	+		R
	2	(+)	(+)	+	+		R
	3	(+)	(+)	+	+		R
	4	(+)	(+)	+	+		R
	5	(+)	(+)	+	+		R
	6	(+)	(+)	+	+		R

40 (+) = light +

Observed 1152 B1-2-3

Try W895 lac − + W1177 + ~~W1394~~ Y10
Hfr Lac − SR lac − SR lac + S^S

∴ B1 = pure S^R.
Mal⁻ : W1177 plus recomb!
Mal⁺ : W1394

B2 - pure S^R also but pure W1394, does not contain W1177. Might be 2344 x W1394!

∴ Either a 3-coincidence or Hfr parent potentiates crossing after!!

Lp^s experiments

4/27/54 ff Has seven irradiated plates of W478 on Bgal, E (M−) suspected sensitives marked.

(1) Picked possible sensitives, streaked on E O; spotted in order on complete → replicated to D(milk)

→ 31 Lp^s ; 2 auxotrophic; all salt+

(2) Several plates replicated directly to D(milk) to pick up non-Lp^s auxos

(5 plates, ca 150 col/plate)

→ 3 possible distinct auxos.

478-4
Both sensitive & resistant on parents
original col
auxotrophic
Trypt − W2475

478-5
Sensitive
4 auxotrophic
Trypt − W2476

478-1
Grew in D(milk)
discarded

478-2
resistant
AA2−

478-3
resistant
Trypt −

5/6/54 ff Started E irradiated plates of W478 - before
16 Lp^s obtained; nutrition checked by E ML

5/11/54 ff 478 UV(8). Procedure as before. 29 Lp^s obtained; nutrition checked by E ML

Tu notes

1152A

DATE: May 8, 1954 REF: 1152

1152 B suggested repeat design. W1177 x W1941 supersuc of Hfr (W2364)

1. Y10 x W1177 x W1941
   F- F- Hfr
   lac+ S^S lac- S^R lac- S^S
   Grow together 1:1:10 12 30 PM - 5 PM.
2. Y10 x W1177 (both F-)
3. W1941 x W1177 (both lac-) Then streak out on EMB lac ind. or plate
4. W1941 x Y10. (both S^S)

5/10:

1. 5 plates (> 200 each) all lac+
2. 1 all -
3. 1 all -
4. 1. N. G. -

Parents checked for lac, S: o/k

Hfr x F⁻ (xF⁻) & motile 1153.

DATE: April 28, 1954. REF:

Desiderata for zygote isolations:
① 10² for both parent motile
② 3 component mixtures as technical control.

10³⁰ - 5³⁰ MX Hfr.

A W2341 mot (TCN) x W1177
B " " x W1394 [128] 40s. cell isol.
C " x 2401. or ?

8:30 A 30. Dilute ca 1:50 to prepare for isolations.
Hfr. 10:30

~~D Freshcultures~~
~~W2401 x W2344/Mot (xF⁻) 12³⁰ - 4³⁰~~
M1 OK. on plating (i.e. test of s.c. of 2344 mot,)

5/1/54 D. 9A1 Older cultures as inoculum. — N.1. Hfr. [to 3:25. Delete due to setup: 4:10].
W2401 x W2344/Mot 1:1:20

[Should study rate of crossing at low cell density & motile and immotile parents, perhaps in viscous medium.]
Could be done in a competition experiment! Cf. W1895, W2334 mot + immot.
No — need some lac-linked marker (V₆, V₁ ?)].

1153 B ([128]). Random single isolates. 40 placed. F1-G1-H1 flowed together. No lac++ (as indicative of lac+ Gal+ recombinants).
5 N.G. (A3,4; B2; D2; G5) agree with drops.
Described as actively motile at n=2⁵: C1, C5, D3A D5, These and G2, H4 appear to be v. slower lac+, others lac⁻. Have DC G streak out on EMB lac, M+L; repl to lac sus., Mal

DATE:

REF:

		1	2	3	4	5	6	7	8	9	10
		Anderson isolates	Lac	SM	Mal						
A	1										
	2			thus +.							
	5										
B	1										
	3										
	4										
	5										
C	1		±	S	+						
	2										
	3										
	4										
	5		±	S	+						
D	1										
	3		±	S	+						
	2B		±	S	+						
	4										
	5		±	S	+						
E	1										
	2										
	3										
	4										
	5										
F	1										
	2										
	3										
	4										
	5		±	S	+						
G	2		±	S	+						
	3										
	4										
	5										
H	1										
	2										
	3										
	4		±	S	+						
	5										

no recombinations evident (strong lac+) (vs W1394). pos.

DATE: REF:

		1	Droplets	3	Scores	5	6	7	8	9	10
11530		129			Lac	Mal	Gal	SM	MM.		
1	A	1 pair !	28		−	−	+	R	−		
2	B	1 pair !	28		− +	−	+	R (lac+, −)	−		
3	E	2	28		−	−	+	R	−		
4		3 mut ?	28		+ −	−	+	R	−		
		4	1		+	+	−	S	+		
	F	1 clump	28		−	−	+	R	−		
		2	1		+	+	−	S	+		
		3	1		+	+	−	S	+		
		4	1		+	+	−	S	+		
		5	1		+	+	−	S	+		
5	G	1 pair !	1		+	+	−	S	+		
6		2	28		−	−	+	R	−		
	A	2	1		+	+	−	S	+		
		5	1		+	+	−	S	+		

no positive pairs

∴ definite correlation of pairs with recombination. Note disappearance of the Hfr cell!

∴ 4 pairs have given 2 zygotes! B, E the Hfr parent disappeared, but gave Z. A1: no recomb. detected so far. G: both survived, no Zyg. These pairs are almost certainly significant: hunt them further. Save 1153D-1-6 for later complete survey of markers.

E. coli : motility observations and isolations.
and other notes on strains

DATE: REF:

4/28. E. coli C - 4th passage (see 1158) gave actively motile culture, still coccoid. Continue passage at R.T. (1157B4)
(This culture started from micro-cool of motile cell → semimotile clone. Then to agar ō gelatin at 30°, then to motility agar at 37°.)

cf. W2401: No motile cells seen at all. No motility in gelatin or agar at 3 days at 37, 30, R.T., or 20°. Reincubate. [do. 52C sent].
W1895 - sluggish, occ. motile. [Are unselected motile cells F⁻?]
W2284 - [Hfr mut → F⁻]. Highly motile.

Select by pas[redacted]
motility tubes. W...
ō Hfr strains, isolate ...
each stage; F⁻ strain ...

1st passage W1895, W ... isolates
still Hfr (reduced? ...) are
not. DCG is declining ...

May 2 ... E. col...
a) ō gelatin 30°
b) ō gelatin 37.
others rem...

Did cross, 5/8
1895: more [illegible] ō 1817 than ō 1177. All [illegible] lac +.
2344: about equal no. of prototrophs ō 1177 & 1817; also all lac +

Streaked out 1895 M2 & 2344 M2. Picked single col. of each & M2B repeated cross, using fresh tester cultures. Made slants of the 1895 & 2344 cultures used. Results of these crosses:

	[illegible]d	1177	1817
1895	0	1 lac-	65+, 7-
2344	1-	0	11+, 11-

5/23. Note: in recent weeks

♂ = W2344/M1 Hfr ♀ = W2401 F⁻
ung 1. ung 28A

Insertion of λ vectors

W2401 proves λ^R λ_2^R.

cf. W2284 = λ_2^S.

conclusion: Mal⁻ as before? Streak out overgrowth of W2284/λ_2.

DATE: MAY 3, 1954 REF:

Have united ufrag. cross as 1155., the first dilution left at room temperature ca. 11:30AM - 5PM 5/3/54, as 1155 A.

9:05AM Make up control ($F^- \times F^+$) W2331M1 × W2401 1:1:20

B. (Penicomy); B1 (NB) (overn.) (36 h.) MAY 4 1954

132 + buterexp. n.g., throw out

C W2344M1 × W2401 in NB. 1:1:20 9AM 37°.

1155 A has grown to ca 5×10^7 (density) by this point. Redilute for set up = A2. Also fresh dilution from 1155 = A3
R.T. 1:100 for 37°

See next page.

Most pairs give obvs. mixed clones not worth picking. Some of these were however picked apart quite early. V/s.

-	A2-B5	+/-	(A2 not unicell)	No test
×	D1-DYD5	all Lac⁻	Presumably was not a pair.	
	D2-D3	±; +/-	(5 %)	⇒ -
	F1-F2	±; +-		⇒ ✓
	E3 F5 G5	±(-+)-		⇒ ✓
	G4 H5	- +-		

∴ every pair then separated evidenced zygote formation!

H corrected as associated pair

Could pairs come from fission of zygote?

Isolate couples Hfr x F⁻.

1155
A

DATE: May 3, 1954. REF: 1153D!

1	2	3	4	5	6	7	8	9	10

9:30 AM ~~Fresh~~ overnite cultures (not regrown)

W2344M1 x W2401. 1:1:10 9:30 – 11:30 37°. (Refr. for later stock)

A). dilute 1:100 for isolations. Keep this at room temperature.

[130] But most of these pairs give mixed "clones". Cf.

	1153D! 1-4PM	8PM	10AM	Pick	1155A	1 = pair of motile – 1; non mot – 2° Lac	Lacsm	Mal	MH	Gal
10 A 1			n.g.							
2	P	+ –		• A2	A2	± –	R – (rare R+)	+ –	+	– +
3	P	+ ; –	✓ i o		B5	–	R	–	–	+
4	28 fogged									
5	P	– , +	✓		1 D1	–	R	–	–	+
B 1	P	++			D2	+	S	+	+	–
2	P	+ , –	✓		2 D3	+ –	R +, –	–	–	+
3	⊖ ⊖ P	~ ++	0 ✓		D4	–	R	–	–	+
4	A3m	++	✓		D5	–	R	–	–	+
25	A2 28.	28 •	28	• B5						
C 1					3 E3	±	S	+	+	–
2	P →	++	✓		6 F1	±	S	+	+	–
3	2m?	++	✓		7 F2	+ –	R+ –	–	–	+
4	28. E2	o	✓ o o		4 F5	– +	R –	–	–	+
5										
D 1	P? Kedas	– •	✓	• D1	8 G4	–	R	–	–	+
2	P	+	✓	• 2	5 G5	–	R	–	–	+
3	D2	– •	✓	• 3	9 H5	+ –	R+ –	–	–	+
4	D1	– •	✓	• 4						
30	D1	– •	✓	• 5						
E 1	P	+ –	✓							
2	P 2 28	+ –	✓							
3	P 2 28	+ ;	✓	• E3						
4	P		+ –							
5	P → 2+	fu ; +	0 , +	⊗						
F 1	P m	++	✓	• F1						
2	28	– –	✓	• F2						
3	3m	++	✓							
4	E1	+ –	✓							
45		–	✓	• F5						
G 1	P E3	++ ~	✓							
2		++	✓							
3	P 2+	++ , + –	✓							
4	P?	– •		• G4						
5	E3	– •		• G5						
H 1	X P 2+									
2		– +	✓							
3	E4	– +	✓							
4	–									
50	G4	–	✓	• H5						

On restreak, D2, E3, F1 are pure Lac+ (Gal⁻); 5, 8 are pure Lac⁻; 2, 4, 7, 9 are Lac⁻ and Lac+ Gal+ (recombinants). 4 had + only in center spot.
Save initial mixtures in stabs for later analyses.

Did any pairs here give pure 28? D1 G4. Not certainly pairs from records.
Total P isolated: 19.

✓ " samples of pure

Zygotes

DATE: May 5, 1954 REF:

Parent cultures overnight. W2344M1 × W2401 Mix 1:1:20 in Penassay 10:15 AM 37°. plate MAY 6 1954

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	Growth pattern	EMBlac A7	S	Mtl; Mal	Gal	CLASS	
A1	1	+		+	−	♂	
2	0 (28)	·			·		
3	1	+		+	−	♂	
4	0 28	·			·		
5	28	−	R−	−	+	♀	
B1	1	+		+	−	♂	
2	28	(+)	R also R	−	+	●	
3	0 28	·			·		
4	1	+		+	−	♂	
5	28	++ −	R −+	(−+)	+	●	Mal± sic! Mtl −
C1 −I3	28	++ −	R −+	−	+	● ◦ ♀	
2	1	+		+	−	♂	
3	28	++ −	R −+	R −	+	● ◦ ♀	
4	28	−	R−	R −	+	♀	
~~5~~	~~28~~	~~+~~	~~Probable contamination~~		+	~~♀~~ ♀ mix	
D1 −I2	28	− ++	R −+	R −	+	● ◦ ♀	
2	1	+		+	−	♂	
3	0 28	·			·		
4	0 1	·			·		
5	0 1	·			·		
E1	0 28	·					
2	1	+		+	−	♂	
3	1	+		+	−	♂	
(4)	28	−	R−	−	+	♀	V_1^s
5	1	+		+	−	♂	
F1	0 1	·		·			
2	0 28	·		·			
(3)	28	− ++	R −+	−	+	● ◦ ♀	
4	28 · 1 d	·		·			
5 died	1	+		+	−	♂	
G1	0 28	·		·			
2	1	+		+	−	♂	
3	1	+		+	−	♂	
4	0	·		·			
(5)	28	++ −	R −+	−	+	● ♀	
H1	0	·		·			
2	28	−	R −	−	+	♀	
3	28	−	R −	−	+	♀	
4	0 28	·		·			
5	1	+		+	−	♂	
I 1 − H3	1	+		+	−	♂	
2 − D1	1	+	thus S.	+	=	♂	
3 + C1	1	+		+	=	♂	

B2 thus lacks ♂ but appeared odd at first.

~~B5 may have ♂~~

C1, D1, F3, F5 lac⁺ S^R

E4 only pair not yet showing odd recomb.

B5 has ♀, 2 recomb

B2 ♀, recomb

DATE: REF:

1	2	3	4	5	6	7	8	9	10

B2, B5. — both nonmotile in direct test.
DCG also tested C1 D1 E4 F3 G5, nonswarm
& C5, swarm (as expected).

E4 }
B5 } pure Gal+ by streak. E4: V_1^S

B2 originally showed weak Lac+ reaction and some Lac+ S^R.
It appears now pure Gal+ by first streak, but some Lac+ S^R indicated.
Maybe simply Lac+ S^R recombinants in low numbers with ♀ excess.

B5: shows Lac– S^R (♀); Lac+ S^R but also some late Mal+ (♂?). Not evident on early streaking.

E4 though from a pair is not obvious recombinant. (✓ arabinose).

Check single colonies:

				Lac	Mal	Xyl	
B2	Lac+	①	Gal₁ S	+	–	+	Xyl+ Lac+ recombinant
		2		+	–	+	
		3		+	–	+	
		4	all +R	+	–	+	all Mtl–
	Lac–	1		–	–	–	♀ parent
		②		–	–	–	
		3		–	–	–	
		4		–	–	–	
B5	Mal+	①		+	+	+	Xyl+ Lac+ Mal+ recombinant
		2		+	+	+	
		3	all +R	+	+	+	
		4		+	+	+	
	–	1		+	–	–	Xyl– Lac+ Mal– recombinant
		②		+	–	–	
		③		–	–	–	♀ parent.
		4		+	–	–	all Mtl–

all carry the orthotypic markers Gal+ S^R and [illegible] Mtl–.

10/6-7/54. Also found, in B5, a Mal+ Lac– recombinant recover

1156 B5 -- 4 = Mal+ Lac- MH- Gal+ Ara-
SR Xyl+

---

all S^R MH- Ara- Gal+

∴

Lac+ { Mal+ Xyl+

Lac- { Mal- Xyl-

Fresh Zygotes

1157

DATE: May 6, 1954 REF:

W2401, W2344M1 grown overnight.
Regrow 1:10 90⁵ - 100⁵ #4. Dilute & place in separate drops for fresh isolations 135

Early mixture.
Pairs not well defined

A 1-2-5 proved all motile
A 3-4 ditto

B1 ∞ ?

B3 ∞

C1 ∞

C4 ∞

	#	Proved
A1	1	♂
2	2	♂
5	⊙	
3	3	♂
4	4	♂
B1	11	pure ♀
3	5	pure ♀
C1	12	pure ♀
4	13	pure ♀

∴ These pairs ~~are~~ are all ~~illegitimate~~ illegitimate. Were recorded as indecisive.

Hfr x W1177. ~~#58~~ 1157.

Conjugal pairs.

DATE: MAY 8 1954 REF:

W2344M1 x W1177 fresh cultures. Mix 2PM. Redilute and (not) not? examine 3PM. (1 hour mixture)

dao [135]

	Lac	S	Gal	Mal	
1	+		–	+	
2	+		–	+	
3	+		–	+	
4	+		–	+	
5	–	R	++	–	
~~10~~	–	R	++	–	
12	–	R	++	–	
13	–	R	++	–	
14	+	D2	–	+	♂
15	+	D5	–	+	♂
21	+		–	+	♂
22	–	R	±	–	♀
23	+	E4	–	+	♂ other L.
24	+	E5	–	+	♂
25	–	R	±	–	♀
31	+		–	+	♂
32	–	R	±	–	♀
33	–	R	±	–	♀
34	+	H2	–	+	♂

1157 (margin, beside 1–5)

E1, E2, E5, ~~E5~~ (margin, beside 21–25); G1, 2, 3 (margin, beside 31–33)

No recombinants here. all pairs illegitimate, ill defined association. Ф

2 surviving pairs are legitimate. Note lethality of remainder (see protocols). Of 5 other pairs, 1 had no survivor, 3 had ♂ only; ~~1♀~~ 1 ♀ only.

None ++ others S.

Gal ++ = 2401
Gal ± = 1177
Gal – = 2344

Details:

E1: → E2 (less not) → ♀; ↓ E1, E5 ♂, ♀ !

Assoc. ca 1 hour! but no recomb detected.

♂ → G1, more not ♂; ↓ G2, G3. ♀ ♀.

Ф

Assoc ca 15 m. but no recomb detected.

~~These should be saved~~!

= 1158 ~~21–25~~ ~~31–33~~

Hfr x W2401 1158

DATE: MAY 10 P.M. MAY 10 1954 REF: 136

Fresh cultures X: 2:15-3:40 PM.

			Cell Type	EMBlac	Gal	SM	Mal	Mtl
A	1	1hour, mating stop.	1	+		S		
	2		28			R		
	3	plate 01 71 hours.	28 1	−		R		
	4		−		as SM		as SM	as SM
	5		0 28	0				
* B	1	1hour together	28	− ++	+	R	−	−
	2		1			S		
	3	stop	1	+ + −		S		
	4		28			R		
	5		−					
C	1	stop	1	+ −		S		
	2		28			R		
	3	stop	1 v.	+		S		
	4		0 28	0 −				
	5		28			R		
D	1	> 1hour	1, Q 1	+ −		S		
	2		28			R		
	3	stop	0 1	0				
	4		0 28	0				
	5	PAIRED & IR	0 28	0 +				
E	1	stop	1	+ −		S		
	2		28			R		
	3	stop	1 d	+		S		
	4		0 28	0 −				
	5		28			R		
F	1	stop	28	−		R		
	2		1	− +	−	S	11	11
	3	std	1	+ −		S		
	4		28			R		
	5		28			R		
G	1	stop	28 0	0	u			
	2		01	0 −				
	3	stop	28	0		R		
	4		0 28					
	5		0 28					
* H	1	1hour	0 1	0				
	2		28	− ++	+	R	−	−
	3	stop	d					
	4		28	−		R		
	5		−					

MAY 11 A.M. MAY 12 A.M.

stop = stop in pipette

Note: most pairs were s.p.p. of 10 pairs, both survived in 7, both lost in 2, 28 only lost in 1; 1 only lost in one of other pairs

(see over)

1 was "separated by surface tension" (drying): both lost.

of the 5 persistent pairs, 4 were "separated" spontaneously

B1-2 → ♂ ✓ ♀ and ⊗ ✓

H1-2 → ♂ died ♀ " ✓

don't count D4.

A3-5 A' (died) A3 → ♀ only.

1: ♂ = W2344M1

28-♀ = W2401.

D1-2-5 ♂1 → 1 (↓ ♂) ● (↓ ♀) ⊗1 (↓ both died)

← count as "viable pair"

A1-2

1 was manipulated at 1 hour

○− → ♀, ♂ only.

Save: A1-2, 3; B1-2; D1-5; H2; F1-2-5

F2 simply looked peculiar under microscope as if motile type 28 but pure bal⁻ lac⁺ indicated (♂) save for further comparison. Abandon ~~13~~ 147 as these show no particular heterogeneity (among survivors). (probably old)

F2 was plated out on EMB Lac.

Zygote isolations

MAY 8 1954

1156. 15 pairs isolated. 28 failed in 8. 6/7 of
131
remainder, solely from line 28 parent. 1 prob. ♂+♀+R.

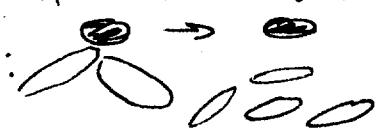
● pair, ═══ ♂ lysidat pipette.

1155 Most pairs not separated (19). ●/4 from l28 parent
130

53D ●/4 partly separated.
129

Total to date (W2344/11 × W2401): ●/16

Queries:

1. These are all fairly late pairs. Earlier should be studied to ensure that zygotes result from pairs, not conc[illegible], and to seek dikaryon cells such as 1156 B2. Notes should be consulted in event of doubt on purity of this cell. None indicated: 

2. Perhaps fuller pedigrees.

3. Review possibilities of line-1 crosses (cf 1158. Thel Fla⁻ 1177 or 1895!

4. Cytology.

5. Find Neurospora resume!

6. Problem of undetected recombinants: why not more?

1 hour fresh zygotes

MAY 11 1954

1159.

DATE: REF:

	1	2	3	4	5	6	7	8	9	10

A 12:30 - 1:45 cross ♂ x ♀ V. young cultures. Diluted ca. 2:50. 146

B. 10:30 - 11:30 " " " . Refr. — 3:10 ± " ... 145

B) — 3-3:40 PM Few or no pairs (Disjunction already?) after refrigeration X.) use 145 for ♂ unless indicated

all 146 stabbed

146	paired 145	Comment	CELLTYPE 146	c.t. 145	lac	146 EMB lac	SM	Mal	Mtl	Gal	SM 146
A 1)	♀ - A5	S1P	0	♂	+			R			
2	♀		0								
3)	♀ - A4	SL. MANIP.	28	♂	+	— (+)		lac +			
4	♀ ♂	lysed?	28			—					
5	♀		28			—					—
B 1)	♀ - A3	Long conversation	0	♂	+						
2	- C1		0	♂d	0						
2 3 }	♀ A1-2		28		d	—					—
4	♀		28			—					—
5	♂		1			±		S	+	+	[illegible] —
C 1)	♀ - C3; C5	[illegible]	28	♂	++	—					—
2	♀		28			++ (—)	+	lac +			—
3	several ♀ ♂ ♂	CLUMP	28 ..			— (+)	3	lac +			+
4	♀ ; C4	lysed?	0	♀	—						
5	♀		28			—					—
D 3 D)	- 5D5♂ NOT SEP	: 0	0		0						
2			0								
3	CLUMP NOT SEP'D - MIXED CLONE NOT PICKED									×	
4			—								
5	♀		28			++ —	4	lac +			—
E 1	♀ - E5	S1P	0	♂	+						
2											
3	- E4♂ NOT SEP'D → MIXED CLONE ♂ NOT PICKED										
4											
4 5			—								
F 1	♀ - F4		0d.	♂	+						
2			—								
3 }	♀ - F5		28	♂	0	—					—
4	♀ · ♀		28 0			—					—
5	♀		28			—					—
G 1 }	♀ - G5		d.	♂	+						
2	♀		28			—					—
5 3	♀ - G4		0	♂	+						
4	♀		28			—					—
5 }	♀		28			—					—
	♀		28			—					—
H 1	♂ H5	S1P	28	♂	+	—					—
3)	♂	3 h. intus	1			+		R S	+	+	—
4)	♀ dense	all OK	28	5S?♂ all OK. +		—		R			—

all others lac — (arrow down col. 8)

all others this

5/13

146 tests for all painted these sugars

DATE: May 12, 1954 REF:

A. Start to isolate numerous pairs, but this was interrupted. Rather late culture.
144 A1 – D3 are isolations. But only A1 – 5 were separated (A5 quite late). B1 – D3 give only viability data: from the untouched pairs, both ♂ and ♀ grew in 9; ♂ only in 3 and both died in 1. This suggests that mortality may be connected with separation but other differences are possible.

In A1-4 ♀ ex conjugant separated early (watch ALR!); A5 at 30+30 ada. Sequence in A1 is not clear from notes. (was it synkaryon?).

B. [143] Pick 5 pairs (rather clumsily) for pedigree analyses. 8 PM

A1: 1 - A ♂, B ♀ (1 cell) — B 1 cell

2A: — A2 ♀ (C♀), B2 ♂, A2, D2 (F2), C2 !, E2 — all grew.

3A: A ♀ (E 1 cell), C, BL ♂, BR ♂, C, D, G, F — all but E grew

4A: B ♀, A ♂, C ♀, D ♀, ! E4 (144 E1-4, F1-4) — B: 1 cell only

5A: — A ♂, B ♀ — both grew.

Chance to correlate growth delay c̄ zygotes.

Note fair survival of these, too.

DATE: MAY 14 1954 REF: 143 - 144

3 rotten 5/16.

143

			EMB Lac	Lac	Gal	Mal M4 Xyl	S
1	A	1	+	+	—	+ -	S
	B	0	·				
	C	0	·				
(2)	A	28	— +	— +	+	—	R —+
	B	11	+ ; +	+ +	— —	+ +	S S
	C	28	—	—	+	—	R
	D	28	—	—	+	—	R
	E	28	—	—	+	—	R
	F	28	—	—	+	—	
3	A	28	—	—	+	—	R
	B	11	+ +	+ +	— -	+ +	S S
	C	28	—	—	+	—	R
	D	28	—	—	+	—	R
	E	0	·				
	F	28	—	—	+	—	R
	G	0					
4	A	1	+	+	—	+	S
	B	0	·				
	C (144E)		— — — —	—	+	— — —	R
	D (144F)		— — — — —	—	+	— — —	R
	E	0					
	F	-					
5	A	1	+	+	—	+	S
	B	d.	·				

[illegible]

6/5/7 Lac+ component died out all parental are +'s here. Concordant.

DATE: MAY 14 1954 REF:

	1	2	3	4	5	6	7	8	9	10
	144	EMBlac		Lac	Mal Mtl	Xyl	Sal	S		
A. 1	·?	−		−	−		+	R		
2 a	1(28); [redacted]	+		− − +	− +		+ − +	R R^{Lac+-}		not separated
b	[redacted], 28	−								
3 a	1-28	+, −		+ −	+ −		+ +	R$^{Lac+/-}$ R^{Lac-}		
b	28	−								
4	28	−		−	−		+	R		
5	28	−		−	−		+	R		
E 5	28	−+		−+	−		+	R Lac+-		

E1-4F1-4 see 143

not clear zygotes.

all parentals Lac+

E5 from A1, but long contact!

A2-3 eff. unseparated

A4 clear test?

$B^- \rightarrow .. B^0_0$ + (1111) saved

no zygote.

Survival not clear.

A5 Long contact, still −! saved

9 A.M.

1160

MAY 14 A.M.

MAY 1954

9A14 1431

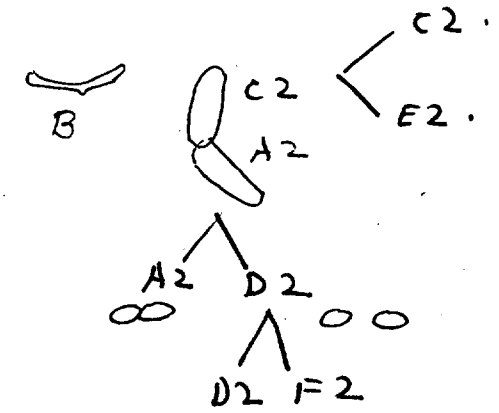
1. ♀ died B A ♂

2. A2 zygote ♀ save as 1160-2-1

B ♂

C D E F ♀

80 flapping



3 A - C - D - F ♀

E - G ♂

B ♂

4 A ♂

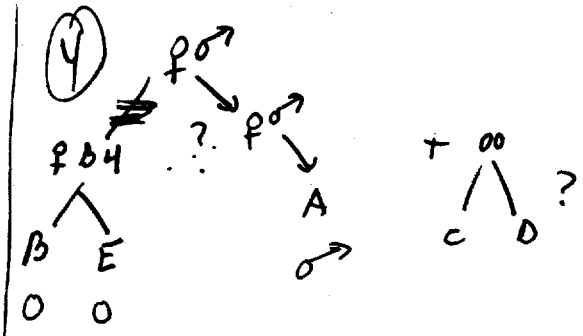
B E ♂

C D ♀

5 A ♂

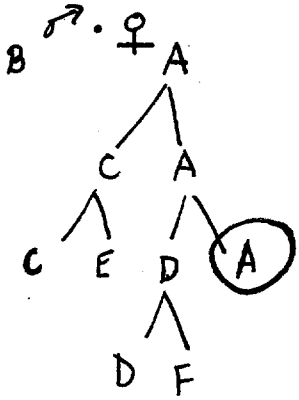
B d.

(4)

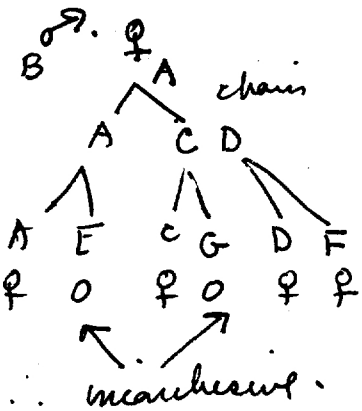


poss. inconclusive also.

(2)



(3)



∴ inconclusive.

116 1

DATE: May 13 1954. REF: 142 - 141

Overnight, x 10^50 - 11^55 37° 1:1:10 Penassay. 142

A1
A2 picked ca 130.

Fresh cross x 12^30 - 3 PM s/c.

Not separated: ~~Reg~~ Refrigerated overnight + separations to 142, 141, A14.

		EMB lac	Gal	Mal	Mtl	Xyl	S
141	A4	−	+	−			S
	A5	−	+	−			R
	C1	+	−	+			R
	3	−	+	−	✓	✓	S
	4	++ −	+	−			R
	5	+	−	+			R lac+/−
	D1	+	−	+			S
	2	−	+	−			S
	4	−	+	−			R
	5	−	+	−			
		−	+	−			
	E4	−	+	−			
	5						
	G1	− [scribbled]	+	−	✓	✓	
	H1	− [scribbled]	+	−			
	2	−	+	−			
	3	−	+	−			
	4	−	+	−			
	5	−	+	−			

all parentals Eaccordant.

1161 is a Mal+ yielder not showing a Mal+ Lac− recomb but exhibiting a Mal− Lac+ recomb. also

~~142 G1 ++ − (Pair of ∅) R (lac?!) Mal Mtl Xyl~~
~~2 + S~~
~~3 + S~~
~~5 + S~~
H4 0
5 − R

	Lac	SM	Mal	Mtl	Xyl	Gal
G1	++ −	R (lac−)	(−+)	−	(−+)	+
2	+	S	+	+	+	−
3	+	S	+	+	+	−
5	+	S	+	+	+	−
H5	−	R	−	−	−	+

Gal among Mal or Xyl needs to be verified.

Re − 10/54. Pure Gal+.

Found at first: A (Smr)

V^S Ara − { Lac − A MtI − S^R Mal − Xyl
Lac + < C MtI + S^S + +
D MtI − S^S − −

No Lac+ S^R, Mal+ S^R (Reexamine for ① Lac+ S^R No ② Lac − Mal + No (440 many colonies spot tested)

DATE: May 14, 1957. REF: 137

	1	2	3	4	5	6	7	8	9	10

		EMBlac		
A	1	0	♂ gd.	
	2	+	♂, ♀d	
	3	+	♂	
	4	±	♂ ♀ d	
B	1	−	N. S. ♂ d.	
	3	+	♀ d.	
D	3	−	♀	
E	5	+	♀ d.	
H	5	++ −		

(G5 ♂) not picked.

X 1:1:20 (fresh) 12:30 - 2:10 (set up c 2:40).
Trapping technique, worked v. well but pipette may be too narrow.

Three viable pairs only: A3-D3 ♂ · ♀ no ⊗
(G5)-H5 ♂ · ⚥ ⊗
B1 (n.s.) ♀ (susp. appearance - restreak c EMBlac.

See protocols for other isolates. V. Poor Viability !!

Raw incidence = 1/3 pairs zygotic

		Lac	Xyl-	MH-	Mal-	S	Gal	
A	1	0						
	2	+	+			S	−	♂
	3	+	+			S	−	♂
	4	+	+			S	−	♂
B	1	−	−			R	+	♀
	3	+	+				−	♂
D	3	−	−			R	±	♀
E	5	+	+			S	−	♂
H	5	++ −	−			R	+	⚥

all concordant

Paris. Typ clas transmigration trap Salmonella. 1163.

DATE: May 19, 1954. REF: 150.

(1) Hfr culture (W2346/M1) seems sluggish. Resolute motile cells = A1-4. A1 n.g. (A3) probably best & be picked

From this serial #, use 6 x 8 coverglass mountings. Lettering in sequence A B C D E F G H.

(2) Set up parents in adjacent deeps: (♂) (♀) ○ next to trap deep, notice that ♂ have to swim through ♀ to reach trap. May have worked moderately well, but settling of ♀ suggests better to allow intimate mixture before trapping. Many very early pairs seem to separate very readily indeed, precluding easy isolation. But no systematic data!

(3) Conventional mixture 1:1:6 2¹⁰ – 3¹⁵ deobtains from ca. 3⁴⁰ – 4:40, assay trap deep

Comment: survival fairly good.

Note: both manip ♀♂ died but also 2 others. Would predict several zzz. but may be some post clone deaths not detected.

Pick sequence:

A 3	D 1
B 1	4
4	5
5	6
6	E 1
	E 2
C 1	3
2	
3	E 4
4	5
5	F 4
6	5
	G 4
	5

all clones to date have agreed & recorded type of cell unmated. Note will be made of any discrep.

B 1 2 3 4 5 6

C 1 2 3 4 5 6

D 1 2 3 4 5 6

E 1 2 3 4 5 6

F 1 2 3 4 5 6

G 1 2 3 4 5 6

H 1 2

please turn page

DATE: 5/20/54. 5/21/54 5/22 REF:

A) Stock	History	Diptype	3	EMB Lac	Lac	Gal	Mal Mtl	S	Lac	Save	10
B { 1	ss.	♂	.	+		—	+	S			
2	7 h.	●									
{ 4	s. doused	♂ ♂.	.	+,+		— —	✓ ++	s		.	
5	late pair	< ♀	.	—	1	+	+—	R	—	.	
6		♀	.	—		+	—	R	—	.	A_2^+
C { 1	SS	♂	.	+		—	+? +?	s		.	
2	2h.	♀ ♂	.	— +	(2) pure	+ —	—	R	—+	.	
3			.	+		—	+	s		.	
{ 4	s.i.d.	♂	.	+		—	+	s		.	
5		< ♀	.	—	3	+	—	R	—	.	
6		♀	.	—		+	—	R	—	.	
D { 1	late pair	♂	.	+		—	+	s			
2	Manip.	●									
{ 4	late pair	♂ ♂	.	+,+		— —	++	s		.	
5	s.s.	♀	.	—	(4)	+	✓ —	R	—	.	
6	2h.	♀		— ++		+	—	R	—+	.	
E { 1	oil bubble	♂	.	+		—	+	s		.	
2		< ♀	.	—	(5)	+	—	R	—	.	
3		♀	.	—		+	—	R	—	.	
{ 4	s.s.	♂	.	+		—	+	s		.	
5		♀	.	—	6	+	—	R	—	.	
F { 4	72h.	♂	.	+		—	+	s			
5		♀	.	●							
G { 4	2h.	♂ R.	.	+		—	+	s		.	
5		< ♀	.	— ++	(7)	+	✓ —	R	—+	.	
6		●									
G { 1	Manip 2-stuck tip	♂	.	+		—	+	s			
2		◎									
H { 1		◎									
2		◎									

all conc.

without second reading only 3/7

These C2 may be a dikaryon. Streaked out on EMB Lac, Gal. Both Gal⁺ and ⁻ are present.

a) spot some of these on plate for 1164 tests

b) check motility!

s.s. = sep. spont

s.i.d. = s.i.d. after pipette

(over)

record on C2 was:

3:54 f0

st. ca 5:15

6:25 $\frac{0^2}{3\ 1}$ presumably separated by slight manipulation.

No comment on behavior at isolation. No basis for question on the isolation - possibility of contamination at isolation is not inherently excluded.

In view of rarity of this event even now (and reduced incidence per zygote for fem pairs, the matter must be strenuously questioned.

No remark is noted of motile cells in the drop.

Why is proportion now so low if true? See next page →

Tests on C2: Streak out on EMB Gal. Gal+ > Gal− colonies.

Pick and stroke: 5 Gal+ 6 Gal−. all are Lac− Mtl− Mal− Xyl− S^R and are thus simply a { None motile in motility agar 10/54 }

Gal−/atheotype recombinant! Streaked on EMB Lac, no Lac+ are noted, but some colonies are pink (Lac− Gal−?). Where crowded with some pink "+" centers.

a) Check Hfr, Lp, mutation. b) Might be Lac+ Gal− Xyl− .... (marked by Gal+ −x ?)

c) Presumably not Lac+ and Hfr would give Lac+ Gal+ recombinants. d) Check motility, Mal+?

DATE: May 23, 1954. REF:

① Already identified as carrying ♀ wt Lac$^{-?}$ Gal^{-} M+l · Mal · Xyl^{-} S^{R}.
In initial spot and especially in first replica to Lac, definite + reaction was indicated, suggested possibility of modified Lac^{+} Gal^{-} Xyl^{-} ck.
Also doubtful possibility of presence of Mal^{0+}.

11 A23. Repick from original ~~spot~~ to stab and 1168C2

a) motility - trace in susp. from agar ~~C2~~, C2', ~~C2A~~ ; no in agar.
b) streak EMB Lac again
c) Mal, M+l
d) λ, x — Gal+ also plus check for Lp, Hfr
e) Compatibility: A24 × 1895M2 ; σ→ status, Lp₂ may interfere c former. nutrition. Should certainly be crossable c Y10 line.

			P25 D(10)	N26 S Lac
1 C2A	1895M2		0	all−
2	2206		+	
3 C2B	1895M2	+++	++++	+? −?
4	2206.		++	+? −

f. Nutrition: ~~both~~ M-H - orthotypy: C2A = Gal^{+} ; C2B = Gal^{-} colonies.

~~[illegible]~~ presumably Hfr. Brownish in Pervasiveness than A.

Y24 → Pure Mal^{-} M+l^{-} Lac^{-} c trace^{+} reaction in background. Reincubate.
Trace reaction comes from interaction of Gal^{+} Lac^{-} / Gal^{-} Lac$^{?}$ colonies = W2502
attempt Gal^{+} revisions of W2502

This recomb. could very readily be missed unnoticed! Ar^{-}.

Note exaggerated orthotypy of e2 cf. e3

segregation of motility, etc.
Note most pathogens motile! (~~[illegible]~~)

11 [redacted] ...um Te
vs. degree of motil

DATE:

	1 non-motil	2 poor-motil	3 high-motil
lac + gal + mal + motil +	1	6	15
+ + + −	0	1	1
+ + − −	0	1	4
− + ± +	0	0	3
− + − +	0	0	0
− + − −	0	0	1
− + + −	0	0	0
Total	1	8	24

No − this rose is

DATE: MAY 20 P.M. REF: 151.

	1	2	3	4	5	6	7	8	9	10
start	12^05 – 1^30		.1 ♂ : 1 ♀ : 10 Scotch.							
			MAY 21 A.M.							
					5/22		concordant			
		History	Cell Type		E.4B lac		Mal Mtl Xyl Gal		S (lac)	
A	1 .		♂		+	✓	– – –	+	S	
cf. standard	2 .	S1d	dense ducts ♀?		–	1 ✓	+ + +	–	R	–
	3 .		♀ dense kind.		–	✓	etc.			
A	4	SS	?		0					
	5 .		♀ Fr.		–	2 ✓				
	6 .		♀ Fr.		–	✓				
B	1 .	loose at first.	♂ v. active		+	✓				
	2 .		●			10				
	3 .	SS.	♀ (recovered)		– ++	✓			R	– +
	4 .		● ♂		+	✓				
(G3)(A6) ♀	5 .	v. loose then tight.	♀		–	3 ✓				
	6 .		♀		–	✓				
C	1	S1P	♂		+	✓				
	2		●		●					
	3		♀		–	✓				
	4 .	S1P.	♂		+	✓				
	5 .		♀		–	4 ✓				
	6 .		♀		++ –	✓			R	– +
D	1	long.	♂ ♂		+	✓				
	2 .		♀		–	5 ✓				
	3 .	sl. manip.	♀		– + (++)	? ✓	Same Mal etc + ? +		R	– +
D	4 .		♂		+	✓				
C3E3 ♀ ♂	5 .	Off first thus dumped thing	gh; Resort		– 7	✓				
	6 .		♀		–	✓				

Pick P21.

D3: no motility seen in direct sweep. None in agar.

D3: all Gal+ but Lac Mal +/–

Spot and colonies on EMB lac.

success

all Gal+ nonmotile

D3 includes
Mal+ Lac− M+ S^S } test of these!
Mal+ Lac+ M+ S^S

Mal− Lac+ M− S^R (1)

perhaps should be screened even more thoroughly.

Mal+ purified all S^S; some unpur. give Mal+S^R "comb" or mutants?

Recombs: (1.s.c.i) Sec 10/1/54

1	Lac− Mal−	H−M(+)	Ara−U1^S
2	Lac+ Mal−	H−M−	Ara+U1^R
3	Lac− Mal+	H−	Ara−U1^S
4	Lac+ Mal+	H−M−	Ara+U1^R

Sec 1183. In routine check, H− segregants found among 11643. Among isolates 1–4, #3 was H−M+ (others presumably H−M− as no growth on Maltose). To try to find other types @ streak growth of mixture through D(H) on EMB Mal, found ca 1% Mal− (2) streak on EMB Mal mm and test mass (all Mal−).

DATE: REF:

	1	2	3	4	5	6	7	8	9	10
			MAY 21 A.M.							
						concordant, no recombinants.				
			Cell Type	Lac		Gal	Mal NH4Yyl		S (lac)	
E	1	sl. manip.	♂ ♂	+ +		− −			S	
10	2		○							
	3		d ?							
	4		♂ ♂	+ +		− −	+ + +			
	5		♀	−	● 8	+	− − −		R	−
	6		d (green)	+						
F	1	lh.	D				etc.			
	2	ss	◉							
20										
	4		♂	+		−				
	5 :	lh.	♀	− 6		+				
	6 :	ss	♀ (F. Gbus.)	−		+				
G	1		♂ ♂	+		−				
	2	long.	● gh.							
	3 :		♀	−		+				
	4		♂	+		−				
30	5	lh.	d.							
	6 of F6.									
H	1		♂							
	2	ss rarely	● gh.							
	4 :		♂	+		−				
	5 :	ss.	♀	−	● 9	+				
	6		d.							

40

Note losses/scars during lethality. Try saturating oil & water.

Counting only complete pairs: only 2 = 1/6. ; add incomplete pairs: ~~1/5~~ 2/4

Complete: 1-2: Incomplete 7-10

2/6. 1/3 3. Total 3/9 only.

50

#8 is certainly not in lethality (fortuitous d. of E6)

& probably 9 also.

DATE: MAY 21 1954 REF: 152

X (old) . 1♂ 1.0♀ 5 ml broth 37° 9 – 10[30] A.M. Trap.
(B) Comparable W2331M2 Examined at same time: no pairs.
Try 2344M2 (F⁻). see 1166.

	1	2	3 (dep)	4 (Lac)	5 (Lac)	6 (Mal MH) A25	7 (Gal)	8 (S (Lac))	9	10
A	1) ×	manip.	○							
	2		○							
B4 –	3 ·		♀	– ++	✓	Mal, MH	+	R (–+)		
	4) ×	loose	♂	+	✓	Mal, MH	–			
	5		○							
	6 ·		♀	–	✓		+	R		
B	4 :	55 early + K3μ	♂ ♂	+ +	(1) ✓	unclear	– –			
	5 :		○ ♀	–	✓		+	R		
	6 : ·		♀ ♀	– + – +	✓		+ +	R R –+		
C	1	lost dep	♂	+	✓		–			
	2		[illegible]		(2) ✓					
	3 ·		♀	–	✓		+	R		
	4	sep: SS	♂ ♂	+ +	✓		– –			
	5		♀	–	(3) ✓		+	R		
	6 ·		♀ ♀	– –	✓		+ +	R R		
D	1 :	SS	♂ ♂	+ +	✓		– –			
	2 ·		♀ ♀	++ – ++	(4) ✓	[illegible] –*	+ +	R R –+	* same + ?	
	3 ·		♀	–	✓		+	R		
E	1 :	ol man.	♂	+ [illegible]	✓	+	–			
	2 ·		♀	–	(5) ✓	–	+			
	3 ·		♀	– ++	✓	–	+	–+		
	4 ×		♂	+	✓	+ MH	–			
	5	distorted	○.					no Mal		
F	1] ×	manip.	♂	+	✓	+ unc	–			
	2		○.							
	3 ·		♀	–	✓	–	±			
	4	SS ♂	○		6 ✓					
	5 :		♀	–	✓	=	+			
	6		♀	–			+			

A }
B } all lethal } manip.
D4 " " manip

D2B: Mal- and seeming mucoid Mal+/-
see over

D2A. Lac +/−, pure Mal −.
sense +, −

D2B. Lac +/− ; Mal − and Mal "v".

D2B1.

↳ 4 of these throughout. Prove to be mucoid, no distinct indication of segregation +/−.

3 Lac + muc. D2B2

1 Lac − muc. D2B3

	Mal	Lac	S
1	—	+	R
2	+	+	S
3	+	—	S

becoming less mucoid. ~~the~~ all auxotrophic.
not segregating.

D2A has ♀, Lac⁺ Mal⁻

D2B has ♀, Lac⁺ Mal⁻, Lac⁺ Mal⁺, Lac⁻ Mal⁺ all Gal⁺.

presumably failure of Mal elimination & crossingover / S/Mal.

of 1161 D3! In both cases, only cases which show Mal⁺, all 4 combinations are seen. Is Lac⁺Mal⁺ ♀ giving reciprocal recombinants?

A 75

5/23/54.

					EMBlac		lac	Mal	MHoz	Xyl.	Gal	S	(lac)
G	1	10	:	ss	♂	+	1	✓	+			−		
	2				♀	−		✓	−			+		
	3	30			♀	−		✓	−			+		
	4	11	:	ss	♂	+	8	✓	+			−		
	5	00			♀	−		✓	−			+		
	6	00			♀	−		✓	−			+ cs Mal		
H	1	1111	:	ss	♂	+	(9)	✓	+			−		
	2	0			♀	−		✓	−			+		
	3	0			♀	−++		✓	−			+	R	−+
	4	11			♂	+		✓	+			−		
	5	0	x	ss	0									
	6	0			0									
J	1	0	:	ss	♀	−		−	− −			+	R	
	2	0			♀	−		−	− −			+	R	

all drops concordant c̄ the morula.

9 pairs complete, 4 zygotic! Note segregation in B4 pedigree.

#4: zyg. ss; ss, sl.mer; ss

5: nonzyg (det.) lost drop; s.p; ss, ss; ss.

Competative pairing

W-2401, W-2344M1; W-1895M2
F- Hfr F-

DATE: May 22, 1954 REF: 153

Yesterday's observations suggest that motile F- (cf. also Salmonella) does not pair with W-2401. This can be properly confirmed only by a competitive pairing experiment. Suggest: menage a trois with Hfr mot; F- mot; F- ~~Hfr~~ non-mot. Pick pairs and diagnose. For simple diagnosis, it should not be essential to separate out the pairs, but would be useful if most of these, as expected, will be bisexual. Similar expt. possible with F+. also permitting "F-duction" test.

Cf. DCG notes and 1154. W-1895 used here is second passage motility, and second colony ~~from~~ reisolate showing F- behavior (with peculiar segregation ratios). (before reisolation).

Ovenight cultures, to 10 ml. Penassay, 37^o 9:25 AM

A. W-2344M1 (.1) + W-2401 (1)

B. do. + W-1895M2 (.1)

C. W-1895 M2 + W-2401.

Previously concerned about lp's of W2401 but EML finds it poss-s. to λ. (possibly Mal-lp_2^r?).
W2338, W2384 being rechecked.

11:40 - 12:40 Isolate pairs. Leave at R.T. Some probable motile - motile pairs seen also.

In controls, no pairing was seen in <u>C</u> as compared to A.
Control for B: streak out on EMB Lac, Test Lac+ on EMB Gal to verify ratios of Hfr/F⁻.

N23: A, B show SR+, not C. on Lac sm.
ca 1%

EMB Gal: Lac.

A ca. = +, - (sic! indicates growth diff.?) + almost = -.

B + > - + = -

C all +. + almost = -.

Sample Lac+ to EMB Gal. (Pick every clone + available to avoid bias on slight difference of appearance!) : 23 Gal+ (F-) : 7 Gal- (Hfr)

Competitive pairing.

153

A 25

			Inoc.	Dup A23	BLac	Lac	Mal	Mth	Gal	S (lac)	
A	1	♀	①	♀							
	2	♂	∞	♀	−	−	✓	−	+	R	
	3		40	♂							
	4		11	♂	+	+	✓	+	−		
	5		11	♂	+	+	✓	+	−		
	6		00	♀	−	−		−	+	R	
B	1	♂		♀	−	−	✓	−	+	R	
	2		50	♀	−++	−++	✓	−	+	R	−+
	3		1+	♂	+±	+±	✓	+ +	+	S	
	4		·	−							
	5		////	♂	+	+	✓	+	−		
	6		60	♀ ∞	0	0					
C	1	♂	////	♀	−++	−++	✓	−	+	R	−+
	2	♂	than 0	♂	+	+	✓	+	−		
	3		60	♀	−++	−++	✓	−	+	R	−+
	4		0	♀	+−	+−	✓	−?	+	R	−+
	5		1^10	♂	+	+	✓	+	−		
	6		0	♀	·	·					
D	1		////	♂	+	+	✓	+	−		
	2		0000	♀	·	·					
	3		//// ////	♂	+	+ 3	✓	+	−		
	4		00000000	♀	−	−	✓	−	+	R	
	5		//// ////	♂	+	+ 4	✓	+	−		
	6		0000	♀	−	−	✓	−	+	R	

SIC 1895

plate of ♂ pink already?

why are Ab ~~pink~~ grey? Recover later.

lac presumed Gal−

all rare exc. B3

4-m streak pure Mal−

		Gal	disp	Blac	Lac	Mal	Xyl	S(lac)	Cal	
E	1									
	2		fat mat							
	3			+	5	+	+		−	
	4			−		−	−	R	+	
	5			+	6	+	+		−	
	6			−		−	−	R	+	
F	1			+	7	+	+		−	
	2			−		−	−	R	+	
	3									
	4		mat!	+		+	+		−	
	5			+		+	+		−	
	6									
? G	1		(♂)?	+		+	+		−	V_1^R
	2		♂	(++)	9	++	+		+	V_1^S } parental
	3		♂	+	8	+	+		−	
	4		♀	−		−	−	R	+	
	5		−							
	6		−							
H	1									
	2		♂	+		+	+		−	
	3		♀	−		−	−	R	+	
	4									
	5		♂	++		++	+		+	
	6		♂	++		++	+		+	

corrected

∴ all pairs were either Hfr/F^-_{28} or (G1-2) and (H5-6) which carry ~~not~~ F- alone or ~~not~~ Hfr.

Following distribution noted: (complete pairs only):

pairs	singles
8 Hfr/F^-_{28}	7 Hfr_1
1 Hfr/F^-_1	~~1~~
0 F^-_1/F^-_{28}	23 F^-_1

Need replication 2/8 zygotic

Could we detect recombination between G1/G2? Only crosses are Gal+/V_1? Test /75.

DATE: May 25 1954 REF:

A. (Golden cultures) W2502 + W~~45~~ 1895 M2 (W2503)

1 : .1 ; 10 10:45 AM

B. W2332 + W2401 (F deuteron).

No pairs but possible mixups & inadvertent use of T2 both.

MAY 24 P.M.

9[15] Some EMB arabinose tests on Lac⁻ exconjugants.

1163 B5 B6 C5 C6 E2-~~B~~ E5 E3

1164 A2-3 A5-6 B5-6 C3 D5-6 F5-6 H5

152 1165 ~~A~~6 C3 C5-6ab F356 G23 G56 J1-2

153 1166 D4 D6 E4 E6 F2 G4 H3

all were Ara⁻ except ? (B6) 1163

Thus Lac⁻/Ar⁺ recombinants would not add appreciably to total.

63C2A,B Ar⁻ | 64D3 ~~64D~~
Mal⁺Lac⁺ — Ar⁺
Mal⁺Lac Ar⁻

15 ~~B~~
A6 - Lac Gal⁺
Mal⁻ MtL⁻
S^R

1164D3:

Lac⁺ MtL⁻ Mal⁻ S^R all Gal⁺ (Ar⁺)

Lac⁻ MtL⁺ Mal⁺ ~~[illegible]~~ S^S ~~[illegible]~~ recheck, might be mixed. (Ar⁻)

Lac⁺ MtL⁺ Mal⁺ S^S 2? S^R SIC. " " " " (Ar⁺)

No Lac⁻ ~~MtL~~ Mal⁻ picked

MtL-Mal coincid. all but 2 concord S.

F - ductus ; TZ

1167

DATE: 5/24/54.

REF: dd 1113. (139).

W2206 is recorded as very futile F+. Use for F- ductus in chance of also detecting recombinations from pairs.

A. ♂ overnight cultures & ♀TZ .1:1:10 in Nevessary 8:25 PM 37°

B ♀ & ♂TZ 1:.1:10

C W2206 & ♀TZ .1:1:10

Repeat 2:30- & fresher mix.

Concl. (1) W2206 insuff. motile (2) TZ at these levels inhibits motility in these crosses (cf previous observations?).

Pass W2206 again - Recover later if necessary.

T. O. isolates.

DATE: May 25 & 26, 1954. REF: (140)

	1	2	3	4	5	6	7	8	9	10

A 10[30] – W2332, 1 ; ♀ 1:5 — 12 N. overnight cultures

B 12:10 2502, 2503 1:1:5 ⎫ Freshly inoculated – not fully grown.

C " 2332, ♀ 1:1:5 ⎭ — 1:20 PM. (" ").

to 2 PM.

What date on F+/F– pairs?

B) T.O. first batch exc.

5/25 140 H-1-2 s.i.p. F1 –< shaped cell.

few if any pairs were noted.

5/26 12 N. (C) (F+ ; ♀) (B) (28AF– × "~~F+~~ mot Hfr)

A 1/5) but mot — A4 mot snakes

2/3) ♂ mot — ~~C3~~ " "

B 1/2) s.i.p. — G3 s.i.p → 3 cells

C1 ♀? (from s.i.p. pr.) — G2 ⌝

D1 Y cell — F 2/3) R.L. —o

D2/D3) b : –cY prob. mot. — F4/5) —o

D4 ♀ from mot. one. — H 3/4) ╲o

~~AF4 ...~~

stab —

DATE: 5/28/54. P27 A28. REF:

140

	Exp.	Inoctype	Diptype	Blac	Disposition
A 1	68C	mot?	♂	−	False pairs.
2)	"	mot	♂	−	
3	"		♂	−	
4.	68B	snake	O		"
5	C	mot?	♂	−	
B 1)	" C	s.p mot	♂	−	"
2	" C		♂	−	
C 1 .	"	♀	O		"
C 3.	B	mot snake	O		"
4	C		♂	−	"
D 1.) ✗	C	✗ mot	♂	−	
2)	C	mot	♂	−	
3	C		♂	−	
4.	C	♀	O		
1.	Bo	✗ mot	♂	+	
F 2)	B		♂	+	
3	B	╲o ⊙	♀	−	~~[illegible]~~ test for recomb.
4)	B		♂	+	
5	B	╲o.	♀	−	"
G 2	B	⌐	O		
3 ~~[illegible]~~	B	...	♂	∝ . +	
H 1)	Bo	s.p ♀		−	"
2	"			+	
3)	B	╲o		+	
4	B.			−	

Many initially cool pairs evidently iso ("false") ♀ still usually invisible.

Impression that W2502 induces clumping of F⁻ cells. of 1170 also. (♀ means motile (recount))

Try streaks of illeg. mating?

No valid F+/F⁻ pr. in this set. possible 2502 × 2503 pairs to save and check readers of motile side: F¹2-3, ²4-5, H1, 2³ H⁴3, 4.

In particular, streak out Lac⁺ for presence of ⁻. ~~[illegible]~~

Save: 1168. — all pure Lac⁺

1169A.

F Lenny.

DATE: May 27, 1954. REF:

9^{30} count. older culture (140)

A. W2206 M1 .1 : ♀ 1. : 10 broth − 11^{30} Numerous pairs!

11^{30} − 12^{30}.

~~B W2332 .1 ♀ 1 : "~~ leave out in view of A success.

C W2502 1 : W2503 .1 : 10 " CCC, D. 6−8 PM.

CC, AA 12^{45} AA − 2^{10}. C showed very few pairs.

D = ♂ × ♀ 2½° D. Y10 × 2502. 6−8 PM. 140 E1-2. F1 X. F2X.

Late PM: numerous clumps & pairs noted but not now picked.

[140] − [154]		Inoculum.	deoptype A/B		Blac Aq.	# dup. VF
1169A. (F+/♀) B3		♂	−		0	
~~4~~	b1	♂	pd		+	
	2	♀	28d	−	0	
	3	♀...	28d	−	0	
~~===~~						1169-
separated p27. B4		♂	1.		+	A1
	b4	♀ 7.)	28 d		0	
	b5	♀ 1.)	28		−	B1
B5		4♂	1.		+	A2
	a1	4♀	d		0	
C2		♂	d.		0	
	a2	4♀	pd ♀		−	B2
	a3	8♀	pd 28		−	B3
see C5		8♂	1.? pd		0	
	a4	4♀ not cult.	(1).		+	~~A1~~
D5		8♂	1.		+	~~AB~~
	c1	16♀	d.		−	B4
	c2	16♀	28.		−	B5
E2		6♂	1.		+	A4
	c3	8♀	28		−	B6
	c4	not? ♂	1.		+	A5
	c5	"	1.		+	A10
E3		8♂	1.		+	A6
	d1	8♀	28		−	B7
E4		♂	1.		+	A8
	d2	16♀	28		−	B8
E5		8♂	1.		+	A8
	d3	12♀	28		−	B9

1169A.

DATE 140 - 154 REF:

	1	2	Lagotype A28	4/ac	5	6	7	8	9	10
G5		6 ♂	pd	~~O~~	~~[illegible]~~					
	d4	5 ♀	18	—	B10					
	5	20 ♂?	1	+	A22					
H5		all ♂? (same mating).	1.	+						

10

1140.

Precedent.

Note: if W2206 is still infective, these pairs should be examined. The only way is to compare plating of ~~W~~ 1169AA ~~with~~ for F deletion. This culture W2206M1 is mass culture of first passage of the stab stock of W2206. Also plate out single colonies, etc., for comparison of fertility (W2206 was recorded as highly fertile).

69AA also plated on EMB Bac sm. No SRT note, no special point now in looking for recombinants among 1-cell progeny.

Respot series A, B.

69C = plating of 1169AA (refrigerator day) as control. Spot single colonies on ~~pl~~ EMB.

To EML A2 for F test. A = ♂ component } 8/10 F+ EML.
B = ♀ " }
C 4/5 F+ EML.

1169
AA.

1245 – 210 PM. F^+/♀. lac A29 [138]

		Inoculum	digs		Dis. sabe F+
A 1	]		0 I		
2			0		
4		♀	28	–	
B 3		♀	28	–	B13
B 1		♀	28	–	B12
2		♂	0		~~A11~~
B 3	]	♂	0		
4		♂	0 (m.)		
5	]	♀	28	–	B13
C 1		♂	1. 28 1.	+ +	A12 13
2		♀	28	–	B14
5		♀	28	–	B15
C 3		v.s.	0		
4		♂	1	+	A11
D 1		♂	1	+	A14
2		♀	28	–	B16
D 3	]	♀	28	–	B17
E 1		♂	1	+	A15
2		♀	28	–	B18
E 3		♀	28	–	B19
4		♂	1 pd	+	A16
5		♀	28	–	B20
F 1		♂	1	+	A17
2		♀	28	–	B21
F 3		♂	1	+	A18
4		♀	28	–	B22
5		♀	28	–	B23
G 1		♂	1	+	A19
2		♀	28	–	B24
3		♂	1	+	A20
4		♀	28	–	B25
5		♀	28	–	B26
H 1		♀ ♀	28, 28	– –	B27, 28
2		♂	1	+	A21
3		♀	0		
H 4		♂	0		
5		♀	0		

why such less digging than 140? all inner.

DATE: May 28, 1954. REF: [155]

Y10M1 + W2502 .1 : 1 : 10 P enzyme 37° 100 – 245 ...
(A B C1 D E1). 2:30 – 3:45 min NB (C4 E4 F G H).

Three 16 "pairs" isolated, but many proved unviable. On early inspection, 8 PM, record as mixed only:
A1, C1, H1 and these were separated & later proved correct.
However A1 B1 E1 may still have some mg28 type cells.
∴ Pick A1-2, C1-2, H1-3 and B1, E1. in this sequence.
W2502 though nonmotile is not morphologically quite so destructive from line 1 as is W2101. Should compare directly.

		dsps	Lac P30	same streak out
A1 2)	(mixed still)	1, 28? 28	+ –	→ Lac+, few (and Lac-S^R).
C1 2)		1 28	+ –	→ pure Lac+
H1 2 3)	(2.28)	1 ~~28~~ 1 28	+ + –	→ pure + → pure +
B1		1-28?	–+	→ Lac+, –
E1		1-28?	+–	→ "

evidently not!

B1, E1.
① Streak out A1, C1, H1-2 on EMB Lac for lac⁻ recomb. (Gal⁺ or S^S)
② Make ~~as Epistac~~ /mm.

A1 is presumably a mixture of parents only as Lac⁻ = S^R. Others show no lac⁻ in single cell progeny of –. ~~but table shows in B1, E1~~ For more efficient tests see ff. i Lac⁺/V, R.

Mal +/− prototrophs.

1171

DATE: May 30 1954 REF: 1163-64.

A) Esther crossed W2574 x W2118 on EMS Mal. Picked Mal+ to EMS Gal) (mod. crowded), replica to EMS Mal, 1 sun. 45 Mal+. Of these, 9 also had Mal+ S^R. DC 6 is checking 8 of these for concurrence of other classes.

9 also had Mal− S^R thus Mal− S^R/Mal+ S^S. (presumably "twins"). check Lac concurrence.

1-8, DCG: #3 also had Mal− S^R #6,8 also had Mal+ S^S. Not clear whether twins

B) Remainder, look for twins (non crossover) 11-21. checkout on EMS Mal. (Number not clear) 10 had Mal+/−

Lac	Mal	
+	+	3
	−	2
−	+	2
	−	0
−(+,−)		4

No indication of significant twinning. Results altogether inconclusive.

If zygote data (not prototrophs)

DCG 1171.

1171 A

1171A 1-8 (known to contain mal + S^R) ; Streaked out on S mal; picked 10 col. from each streak. Spotted each on S gal; replicated to S mal and S mal S/4. Results:

1: all mal + S^R

2: " " "

3: 5 mal + S^R; 5 mal - S^R

4: all mal + S^R

5: " " "

6: 9 mal + S^R; 1 mal + S^S

7: all mal + S^R

8: 1 mal + S^R; 9 mal + S^S

1171A 11-21 Streaked out on S mal to isolate mal +. In most of these streaks no mal - appeared. Spotted mal + and mal - (mal - differing from S mal S/4 plate) on S lac.

	mal +	mal -		mal +	mal -	
11	lac +	lac +	16	lac -	lac +	
12	+	-	17	-	+	
13	+	-	18	+	+	
14	-	+ and -	19	+	+	
15	-	+ and -	20	-	+ and -	these two alike
			21	-	+ and -	

1171 B

Picked ~~scribbled~~ mal+ col. to Slac; replicated to Smal and Smal SM.

Only 3 contained S^R components

#8 & #15 contained mal- S^R

#14 mal+ S^R

8 & 15; Spotted mal+ and mal- on Slac;

	mal+	mal-
8	lac-	lac-
15	lac-	lac+ and -

14; Streaked out on Smal (→ two mal-); picked 16 colonies to Slac; replicated to Smal and Smal SM. All 16 were mal+ S^R

Misc crossing tests 1172

5/31/54.

		D(0)	Lac:
1.	W2206M1 × ♀	0	
2	1165D2B2 × Y10M1	few ?	
3	1164D3— × "	+	
4	♂ × ♀	see 1171B +	
5	W2581 × Y10M1		1+/>100 —. pure+ col (ortho.)
6	W2583 × W1177MY		ca 1–2% +/— colonies
7	W2583 × W2407.		Probable lac+/— but notches + (ortho is only —).

Mix 3-4 h., plate is washing on D(0), or dilute on EMS Hap EMS Hap lac T5.

∴ W2502 is verified as Hfr + ortho type pattern seems similar.

6) should be most profitable for detection of zygotes. also repeat plating on EMS lac dilut.

♂ nonmotile × ♀ motile
line 28A line 1

173

DATE: July 2, 1954. REF: 158-159

Back to Lab!

ratio 1:10

W2583 × W2639. 1135 — [illegible] hrs.
♂ ♀ W894Mal+motF!!

Final pairs	cell cool.	Drop	EMB lac		Mal	Mtl	Gal	Sm	
save A 2	1	1	(+ –)	Ⓩ:	+		–	S	motile.
save 3	1	1	–		→ pure V, S				
4	1	0	0						
5	28	28 0 pd	0						
6	28	0	[illegible]						
B 3	28		+			s			
4	1		–						
5	1 lost		0						
C 2	1		–						
B 6 ss	28		+						
C 3	28		+						
4	1								
B 2	1		–						
D 1	28		+						
2	1		–						
C 5	1		–						
D 3	large 1	0							
D 5 sip	28	0							
6	1	0							
E 3	1	0							
4	28	0							
E 1	1		–						
2	28	0							
5	28	0							
6	1	0							
F 1	1	n.g	–						
2	28		+						
5	1	? 0							
F 3	28	? 0							
4	1	? 0							
G 1	28	0							
G 4	? 1	0							
5	28	0							
H 3, 4	1, 28	0 0							
H 5, 6	28, 1	0 0							

concordant. ! others no growth! (not 0.) why?

why this half

parental:
all Lac– S^s, Lac+ S^r
Mal+ Mal–
Gal– Gal+
no other recombinants.

Ⓩ was from long duration pair set., unfortunately others inviable.

[illegible]

should check

DATE: July 3, 1957. REF: 159

		2 Cellios.	3 Digg.	4 Blac	5 Mal	6 Gal	7 MtL	8 SM	9	10
A 2	manip	28	28	+						
3		1	1	−						
4	sip	28	28	+						
5		1	0							
B 1	sip.	28	0							
2		1	1	−						
B 4	manip	28	28	+						
5		28	28	+						
6		1? (28)	0							
A6		1	1	−						
C 1	SS	28	28	+						
2		1	~							
B3		1	1	−						
C 3	sip	1	1	−						
4		28	28	+						
C 5	sap	28	28	+						
6		1	1	−						
D 1	SS	1	1	−						
2		28	28	±						
3		1	1	−						
D 4	SS	28	28	+						
5	(Hemmory?)	1	1	±						
6		1	1	−						
E 1	slm.	28 ↔	1	−						
2		1	1	−						
3		1	0							
E 4	SS	28	28	±						
5		1	1	−						
F 1	SS	28	28	±						
2	SS,	1	1	−						
3		1	0							
F 4		28	28	±						
5	manip	1	1							
6		1	0							
G 1	sip	28	28	±						
2		1	1	−						
G 3	manip	1	0							
4		28]	28	+						
5		1	0							
6		28	28	+						
H 1	sip	1	0							
2		28	28	+						
3		1	0							
H4, 5	SS	1	(28) → 28 0	H4 +						

(1PM − 3PM ×) (Isolate 3 − 4 10)
× ratio 1:10
(Separate 5 − 5 30 PM)

all Lac− are Mal+ MtL+ S^S Gal+
Lac+ are
all Lac+ are Mal− S^R Gal+
No recombinants !

manip, sip or [illegible] should be inviable
O ok but not recomb.

all coincident. No Z!
poor viability again!

1174

DATE: July 5, 1954. REF: 160

W2583 x W894 Mal+ M3+ = W2639

V_1^R Gal+ Lac+ W2502

B same cross at 4 hours plate nEMBlac, just two Lac±V(?) pass.

Cross 10:1 12 30 to 2 30 37°

cell drop cell bxdxEMBlac →

1 - all Lac -

28 - all Lac+

except D5 = Lac±/-.

A 1	0	28
2	(1)	1
3	(1.)	1
A 4	(1)	1
5		0
6	0 ~~none~~	1
B 1	0.	28
2	(1)	1
3	(1)	1
B 4	(1)	1
5	0	28
6	(1)	1
C 3	(1)	1
D 6	(1)	1
C 1	(11)	1
2	00	28
C 4	0	28
5	(1) (1)	1 1
6	(1) (1)	1 1
D 1	(4)	1
2	00	28
D 3	0	28
4	0000	28
5	(1)	1 ←
E 1	00.	28
2	(1)	1
3	(1)	1
E 4	000.	28
5	(1)	0
F 1	00.	28
2	(1)	0
E 6	(1)	0
E 3	large motile	1
F 4	00	28
5	0	0
G 1		1
G 4	00	28
2	(1)	1
3	(1)	0
H 1	(1)	1
2	00.	28
3	(1)	1
G 5	(1)	1
G 6	(1)	1
H4-5	G-	28,1 28

prob not saved

H4 found mixed, record — 4: . 5: 0.

Cross O - fams 1175

DATE: July 4, 1954 REF:

10 ml penassay, 1/2 ml ivg-X irradiated ± 1/2 ml W2581 (ivg 20) 1 hr 37°

7/5. Agar motility tubes.

Examine cultures carefully P6.

1175:	ivg.	X: motility	Control: motility	
1	3	0		
2	4	0		
3	9	0		
4	10	0		
	11	+	+	(occ. cells)
5	17	0		
6	18	0		
	27	+	+	(rare)
7	31	0		
	51	Rough +	+	as known
	53	++	++	
8	54	0		

In preliminary tests in motility agar, same results. No T or S. 48h. 30° same.
Reinoculate into Penassay at 25° for pos. temperature effect.
(also try 34, 38, 40, 42, 49, 50: preliminary controls: nonmotile). (see over)

DCG is repeating extensively.

[redacted] of WS cultures

	tube (48h.)	media
22	Swarm throughout tube	motili present
32	slow	motili present
34	non motili	no motili seen
38	" "	" " "
40	" "	" " "
42	flare	no motili seen
47	Swarm throughout tube	motili present
49	non motili	no motili seen
50	" "	" " "
52	Swarm throughout tube	motili present
55	" "	large proportion motili

[redacted]