

DATE: Apr 27, 1954

REF:

	1	2	3	4	5	6	7	8	9	10
	Control platings 2 and 3 parents.									
A	W2344 + W1177				young cultures 1 ml each per 10 ml 2:10 PM. Plate at 4:50 PM. EM 13 loc					
B	"	"	"	W1394						
	Lac ⁺ Gal ⁻ S ^B	Lac ⁻ Mal ⁻ S ^R +		Lac ⁺ Mal ⁺ S ^R F ⁻						
	110 ⁺ 14 ^r									
10	(better to use 1895; 2344 being Gal ⁻ !)									
C	W2344 + W2401 (for Mot ⁺ S ^R selection)				Inoc motility ± sm.					
	4/28/54.				Young cells. 1:30 PM -					
D	W1895 x W1177				X transferred.					
E	"	"	"	x W1394.						

28: (B) Pick sectorial colonies. Test Mal/S. Mal⁺S^R indicates F⁻/F⁻
 14/11 plates x 150 colonies scoreable Mal⁺S^S " Hfr/F⁻
 But Gal⁻Lp^S character of Hfr parent Mal⁻S^R = F⁻ parent.
 may prejudice results. However at this time,
 the Hfr was probably not overly distinct from W1894. (judging from plate + sm)

40 Est 3-4% recomb in A / sm

A29: 5 only Mal Mal⁺. 6 were pure Mal⁻. 3 clearly Mal⁺S^S/Mal⁻S^R.
 of remaining 2, many pure ~~to~~ Mal⁺S^R, other is Mal⁺S^R/Mal⁻S^R.
 Neutral to characterize components.

1152 B1-2 : checked by DCG next page

DATE:

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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

Oscules
 1152 B1-2-3

	1	2	3	4	5	6	7	8	9	10
10	B1 + ①	mal +	mal +	lac +	mal +	S	V5			
	2	+	+	+	+	all normal	S			
	3	+	+	+	+		S			
	4	+	+	+	+		S			
	5	+	+	+	+		S			
	6	+	+	+	+		S			
20	B1 - 2 ①	-	-	-	-		R			
	3 ②	-	sl	+	-		R			
	4	-	sl	+	-		R			
	5	-	sl	+	-		R			
	6	-	sl	+	-		R			
30	B2+ (dark)	+	+	+	+		S			
	2	+	+	+	+		S			
	3 (+)	(+)	(+)	+	+		S			
	4	+	+	+	+		S			
	5	+	+	+	+		S			
	6	+	+	+	+		R			
40	B2+ (light)	(+)	(+)	+	+		R			
	2	(+)	(+)	+	+		R			
	3	(+)	(+)	+	+		R			
	4	(+)	(+)	+	+		R			
	5	(+)	(+)	+	+		R			
	6	(+)	(+)	+	+		R			

(+) = light+

Y10
~~W1177~~ + W1177 + W1394
 lac⁻ SR lac⁺ S
 Hfr lac⁻ SR

B1 = pure SR.
 Mal⁻: W1177 plus recombs!
 Mal⁺: W1394

B2 = pure SR also but pure W1394, does not contain W1177. Might be 2344 x W1394!

∴ either a 3-coincidence or Hfr parent potentiated crossing after!!

Lp^s coincidents

4/27/54 ff Has given irradiated plates of W478 on Bgal, \bar{c} (M-)
suspected sensitive marked.

(1) Picked possible sensitive,
struck one B O;
spotted in order on complete
↓
replated to D(meth)

→ 31 Lp^s ; 2 auxotrophic; all gal⁺

478-4
Both sensitive &
resistant components
original col
auxotrophic
Trypt - W2475

478-5
Sensitive
& auxotrophic
Trypt - W2476

478-1
Grew in
D(meth)
discarded

(2) Several plates replated
directly to D(meth) to
pick up non-Lp^s auxos
(5 plates, ca 150 col/plate)
↓
3 possible double auxos.

478-2
resistant
AA2-

478-3
resistant
Trypt -

5/6/54 ff Started \bar{c} irradiated plates of W478 as before.
16 Lp^s obtained; nutrition checked by EML

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

to notes

DATE: May 8, 1954

REF: 1152

1152 B suggested W1177 x W1394 impregnate of Hfr (W2304) repeat design.

1. $\gamma 10 \times W1177 \times W1941$
lac⁺ S⁺ lac⁻ S⁺ Hfr - S⁺

Grow together 1:1:10 ~~17:30 PM~~ 12:30 PM - 5 PM.

2. $\gamma 10 \times W1177$ (both F⁻)

then streak out on EM13 lac sm. or plate

3. $W1941 \times W1177$ (both lac⁻)

4. $W1941 \times \gamma 10$. (both S⁺)

Sept 10:

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

parents checked for lac, S: O/R

30

40

50

DATE: April 28, 1954.

REF:

Residuate for zygote isolation:

- ① 10⁸ both parents motile
- ② 3 component mixtures as technical control.

10³⁰ - 2³⁰ Mx

10
 A W2341 mot (TCN) x W1177
 B " " " x W1394
 C " " " x 2401.

Ref.

128 40s. cell count.
 ← a?

8:30 A 30. Dilute ca 1:50 to prepare for isolation.

Ref. 10:30

20
~~D Fresh cultures~~

~~W2401 x W2341 Mot (s.c.) 1:2:20 = 4:3:4~~
 " " OK. arplating. (i.e. test of s.c. of 2344 mot.)

5/1/54 D. 9A1 Oldu cultures as inoculum. - N.I.

Ref. [to 3:25. Dilute due to setup: 4:10].

30 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.

40 No - need some lac-linked marker (V₆, V₁?)].

1153 B (128). Random single isolates. 40 placed. F1-G1-H1 flawed together. No lac++ (as indicative of lac+Gal+ recombinants).

5 N.G. (A3,4; B2; D2; G5) agree with depts.

50 Described as actively motile at n=2⁵. C1, C5, D3A D5, These and G2, H4 appear to be v. slow lac+, others lac-. Have DC G streak out on EMS lac, M&L; repl to lac sup., Hal

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	1	2	3	4	5	6	7	8	9	10
	<i>Acetone isolates</i>									
		<i>hc</i>	<i>SM</i>	<i>Mal</i>						
A	1 2 5									
B	1 3 4 10		<i>thus T.</i>							
C	1 2 3 4	±	S	+						
D	1 2 3 4 5	±	S	+						
E	1 2 3 4 5	±	S	+						
F	30 2 3 4 5									
G	2 3 4	±	S	+						
H	40 2 3 4 5	±	S	+						

*no recombination's
evident (strong hc+)
(non W1394) pres.*

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	1	2	3	Score	5	6	7	8	9	10	
1153D	<u>129</u>	hopeless		lac	Mal	Gal	SM	MH.			
1	A 1 pair!	28		-	-	+	R	-			
2	B 1 pair!	28		- +	-	+	R (lact, -)	-			
3	E <u>2</u> <u>3</u> int? <u>4</u> <u>5</u>	28		-	-	+	R	-			
4		28		+ -	-	+	R	-			
		1			+	+	-	S	+		
		28			-	-	+	R	-		
	F <u>1</u> dump <u>2</u> <u>3</u> <u>4</u> <u>5</u>	1		+	+	-	S	+			
		1		+	+	-	S	+			
		1		+	+	-	S	+			
		1		+	+	-	S	+			
		1		+	+	-	S	+			
5	G <u>1</u> <u>2</u> pair	1		+	+	-	S	+			
6		28	28	-	-	+	R	-			
A	<u>2</u> <u>5</u>	1		+	+	-	S	+			

no
parent
parent

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

∴ 4 pairs have given 4 2 zygotes! B, E the Hfr parent reappeared, but gave 3. A: 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 isotonics.
and other notes on sterility

1154

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4/28. *E. coli* C - 4th passage (see 1153) gave entirely motile culture, still coaroid. Continue passage at R.T. (1157B4)

(This culture started from micro-cool of motile cell → semi-motile clone. Then to agar's 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°. Remincubate. [do. 52C sent.]

W1895 - sluggish, occ. motile. [Are unselected motile cells F⁻?]

W2284 - [Hfr mot → F⁻]. Highly motile.

Select by passage through motility tubes. W111, [redacted] W231, W242, W245, W246, W247, W248, W249, W250, W251, W252, W253, W254, W255, W256, W257, W258, W259, W260, W261, W262, W263, W264, W265, W266, W267, W268, W269, W270, W271, W272, W273, W274, W275, W276, W277, W278, W279, W280, W281, W282, W283, W284, W285, W286, W287, W288, W289, W290, W291, W292, W293, W294, W295, W296, W297, W298, W299, W300, W301, W302, W303, W304, W305, W306, W307, W308, W309, W310, W311, W312, W313, W314, W315, W316, W317, W318, W319, W320, W321, W322, W323, W324, W325, W326, W327, W328, W329, W330, W331, W332, W333, W334, W335, W336, W337, W338, W339, W340, W341, W342, W343, W344, W345, W346, W347, W348, W349, W350, W351, W352, W353, W354, W355, W356, W357, W358, W359, W360, W361, W362, W363, W364, W365, W366, W367, W368, W369, W370, W371, W372, W373, W374, W375, W376, W377, W378, W379, W380, W381, W382, W383, W384, W385, W386, W387, W388, W389, W390, W391, W392, W393, W394, W395, W396, W397, W398, W399, W400, W401, W402, W403, W404, W405, W406, 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5/23. Note: in recent weeks

$$\sigma \rightarrow = \underset{\text{wg 1.}}{W2344M1} \text{ Hz} \quad \text{f} = \underset{\text{wg 28A}}{W2401} \text{ F}^-$$

Question of λ reactions

$$W2401 \text{ proves } \lambda^R \lambda_2^R.$$

$$\text{cf. } W2284 = \lambda_2^S.$$

emulated i Mal⁻ as before?

Struck out overgrowth of
 $W2284/\lambda_2$.

~~slip page~~

DATE: MAY 3, 1954

REF:

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

9:05 AM Make up control (F x F) W2331M1 x W2401 1:1:20

B. (Pencam); B1 (V/B) (norm) (36 h.)

MAY 4 1954

10 C. 132 but exp. n.g., throw out W2344M1 x W2401 in V/B. 1:1:20 9 AM 37°

1155 A has grown to ca 5×10^7 (density) by this point.

Dilute for set up = A2. Also fresh dilution from 1155 = A3
A.T. 1:100 Inc 37°

See cont page.

Most pairs give obv. mixed clones not worth picking. Some of these come however picked apart quite early. V.z.

- A2-B5 +/- - (A2 not uniall) No test
- * D1-D4 D5 all lac - Presumably was not a pair.
- D2-D3 ±; +/- (5 x, → -
- F1-F2 ±; +/- → ✓
- E3 F5 G5 ± (-+) - → ✓
- G4 H5 - +- → ✓

40 1. every pair then separated evidenced zygote formation!

45 H counted as associated pair

50 Could pairs come from fission of zygote?

DATE: May 3, 1954.

REF: 1153D!

overnite
9:30 AM Fresh cultures (not regrown)
W 2344M1 x W 2401. 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".

P.ich 1 = pair of isolate - 1; use most - 28°
1155A lac lac⁺ Mal MH bal

	1	2	3	4	5	6	7	8	9	10
A	1-4 PM		8 PM	10 AM						
A1	P	+	-			A2	A2	±	-	R (rare)
A2	P	+	-			A2	B5	-		R
A3	P	+	-			A2	D1	-		R
A4	P	+	-			A2	D2	±		S
B	P	+	-			A2	D3	+	-	R ⁺
B1	P	+	-			A2	D4	-		R
B2	P	+	-			A2	D5	-		R
B3	P	+	-			B5	E3	±		S
B4	P	+	-			B5	F1	±		S
C	P	+	-			B5	F2	±	-	R ⁺
C1	P	+	-			B5	F3	-	-	R ⁻
C2	P	+	-			B5	G4	-		R
C3	P	+	-			B5	G5	-		R
C4	P	+	-			B5	H5	+	-	R ⁺
D	P	+	-			D1				
D1	P	+	-			D1				
D2	P	+	-			D1				
D3	P	+	-			D1				
D4	P	+	-			D1				
E	P	+	-			E3				
E1	P	+	-			E3				
E2	P	+	-			E3				
E3	P	+	-			E3				
F	P	+	-			F1				
F1	P	+	-			F1				
F2	P	+	-			F1				
F3	P	+	-			F1				
F4	P	+	-			F1				
G	P	+	-			F5				
G1	P	+	-			F5				
G2	P	+	-			F5				
G3	P	+	-			F5				
G4	P	+	-			F5				
H	P	+	-			G4				
H1	P	+	-			G4				
H2	P	+	-			G4				
H3	P	+	-			G4				
H4	P	+	-			G4				

On restriction, D2, E3, F1 are pure lac⁺ (bal⁻); 5, 8 are pure lac⁻; 2, 4, 7, 9 are lac⁻ and lac⁺ bal⁺ (recombinants) 4 had⁺ only in center spot. Save initial mixtures in stab for later analysis

Did any pairs here give pure 28?
Total P isolated: 19.

D1 G4. Not certainly pairs from acids

✓
summed up

DATE: May 5, 1954

REF:

Parent cultures overnight.
 P. massary 10:15 AM 37°

W 344M1 x W 2401. 1/1:20 in

Plate MAY 6 1954

134

EM/Blac S

MR; Mal

Gal

CLASS

Growth pattern

AT

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+

R-

R also R

R+

R+

R+

R-

R+

R-

R+

R+

R-

R-

others S.

Maltic MR-

probable contamination

V₁

B2 thus lacks ♂ but appeared odd at first.

B5 may have ♂

♂ C1, D1, F3, F5 lact ♂

♀ E4 only pair not yet showing odd recomb.

♂ B5 has ♀, 2 recomb

♀ B2 ♀, recomb

♂ B5

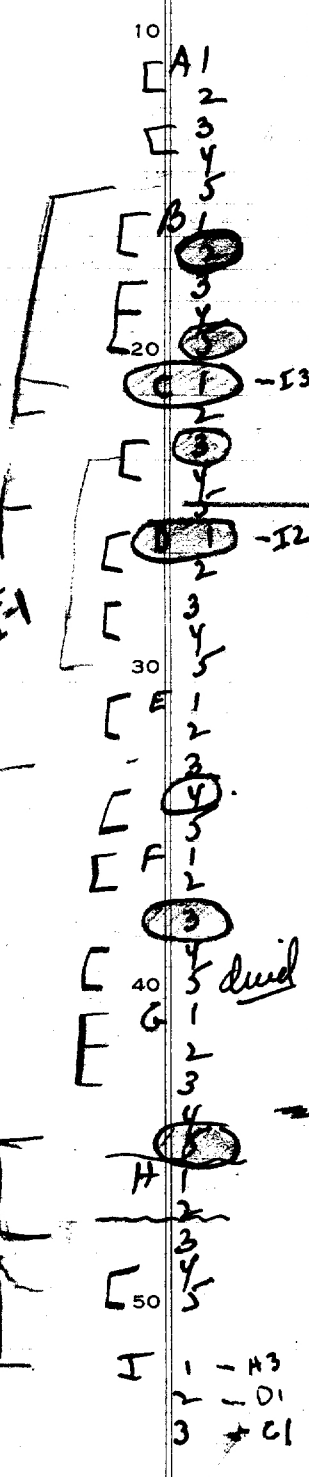
♂ B5

♂ B5

♂ B5

♂ B5

♂ B5



DATE:

REF:

1 2 3 4 5 6 7 8 9 10

B2, B5. - both nonmotile in dinit test.
 DCO also tested E1 D1 E4 F3 F5, non-swarm
 & E5 - swarm (as expected).

10 E4 } pure Gal⁺ by streak. E4: V₁^S
 B5 }

B2 originally showed weak lac⁺ reaction and some lac⁺SR.
 It appears now pure Gal⁺ by first streak, but some lac⁺SR indicated.

20 Maybe simply lac⁺SR ~~is~~ recombinants in low numbers with ♀ excess.

B5: shows lac^(♀)SR; lac⁺SR but also some late Mal^(P?). Not evident on early streaking.

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

30 Check growth colonies:

Strain	Col ^S	lac	Mal	Xyl	Notes
B2	①	+	-	+	Xyl ⁺ lac ⁺ recombinant
	2	+	-	+	
	3	+	-	+	
	4	+	-	+	
B2	1	-	-	-	all Mal ⁻
	2	-	-	-	
	3	-	-	-	
	4	-	-	-	
B5	①	+	+	+	Xyl ⁺ lac ⁺ Mal ⁺ recombinant
	2	+	+	+	
	3	+	+	+	
	4	+	+	+	
B5	1	-	-	-	Xyl ⁻ lac ⁺ Mal ⁻ recombinant
	2	-	-	-	
	3	-	-	-	
	4	-	-	-	

all carry the orthotypic markers Gal⁺ SR and Mal⁻.

10/6-7/54. Also found, in B5, a Mal⁺ lac⁻ recombinant occur

1176 B5 - 4 = Mal+ Lac- MH- Gal+ Ara-
SR Xyl+

all SR MH- Ara- Gal+

∴
Lac+ { Mal+ Xyl+
Lac- { Mal- Xyl-

DATE: May 6, 1954

REF:

W2401, W2344M / germozymyot.
 Regrow 1:10 9⁰⁵ - 10⁰⁵ AM.
 fresh isolations

135

Dilute & place in separate drops for
 early mixture.
 Pairs not well defined

10 A 1-2-5 proved all motile
 A 3-4 ditto
 B 1 ♂ ?
 B 3 ♂
 20 C 1 ♀
 C 4 ♀

A 1 # 1 proved
 2 2 ♂
 5 0 ♂
 30 3 ♂
 4 4 ♂
 B 1 11 pure ♀
 3 5 pure ♀
 C 1 12 pure ♀
 4 13 pure ♀
 40

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

50

Hfr x W1177.

~~#58~~
1157.

Conjugal pairs.

DATE: 1954 MAY 8

REF:

W2344M1 x W1177 fresh cultures. Mix 2PM. Redelite and
(not) not?
examine 3PM. (1 hour mixture)
do 135

10

lac S Gal Mal

1	+		-	+
2	+		-	+
3	+		-	+
4	+		-	+
5	-	R	++	-
7p	-	R	++	-
12	-	R	++	-
13	-	R	++	-
14	+	D2	-	+
15	+	D5	-	+
21	+		-	+
22	-	R	±	-
23	+	EV	±	+
24	+	ES	-	+
29	-	R	±	-
31	+		-	+
32	-	R	±	-
33	-	R	±	-
34	+	H2	-	+

all pairs illegitimate defined associated

recombinants here.

Φ

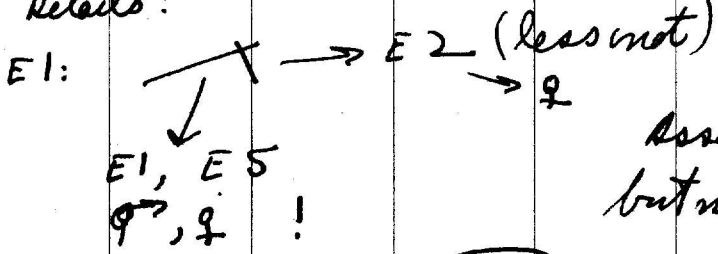
1157

E1
E2
E3
E4
E5
E6
E7
E8
E9
E10
E11
E12
E13
E14
E15
E16
E17
E18
E19
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E30
E31
E32
E33
E34

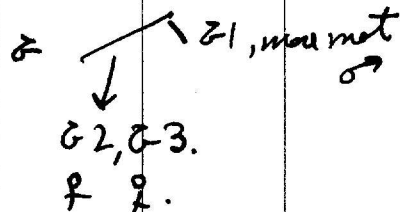
2 surviving pairs are legitimate. Note lethality of remainder (see protocols). of 5 other pairs, 1 had no survivor, 3 had ♂ only; 1 ♀ only.

Mme ++ others S.

Details:



Assoc. ca 1 hour! but no recomb detected.



Φ

Assoc ca 15m. but no recomb detected.

There should be saved!
 = 1158 21-25
 31-33

Gal ++ = 2101
 Gal ± = 1177
 Gal - = 2344

50

1 was "separated by surface tension" (dyeing): both lost.
of the 5 persistent pairs, 4 were "separated" spontaneously

B1-2 → ♂ ✓ ♀ and ⊗ ✓

H1-2 → ♂ died ♀ " ✓

don't count Dy.

A3-5 A' died A3 ⊗ | → ♀ only.

28-♂ = W2344M1
28-♀ = W2401.

D1-2-5 d → ↓ ♂ ↓ ♀ ↓ both died

← count as "viable pair"

A1-2
1 was manipulated at 1 hour
O- → ♀, ♂ 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⁻bac⁺ indicated (♂)
save for further comparison. Abundant 147 as
these show no particular heterogeneity (among survivors).

F2 was plated out on EMB Col.

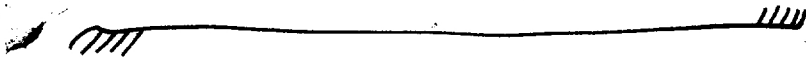
(probably old)

Zygote isolation

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, ~~male~~ ♂ pipet at pipette.



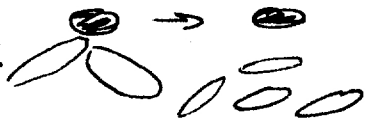
1155 Most pairs not separated (19). 4/4 from line 28 parent
[130]

53 D 4/4 partly separated.
[129]

Total to date (W2344M1 x W2401): 13/16

Queries:

1. These are all fairly late pairs. Each is should be studied to ensure that zygotes result from pairs, not concourse, and to seek diakaryon cells such as 1156 B2. Notes should be consulted in event of doubt as purity of this cell. None indicated.



2. Perhaps fuller pedigrees.

- 3. Review possibilities of line-1 crosses (cf 1158. Third Fl^{a-} 1177 or 1895!
- 4. Cytology.
- 5. Find previous resume!
- 6. Problem of undetected recombinants: why not more?

May 12, 1954

DATE:

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 ♀ gave in 9; ♂ only in 3 and both died in 0. This suggests that viability maybe connected with separation but other differences are possible.

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

B. [143] Pick 5 pairs (rather clumosity) for pedigree analysis.

A1: 1 - A, B, ^{1 cell}
 ♂ ♀

B 1 cell

2A: — A2♀ B2♂ A2 D2. C2! E2
 C♀ F2

all grow.

3A: A C BL BK C D
 ♀ ♂ ♂ ♂ ♂
 E 1 cell

all but E grow

4A: — B♀ A♂ C D E4
 ♀ ♀

B. 1 cell only

5A: — A♂ B♀

both grow.

chance to correlate growth delay & zygotes.

Note fair survival of these, too.

DATE: MAY 14 1954

REF:

	1	2	3	4	5	6	7	8	9	10
	<u>144</u>									
		EMStac		Lac	Mal	M ⁺	Xyl	Zal	S	
A. 1	?	-		-	-			+	R	
2 a	1-28	}	+	-	-	+	+	+	-	R R ⁺
b	28			-						
3 a	1-28	}	+, -	+	-	+	-	+	+	R ⁺ R ⁻
b	28			-						
4	28	-		-	-			+	R	
5	28	-		-	-			+	R	
E 5	28	-+		-+	-			+	R	R ⁺

not separated

EI-4FI-4
see 143

all parents concordant

not clear zygotes

E5 from A1, but long contact!

A2-3 off. inseminated

A4 clear test?

β → β° + (||||)

no zygote.

Survival not clear.

A5 long contact, still -!

saved

saved

9 A.M.

1160

MAY 14 A.M.

MAY 1954

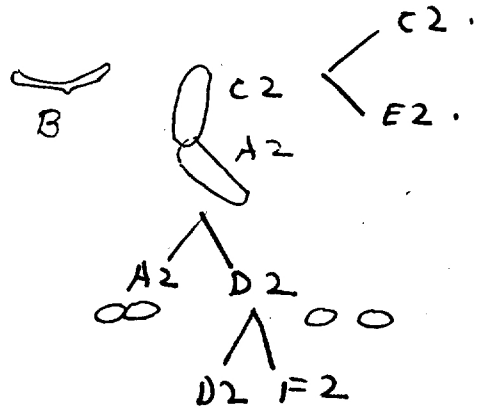
9AM 1431

1. ^B ♀ died A ♂

2. A2 zygote ♀
B ♂
CDEF ♀

same as 1160-2-

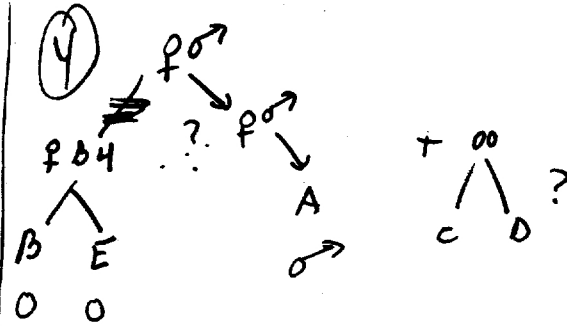
♂ stepping



3. A-C-D-F ♀
E-G O
B ♂

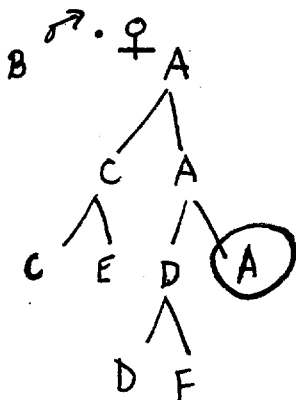
4. A ♂
B E O
C D ♀

5. A ♂
B d.

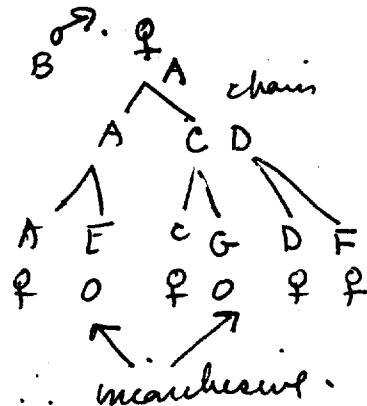


poss. monocherive also.

(2)



(3)



DATE: May 13 1954.

REF: 142-141

Ovenyjit, x 10⁵² - 11⁵⁵ 37° 1:1:10 Pineassy.
 A1 picked ca 130.
 A2

Freshcross x 12³⁰ - 3 PM SIC.

Not separated: ~~any~~ Refrigerated ovenyjit +
 separations to 142, 141, A14.

	1	2	3	4	5	6	7	8	9	10
141	A4	EMB lac	Gal	Mal	MAL	Xyl	S			
	A5	-	+	-	-	-	R			
	C1	+	-	+	✓	✓	S			
	3	-	+	-			R			
	4	++	+	-			R			
	5	+	-	+			S			
20	D1	+	-	+			S			
	2	-	+	-			R			
	4	-	+	-						
	5	-	+	-						
	E4	-	+	-						
	5	-	+	-						
	G1	-	+	-	✓	✓				
30	H1	-	+	-						
	2	-	+	-						
	3	-	+	-						
	4	-	+	-						
	5	-	+	-						

all parents
 EMB lac

1161 is a Mal+
 yields not showing a
 Mal+ lac- recomb but
 exhibiting a Mal- lac+
 recomb. also

142

	1	2	3	4	5	6	7	8	9	10
G1	++	+	+	+	+	+	+	+	+	+
2	+	+	+	+	+	+	+	+	+	+
3	+	+	+	+	+	+	+	+	+	+
5	+	+	+	+	+	+	+	+	+	+
40	H4	0								
	5	-								

Pair of R (lac?) Mal MAL Xyl
 S
 S
 S
 R

	1	2	3	4	5	6	7	8	9	10
		lac	SM	Mal	MAL	Xyl	Gal			
G1	++	-	R (lac)	(+)	-	(+)	+			
2	+		S	+	+	+	-			
3	+		S	+	+	+	-			
5	+		S	+	+	+	-			
50	H5	-	R	-	-	-	+			

Gal among
 Mal or Xyl needs to
 reconfirm.

Re - 10/54. Pure Gal+.

Found infant: A
 lac - MAL - S^R Mal - Xyl

US are
 lac+ MAL + S^S + +
 MAL - S^S - -

No lac+ S^R Mal + S^R (Re-examine for
 ① lac+ S^R No
 ② lac- Mal + No
 many colonies
 spods tested)

stb

DATE: May 14, 1957.

REF: 137

	1	2	3	4	5	6	7	8	9	10
A	1	CMRbac	♂ ad.							
B	2	+	♂, ad							
	3	+	♂, ad							
	4	+	♂, ad							
	5	+	♀ ad.	N.S.	♂ ad.					
D	5	+	♀							
E	5	+	♀ ad.							
H	5	+	-							
(G5)										

X 1:1:20 (fresh) 12:30-2:10
 (set up to 2:40).
 Trapping technique, worked
 v. well but petiole maybe
 too narrow.

Three viable pairs only: A3-D3 ♂. ♀ no ♂
 (G5)-H5 ♂. ♀ ♂
 B1 (n.s.) ♀ (suspect appearance -
 resemble CMRbac.)

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

Now incidence = 1/2 pairs zygotic

	1	log	Xyl-Mal-	S	cal	
A	2	+	+	S	-	all concordant
B	3	+	+	S	-	
	4	+	+	S	-	
D	3	-	-	R	+	
	3	+	+	R	-	
E	3	-	-	R	+	
	3	+	+	S	-	
H	5	+	+	R	+	
	5	+	-	R	+	

Pairs. Try also
 Hausman's trap
 Salmorella.

1163.

DATE: May 19, 1954.

REF:

150

① The culture (W234641) seems sluggish. Perolate motile cells = A1-4. A1 n.g. (A3) probably best to be put up
 From this serial #, use 6x8 coverless markings. Letting in
 sequence A B C ... D ... E ... F ... G ... H ...

② Set up parents in adjacent drops: (♂) (♀) (O) next to trap
 drop, note that ♂ has to swim through ♀ to reach trap. May
 have worked moderately well, but setting 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!

Conventional mixture 1:1:6 2¹⁰ - 3¹⁵ isolation from
 ca. 340 - 4:40, using trap drop

③
 B
 C
 D
 E
 F
 G
 H
 I
 J
 K
 L
 M
 N
 O
 P
 Q
 R
 S
 T
 U
 V
 W
 X
 Y
 Z

see next page

Comment: survival fairly good.

Note: both manip ♀♀ died but also
 2 others. Would predict several zyg.
 but may be some post clonal lethality not
 detected.

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

all clars to
 date have
 agreed &
 recorded type of
 cell manipulated.
 Note will be made
 of any discrepancy.

DATE: 5/20/24

5/21/24 5/22

REF:

A) state	History	Age type	EMBLac	lac	Gal	MALTR	S ^B (lac)	save	10
B	1 ss.	♂	+		-	+	S		
	2 7h.	●							
	4 s. downed	♂	++		-	++	S		
	5 late pair	♀	-		+	-	R	-	
	6	♀			+	-	R	-	A2+
C	1 ss	♂	+		-		S		
	10 2h.	♀	-	2	+	++?	R	-	
	3	♂	+		-	+	S		
	4 s.i.d.	♂	+		-	+	S		
	5	♀	-	3	+	-	R	-	
	6	♀			+	-	R	-	
D	1 late pair	♂	+		-	+	S		
	2 Manip.	●							
	29 late pair	♂	++		-	++	S		
	5 s.s.	♀	-	4	+	-	R	-	
	6 2h.	♀	++		+	-	R	-	
E	1 only	♂	+		-	+	S		
	2 bubble	♀	-	5	+	-	R	-	
	4 s.s.	♂	+		-	+	S		
	5	♀	-	6	+	-	R	-	
F	4 72h.	♂	+		-	+	S		
	5	♀	●						
G	4 2h.	♂	+		-	+	S		
	5	♀	-	7	+	-	R	-	
	6	♀	++						
H	1 Manip	♂	+		-	+	S		
	2 2 sticks tip	♀							

all conc.

These C 2 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!

at front second reading only 3/7

50
 s.s. = s.p.
 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 isolates - possibility of contamination at isolation is not inherently excluded.

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

No unamb. 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 EM13 Gal. Gal⁺ > Gal⁻ colonies.

Pick and streak: 5 Gal⁺ 6 Gal⁻. all are

lac⁻ M⁺ Mal⁻ Xyl⁻ S^R and are thus simply a

} Non-motile in motility agar
10/54

Gal⁻ / atthotype recombinant! Streaked on EM13 lac, no lac⁺ are

noted, but some colonies are pink (lac⁻ Gal⁻?). When crowded with some pink "+" reactions.

- a) Check Hfr, Lp, mutations.
- b) Might be Lac⁺ Gal⁻ Xyl⁻ (checked by Gal⁺ -x?)
- c) Presumably not lac⁺ and Hfr would give lac⁺ Gal⁺ recombinants.
- d) Check motility, Mal⁺?

DATE: Mar 20, 1954.

REF:

① Already identified as carrying ϕ and lac^- Gal⁻ M^H Mal⁻ Xyl⁻ SR.
 In initial spot and especially in first replica to lac , definite +
 reaction was indicated, suggested possibility of modified lac^+ Gal⁻ Xyl⁻, etc.
 Also doubtful possibility of presence of Mal⁺.

11A23. 10 Replicate from original spot to stab and 1163C2

- a) motility - none in susp. from agar #C2, C2', C2A etc. ; + in agar.
- b) streak EM13 lac again
- c) Mal, M^H
- d) X, X - Gal⁺

e) Compatibility: M^H x 1895M2; ♂
 1 C2A 1895M2
 2 2206
 3 C2B 1895M2 + + +
 4 2206

p25	M ^H
0/0	Slac
0	
+	all -
+++	+? -?
++	+? -

also please check for h_p , Hfr status, h_p , may interfere & former. nutritional. Should certainly be crossable & Y10 line.

f. Nutrition: both M-H -
 presumably Hfr .

orthotypy: C2A = Gal⁺; C2B = Gal⁻ colonies.
 Brougher is necessary than A.


924 → Pure Mal⁻ M^H⁻ lac^- & lac^+ reaction in background. Reincubate.
 True reaction comes from interaction of Gal⁺ lac^- / Gal⁻ lac^+ colonies
 attempt Gal⁺ recessive of W2502

W2502

40 This recomb. could very readily be missed unaided! ar.

Note exaggerated orthotypy of e2 of e3

Aggregation of
 metals, etc.
 Note most pathogens
metals! (~~part of~~)

11  amount
 vs. degree of metal

DATE:

	mm ¹ metal	pm ² metal	high ³ metal
loc + gal + metal + soil +	1	6	15
+ + + -	0	1	1
+ + - -	0	1	4
- + + +	0	0	3
- + - + 10	0	0	0
- + - -	0	0	1
- + + -	0	0	0
Total	1	8	24

No - this case is

all Gal^+ nonmotile

D3 includes

Mal^+	Lac^-	MH^+	S^S
Mal^+	Lac^+	MH^+	S^S

} first of these!

~~✗~~

$\text{Mal}^- \text{Lac}^+ \text{MH}^- \text{S}^R$ (A)

perhaps should be reviewed even more thoroughly.

Mal^+ purified all S^S ; some unpur. give $\text{Mal}^+ \text{S}^R$ "contaminants" or mutants?

possible: (I.S.C.I)

See 10/1/54

- | | | | |
|---|-----------------------------|----------------|-----------------------------|
| 1 | $\text{Lac}^- \text{Mal}^-$ | H-M^+ | $\text{Ara}^- \text{U}_1^S$ |
| 2 | $\text{Lac}^+ \text{Mal}^-$ | H-M^- | $\text{Ara}^+ \text{U}_1^R$ |
| 3 | $\text{Lac}^- \text{Mal}^+$ | H^- | $\text{Ara}^- \text{U}_1^S$ |
| 4 | $\text{Lac}^+ \text{Mal}^+$ | H-M^- | $\text{Ara}^+ \text{U}_1^N$ |

See 1183. In routine check, H^- segments found

among 1164D3. Among isolates 1-4, #3 was ~~$\text{H}^- \text{M}^+$~~ $\text{H}^- \text{M}^+$ (others presumably $\text{H}^- \text{M}^-$ as no growth on Major). To try to find:

other types (1) streak growth of mixture through D(H) on EMB Mal,

found ca 1% Mal^- mass (all Mal^-).

(2) streak on EMB Mal and test

DATE:

REF:

	1	2	3	4	5	6	7	8	9	10
E 10	1	al. manip.	Cell type ♂ ♂ d?	lac ++		Gal --	Mal NH ₄ yl	S _S (lac)		
	2									
	3									
F 20	4	lh. ss	D ●	++	8	--	+ + +	R	-	
	5									
	6									
G 30	1	lh. ss	D ●	+	6	-	+	-	-	
	2									
	3									
H 40	1	lh. ss	d.	+		-	+	-	-	
	2									
	3									
H 50	4	lh. ss	d.	+	9	-	+	-	-	
	5									
	6									

concordant, no recombinants.

←



Note losses from drying, lethality. Try saturating oil in water.
 Counting only complete pairs: only 2 = 1/6; add incomplete pairs: ~~4/6~~
 complete: 1-6: incomplete 7-10
 2/6. 1/3 3. Total 3/9 only.
 #8 is certainly out on lethality (further d. of E6)
 & probably 9 also.

DATE: MAY 21 1954

REF: 152

X (old), 1 ♂ 1.0 ♀ 5 ml broth 37° 9' - 10³⁰ AM. Trap.
 (B) Compressible col 2331M2 Examined at same time: no pairs.
 Try 2344M2 (F-). see 1166. (A25)

	1	2	3	4	5	6	7	8	9	10
			dep	LAC	Lac	Mal MAL	Gal	S (lac)		
A 10	1) X manip.		♂							
	2) X		♀	-	✓					
	3) .		♀	+	✓	Mal, Mal	+	R (-)		
	4) X base		♀	-	✓					
	5) X		♀				+	R		
	6) .		♀							
B	4) .	57 early	♂	+	+	Concen.	-			
	5) .	43 μ	♀	-	①		+	R		
	6) .		♀	+	+		+	R R +		
			♀							
C 20	1) .		♂	+	-		-			
	2) .	lost dep	♀	-	②		+	R		
	3) .		♀							
	4) .		♂	+	+		-			
	5) .	SS	♀	-	③		+	R		
	6) .		♀	-			+	R R		
			♀							
D 30	1) .		♂	+	+		-			
	2) .	SS	♀	+	+	Mal *	+	R R - +		* same + ?
	3) .		♀	-	④		+	R		
			♀							
E	1) .	sl man.	♂	+	⑤		-			
	2) .		♀	-			+			
	3) .		♀	-			+			
	4) X		♂	+			-			
	5) X	distorted	♀			one		as Mal		
			♀							
F 40	1) X manip.		♂	+			-			
	2) .		♀	-			+			
	3) .		♀							
	4) .	SS	♀	-	6		+			
	5) .		♀							
	6) .		♀				+			

A) all lit
 B) all lit
 D4 " " manip

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

50

D2A. $lac^+/-$, pure Mal⁻.
 same +, -

D2B. lac^- ; Mal⁻ and Mal⁺.

D2B1.

4 of these should be out. Prove to be mucoid, no distinct indication of segregation +/-.
 3 lac^+ muc. D2B2
 1 lac^- muc. D2B3

Mal	lac	S
pure Mal ⁻	+	R
pure +	+	S
3 -	-	R

	Mal	Lac	S
1	-	+	R
2	+	+	S
3	+	-	S

becoming less mucoid. all are all auxotrophic.
 not segregating.

D2A has ♀, $lac^+ Mal^-$

D2B has ♀, $lac^+ Mal^-$, $lac^+ Mal^+$, $lac^- Mal^+$ all lac^+ .

presumably failure of Mal elimination & crossing over $\frac{S}{Mal}$.

of 11640 3! In both cases, only ones which show Mal⁺, all 4 combinations are seen. Is $lac^+ Mal^+$ Hfr giving reciprocal recombinants?

5/23/54.

		A 25						
		EMB lac	lac	Hal ^r Hal ^{r2} Xyl.	Gal	S	lac)	
G 1)	1	+	✓	+	-			♂
	2	-	1 ✓	-	+			♀
	3	-	✓	-	+			♀
	4	+	✓	+	-			♂
	5	-	8 ✓	-	+			♀
	6	-	✓	-	+			♀
J	1	+	9 ✓	+	-			♂
	2	-	✓	-	+			♀
	3	-	++ ✓	-	+	R	-	♀
	4	+	✓	+	-			♂
	5	0						0
	6	0						0
J	1	-	-	-	+	R		♀
	2	-	-	-	+	R		♀

all deep concordant in the formula.

9 pairs complete, $\frac{1}{2}$ zygote! Note segregation in B4 pedigree.

4: zyg. SS; SS, sl. men; SS

5: unzyg (det.) lost deep; s.p; SS, SS; SS.

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

DATE: May 22, 1954

REF: 153

Yesterday's observations suggest that motile F- (cf. also ⁷Salmonella) does not pair with W-2401. This can be properly confirmed only by a competitive pairing experiment. Suggest: manage a trois with Hfr mot; F- mot; F- 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.

10

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

10

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

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

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

C. W-1895 M2 + W-2401.

Previously concerned about bp^s of W2401 but EML finds it non-s.d to λ. (possibly Mbl-bp₂?)
W2338, W2384 being incubated.

20

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

In controls, no pairing was seen in C compared to A.

Control for B: streak out on EMBS lac, Test lac⁺ on EMBS lac to verify ratio of Hfr/F⁻.

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

EMBS Gal:

A	ca. = +, -	(sic! indicates growth diff.?)	Lac.
B	+ > -		+ almost = -.
C	all +.		+ = -
			+ almost = -.

Sample lac⁺ to EMBS Gal. (Pick every chain + available to avoid bias on slight difference of appearance): 23 Gal⁺ : 7 Gal⁻
(F-) (Hfr)

50

Competitive pairing

153

Start

A 25

lots of
offspring
already

	Proc.	Age	Sex	Bl Lac	lac	Mal	MH	Gal	S (lac)
A 1	①	0	♀	-	✓	-	-	+	R
A 2	00000000	0	♀	-	✓	-	-	+	R
A 3	40	0	♂	-	✓	-	-	+	R
A 4	11	0	♂	+	✓	+	-	-	R
A 5	11	0	♂	+	✓	+	-	-	R
A 6	00	0	♀	-	✓	-	-	+	R
B 1	0	0	♀	-	✓	-	-	+	R
B 2	50	0	♀	++	✓	-	-	+	R
B 3	1+	0	♂	++	✓	+	+	+	S
B 4	.	0	-	++	✓	+	+	+	S
B 5		0	♂	+	✓	+	-	-	R
B 6	60	0	♂	0	✓	+	-	-	R
C 1		0	♀	++	①	-	-	+	R
C 2	0	0	♂	++	①	+	-	-	R
C 3	60	0	♀	++	①	-	-	+	R
C 4	0	0	♀	+-	①	-	-	+	R
C 5	10	0	♂	+	②	+	-	-	R
C 6	0	0	♀	.	②	+	-	-	R
D 1		0	♂	+	✓	+	-	-	R
D 2	0000	0	♀	.	✓	+	-	-	R
D 3	0		♂	+	3	✓	+	-	R
D 4	0	00000000	♀	-	3	✓	-	+	R
D 5	0		♂	+	4	✓	+	-	R
D 6	0	0000	♀	-	4	✓	-	+	R

SIC 1895

why are
A6
recovered
later?

lac⁺ personal
Gal⁻

all race
etc. B3

off-m
strains
pure Mal⁻

	Gen	diag	Blac	loc	Mal xyl	S(lac)	Gal
E 1	0	0					
2	⊖ at met	0					
3	2+	♂	+	5	+	+	-
4	0	♀	-		-	-	R +
5	+	♂	+	6	+	+	-
6	00	♀	-		-	-	R +
F 1	+	♂	+	7	+	+	-
2	0000	♀	-		-	-	R +
3	0	0					
4	0000 met!	♂	+		+	+	-
5	⊕	♂	+		+	+	-
6	000	0					
? G 1	00 met // 00	(♂)?	+	(++) 9	+	+	-
2	- met // //	♂	(++) 9		++	+	+
3	+	♂	+	8	+	+	-
4	0000	♀	-		-	-	R +
5		-					
6		-					
H 1	0	0	#				
2	⊖ m.	♂	+		+	+	-
3	0000	♀	-		-	-	R +
4	⊕	0					
5	(111)	♂	++		++	+	+
6	*0	♂	++		++	+	+

$V_1 R, V_1 S$ } parental

conver.

∴ all pairs were either Hfr/F₂₈⁻ or (G1-2) and (H5-6) which carry met F- alone or i Hfr.

Following distribution noted: (complete pairs only):

pairs	singles
8 Hfr/F ₂₈ ⁻	7 Hfr ₁
1 Hfr/F ₁ ⁻	2
0 F ₁ /F ₂₈ ⁻	23 F ₁ ⁻

Need replication 2/8 zygotic

could we detect recombination between G1/G2? Only answers are Galhp/V₁? Test /75. ✓

DATE: May 25 1954.

REF:

	1	2	3	4	5	6	7	8	9	10
A.	(old cultures)		W 2502 + W 1895 M2							
					(W 2503)					
			1	:	.1		:10	10:45 AM		

B. W 2332 + W 2401 (F denton).

10

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

20

30

40

50

MAY 24 P.M.J

9¹⁵ Same EMB agarose tests on lac^- & Ar^- variants.

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

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

152 ¹¹⁶⁵ A6 C3 C5-6ab F356 G23 G56 J1-2

153 ¹¹⁶⁶ D4 D6 E4 E6 F2 G4 H3

all were Ar^- except? (B6)

These lac^-/Ar^+ recombinants would not add appreciably total.

63 C2 A, B	64 D3 G4 D
Ar^-	$Mal^+ lac^+ - Ar^+$
	$Mal^+ lac^- Ar^-$

116403:

$lac^+ MH^- Mal^- SR (Ar^+)$ all lac^+

$lac^- MH^+ Mal^+ S^S$ ~~reverts~~ ^{SIC.} ~~might be mixed.~~

$lac^+ MH^+ Mal^+ S^S$ 2? SR " " " "

15 ~~B~~
A6 - $lac^+ Mal^+$
 $Mal^- MH^-$
SR

(Ar^-)
(Ar^+)

No lac^- ~~reverts~~ Mal^- picked

$MH^- Mal^-$ ~~reverts~~ ^{and all but} ~~reverts~~ ^{reverts}

DATE:

5/24/54.

REF: see 1113.

139.

1 2 3 4 5 6 7 8 9 10

W2206 is recorded as very fertile F⁺. Use for F-direction in chance of also detecting recombinants from pairs.

A. ~~W2206~~ $\sigma \rightarrow \text{d} \text{ } \text{f} \text{ } \text{TZ}$ ^{overnight cultures.} 1:1:10 in presence 8:25 AM 37°

B. $\text{f} \text{ } \text{ } \sigma \rightarrow \text{TZ}$ 1:0:10

C. W2206 $\text{f} \text{ } \text{f} \text{ } \text{TZ}$ 1:1:10

Repeat 2:30 - & further more.

Encl. ① W2206 misuff. motile ② TZ at these levels inhibits motility in these strains (cf previous observations?).

Pass W2206 again - Recount 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 full grown.			
C	"	2332, ♀	1:1:5		- 1:20 PM.	(" ")			
					to 2 PM.				

what date
on P+ / F-
pairs?

5/15
20
B) T.O. first batch exc.
140 H-1-2 s.i.p.
few if any pairs were noted.

F1 ← shaped cell.

5/26 12N (C) (F+ ; ♀)

A 1) ^{just} not
5) ^{not}
302) ♂
3) not

B 1) sip.
2)

C 1 ♀? (transip m.)

D 1 Y cell

D 2) b: - 24

D 4 ♀ from not me.

~~FF 2332~~

prod. not.

(B) (28AF- x " not 1/2)

A 4 not snubed
B 3 " "
G 3 sip → 3 cells
G 2 7
F 2) - 0
3) h.l.
F 1) - 0
H 3) 0
4)

stab

5/28/54.

DATE:

P27

A28.

REF:

140

	1	Exp.	Inotype	Keytype	Blac	Disposition	8	9	10
A	1	68C	not?	→	-	<u>False pair.</u>			
	2)	"	not	→	-				
	3)	"	not	→	-				
	4)	68B	scale	→	-		"		
	5)	C	not?	→	-		"		
B	1)	"C	sip not	→	-	"			
	2)	"C		→	-	"			
C	1.	"	♀	0		"			
C	3.	B	not scale	0		"			
D	1.)	B	> not	0	-	"			
	2.)	C	not	0	-	"			
	3.)	C		0	-	"			
	4.)	C	♀	0	-	"			
F	1.)	B ₀	♀ not	0	+	fact fact for recomb.			
	2.)	B ₀	✓	0	+				
	3.)	B	✓	0	+		"		
	4.)	B	✓	0	+		"		
	5.)	B	✓	0	+		"		
G	2)	B	✓	0	X				
	3)	B	✓	0	+				
H	1)	B ₀	sip ♀		-	"			
	2)	"			+	"			
	3)	B	✓		-	"			
	4)	B	✓		-	"			

Many initially cool pairs evidently is (false)

♀ still usually viable.

Impression that W2502 ^{♀ means} induces chipping of ^{metals} F⁻ cells. of 1170 cells. (moment)

Try streaks of ally. meting? Save: 1168. — all pure fact

No valid F⁺/F⁻ pro. in this set. possible 2502 x 2503 pairs to some and check inclusions of metal side: F²-3, 4-5, H¹, 2, H³, 4.

In particular, streak out fact for presence of ~~and~~

Save: 1168. — all pure fact

1169A.

F. Kemp.

DATE: May 27, 1954.

REF:

	1	2	3	4	5	6	7	8	9	10
9 ³⁰	ovnt.		older capture							
A.	W2206 M1 .ol.		♀ 1.	:	10 both	-	11 ³⁰	Numerous pairs! 11 ³¹ -12 ³⁰		
B.	W2332	.1	♀ 1	:	"	-	11³⁰	leave out in view of A success.		
C.	W2502 1	:	W2503 .1	:	10 "			CCC, D, 6-8 PM.		
CE, AA	1245		AA - 2 ¹⁹ .					C showed very few pairs.		
D = ♂ x ♀	410 x 2502.	6-8 PM.						140 E1-2-F1 X. F2X.		
2 ¹⁹	late PM: numerous clumps & pairs noted but not now picked.									

1169A.	140-154	broculum.	deotype	total	area	dep.				
(F+/♀)	B3)	♂	-	0						
	4)	♂	pd	+						
	2	♀	pd	0						
20	3	♀...	pd	0						
	B4									
separated p27.	B4	♂	28	+			1169-			
	b4	♀ 7.)	28	+			A1			
	b5	♀ 1.)	28	+			B1			
	B5	4♂	1.	+			A2			
	a1	4♀	d	+			A2			
30	C2	♂	d.	+						
	a2	4♀	pd ♀	+			B2			
	a3	8♀	pd 28	+			B3			
	C5	8♂	1? pd	+						
see	a4	4♀ w/	⊙.	+						
	D5	8♂	1.	+			B3			
40	c1	16♀	d.	+			B4			
	c2	16♀	28.	+			B5			
	E2	6♂	1.	+			A4			
	c3	8♀	28	+			B6			
	c4	not?	1.	+			AB			
	c5	"	1.	+			A10			
	E3	8♂	1.	+			A6			
	d1	8♀	28	+			B7			
50	E4	♂	1.	+			A8			
	d2	16♀	28	+			B8			
	E5	8♂	1.	+			A8			
	d3	12♀	28	+			B9			

DATE 1/4/01 - 154

REF:

	1	2	3	4	5	6	7	8	9	10
65		6 ♂ 44 5 ♀ 5 200?	dup type A18 pd 18 1	4ac 0/1 - +	0/1 B10 A22					
H5		all ♂? (same parent).	1.	+						

11/10
E. Woodmont.

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

69AA also plated on EM13 bac. No SRT noted, no special point now in looking for recombinants among 1-cell progeny.

Respot series A, B.

169C = plating of 1169AA (refidung day) as control. Spot single colonies on EM13.

To EML A2 for F test.

A = ♂ component	}	8/10 F+	EML.
B = ♀			
C			

4/5 F+ EML.

Time	Sex	Age	lac A29	Disp. site
1245-210 PM <td></td> <td>F+/♀ <td></td> <td>138</td> </td>		F+/♀ <td></td> <td>138</td>		138
		brood		the Fv
A1) 2) 3)	♀	0 0 28	-	
B1) 2)	♀	28 0	-	B18 B17 A11
B3) 4) 5)	♂	0 0(m.) 28	-	B19
C1) 2) 5)	♀	28 28	+ + - -	A1213 B14 B15
C3) 4)	♂	0 1	+ +	A11
D1) 2)	♀	1 28	+ -	A14 B16
D3) 4) 5)	♀	28	-	B17
E1) 2) 3)	♀	1 28	+ -	A15 B18
E3) 4) 5)	♀	28 1 28	- + -	B19 A16 B20
F1) 2)	♀	28 28	+ -	A17 B21
F3) 4) 5)	♀	1 28 28	+ - -	A18 B22 B23
G1) 2)	♀	1 28	+ -	A19 B24
3) 4) 5)	♀	1 28 28	+ - -	A20 B25 B26
H1) 2) 3)	♀	28 1 0	- + -	B27, 28 A21
H4) 5)	♀	0		

why such less dying than 140^{all} imm.

DATE: May 28, 1954.

REF: 155J

1 Y10M1 + W2502 .1 : 1 : 10 necessary 37° 100 - 245 ...
 (A B C | D E I). 2:30 - 3:45 in NB (C4 E4 F6 H).

Thus 16 "pairs" isolated, but many proved invalid. an early suspicion, 8PM, record assumed only:

10 A1, C1, H1 and these were separated & later proved correct.

However A1 B1 E1 may still have some wj28 type cells.

∴ Pick A1-2, C1-2, H1-3 and B1, E1. in this sequence.

20 W2502 though nonviable is not morphologically quite so distinctive from line 1 as is W2101. Should compare directly.

A1 (unmixed) 1, 28? + → lact+, few (and Lac-S^R).
 2) 28 -

C1) 1 + → pure lact
 2) 28 -

30 H1) 1 + → pure +
 2) 28 + → pure +
 3) 28 -

B1 1-28? -+ → lact, -

E1 1-28? +- → "

40 evidently not!

B1, E1.

① streak out A1, C1, H1-2 on EMBS Lac for lac⁻ recomb. (Gal⁺ or S³)

② A1 also as ~~EMBS~~ / ~~sm.~~

A1 is presumably a mixture of parents only as lac⁻ = S^R. Others show no
 50 lac⁻ in single cell progeny of F⁻. ~~but test clones in B1, E1~~ for more efficient tests suff. i. Lac⁺/V₁R.

DATE: May 30 1954

REF: 1163-64.

A) Estlin crossed W2574 x W2111 on EMS Mal. Picked Mal⁺ to EMS Gal (mod. crowded), replica to EMS Mal, Esus. 45 Mal⁺. Of these, 9 also had Mal⁺SR. DCG is checking 8 of these for concurrence of other classes.

9 also had Mal⁻SR thus Mal⁻SR/Mal⁺S⁺.

(presumably "turnis"). Check loc concurrence.

1-8, DCG: #3 also had Mal⁻SR #6, 8 also had Mal⁺S⁺. Not clear whether turnis

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

~~W2574~~
~~W2111~~

Mal ⁺ -	
loc ++	3
+-	2
-+	2
--	0
-(+,-)	4

No indication of significant turning.
Results altogether inconclusive.

if replicate data (not prototrophs)

1171 A

1171A 1-8 (known to contain mal + SR) ; Struck out on S mal; picked 10 col. from each streak. Spotted each on S gal; replicated to S mal and S mal SM.

Results:

- 1: All mal + SR
- 2: " " " "
- 3: 5 mal + SR; 5 mal - SR
- 4: All mal + SR
- 5: " " " "
- 6: 9 mal + SR; 1 mal + S^S
- 7: All mal + SR
- 8: 1 mal + SR; 9 mal + S^S

1171A 11-21 Struck out on S mal to isolate mal+. In most of these streaks no mal- appeared. Spotted mal + and mal- (mal- ^{definitely} from S mal SM plate) on S lac.

	<u>mal +</u>	<u>mal -</u>		<u>mal +</u>	<u>mal -</u>
11	lac +	lac +	16	lac -	lac +
12	+	-	17	-	+
13	+	-	18	+	+
14	-	+ and -	19	+	+
15	-	+ and -	20	-	+ and -
			21	-	+ and -

} these two spots together

1171 B

Picked several mal + col. to S lac; replicated to S mal and S mal SM.

Only 3 contained SR components

#8 & #15 contained mal - SR

#14 - mal + SR

8 & 15: Spotted mal + and mal - on S lac;

	<u>mal +</u>	<u>mal -</u>
8	lac -	lac -

15	lac -	lac + and -
----	-------	-------------

14: Streaked out on S mal (→ no mal -); picked

16 colonies to S lac; replicated to S mal and S mal SM.

All 16 were mal + SR.

5/3/51/.

	D(0)	Lac:
1. W2206M1 x ♀	0	
2 1165D2B2 x Y10M1	few?	
3 1164D3 - x "	+	
4 ♂ x ♀	see 1171B	+
5 W2581 x Y10M1		1+ / >100 - . pure + col (ortho.)
6 W2583 x W1177M4		ca 1-2% +/- colonies
7 W2583 + W2407.		Probable lac+/- but not char+ (ortho is only -).

Mix 3-4 h., plate & wash up on D(0), or dilute on EM13lac T5.
EM5Hsd

∴ W2502 is verified as Hfr + orthotypy pattern seems similar.

6) should be most profitable for detection of zygotes. also repeat plating on EM13lac diluent.

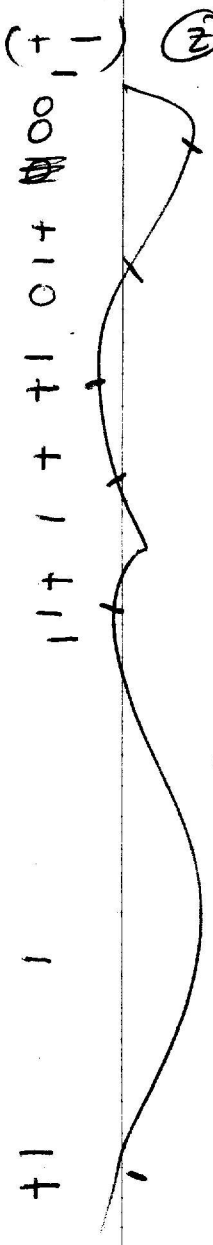
♂ nonmotile x ♀ motile
line 28A line 1

DATE: July 2, 1954.

REF: 158-159

	1	2	3	4	5	6	7	8	9	10
Parents to Lab!	W2583 x W2639		W894M + W07F	1135 - 1135 byjus.						
Final pairs	cell cool.	Drop	EMB lac	Mal	MAL	Gal	SM			
A 1-6	1-6	1-6	(+ -)	+	+	-	S			motile.
B 3-5	28	lost	00	+						
C 2-6	28		+	+						
C 2-4	28		+	+						
B 2	28		+	+						
D 1-5	28		+	+						
D 3	large 1		+	+						
D 5-6	28		+	+						
E 3-4	28		+	+						
E 1-6	28		+	+						
F 1-5	28		+	+						
F 3-4	28		+	+						
G 1-5	28		+	+						
G 4	28		+	+						
H 3-4	28		+	+						

concordant.
 only this half n.g.
 other no growth! (not d. / why?)



all lac-ss, Lac+ s
 Mal+ Mal-
 Gal- Gal+
 no other recombinants.

(2) was from long duration pair set,
 unfortunately, others viable.

should be OK

DATE: July 3, 1954.

REF: 139

	2	3	4	5	6	7	8	9	10
A 2 3 manip	cell. 28 1	dup. 28 1	blac + -	Mal	Gal	MAL	SM	(1PM - 3PM x) (Dedote 3-410)	
4 5 sip	28 1	28 0	+				ratio 1:10	(Separate 5-5 3PM)	
B 1 2 10 dip.	28 1	0 1	-						
B 4 5 6 manip	28 28 28	28 28 0	+ + -						
A 6	28	0	-						
C 1 2 SS	28 1	28 1	+ -						
B 3 20	1	1	-						
C 3 4 sip	28 1	28 1	+ +						
C 5 6 sap	28 1	28 1	+ -						
D 1 2 3 SS	28 1 1	28 1 1	+ + +						
D 4 5 6 SS (known)	28 1 1	28 1 1	+ + -						
E 1 2 slm.	28 1	1 0	- -						
E 3	1	0	-						
E 4 5 SS	28 1	28 1	+ +						
F 2 3 SS	28 1	28 1	+ -						
F 4 5 6 manip	28 1 1	28 0 0	+ + +						
G 1 2 sip	28 1	28 1	+ -						
G 3 4 manip	28 1	0 28	+ +						
G 5 6	28 1	0 28	+ +						
H 1 2 sup	28 1	0 28	+ +						
H 3 5 SS	28 1	0 28	+ +						
HY 1 5	28 1	0 28	+ +						

all lac-are Mal+ Mal+ S⁺ Gal+
 lac+ are
 all lac are Mal- S⁺ Gal+
 No recombinants!
 many sup or
 inviable O should be
 OK but not
 recomb.

all independent, Wo Z!
 non viability again.

DATE:

July 4, 1954

REF:

10 ml penassay, 1/2 ml wg-x ^{37°} incubated ± 1/2 ml W2881 (wg 20 H₂O)

9/5. Noz motility tubes.

Examine cultures carefully Pb.

1175:	wg.	X: motility	control: motility
1	3	0	
2	4	0	
3	9	0	
4	10	0	
5	11	+	+ (occ. cells)
6	17	0	
7	27	+	+ (rare)
8	31	0	
	51	rough +	+ unknown
	53	+ +	+ +
	54	0	

In preliminary tests in motility agar, same result. No Tor S. 48h. 30° same.

Remoiculate into Penassay at 25° for poss. temperature effect.

(also try 34, 38, 40, 42, 49, 50: preliminary controls: nonmotile). (see over)

40 DCG is repeating extremely.

2 Notes of Ws. cultures

	<u>tube (48h.)</u>	<u>notes</u>
22	Swarm throughout tube	motile present
32	slow	motile present
34	non motile	no motile seen
38	" "	" " "
40	" "	" " "
42	flare	no motile seen
47	Swarm throughout tube	motile present
49	non motile	no motile seen
50	" "	" " "
52	Swarm throughout tube	motile present
55	" "	large proportion motile

