

Multiple heterozygotes

Oct 2, 1948

1. W477 x W67 (Lac₁- x Lac₄-). 15 plates.
2. W125 x W478 466 10
3. W133 x W478 466 10
4. W125 x 487 5
5. W133 x 487. 5

1. + and - colonies, variegation?
 → 2. Variegated, c small - sectors. } Restrains on EMS Lac.
 W73, 74

1. No colonies! Later 2 + noted! ~~---~~
2. Numerous +. 64 picked to EMB. 11/64.
3. 1+ in 10 x 200 tests. = 327-3-1 Pure ++
4. Ca 20% +.
5. 0 or 1? + in ca 5 x 200 = 1000 tests.

59, 51, 47, 48 - heterozygous - 6, 1, 61, 20 - streak out on EMS Lac.

60, 43? ? 48h. (EMB.)

H	1	2	3	4	5	6	7	8	9	10	Notes
75	1	1									Small cols. Many nearly + colonies. occ. ⊙
76	2	6									Numerous ⊙
77	3	20									Nearly +
78	4	47									" "
79	5	48									All +; sedover
79	6	51									+ and ⊙ colonies. Also ⊙
80	7	59									⊙ and ⊙ do.
81	8	61									do.
82	? 9	60									mostly v. small +, - colonies. 1 var. ⊙
83	? 10	43									Good var., +, - good growths ⊙

Oct. 6, 1948.

all in lac 5.

1. W478 x W583

2. " W584.

3. W477 x W45.

4. ~~W477 x W186.~~

Plates made 1 in black (Xyl; lac) are repeated 10/7/48.

2. 40 tested; 11 selected for further test. S.O. EMSlac

3. No yield

1. xylose: 72 tested

5, 6, 7, 8, 12, 35, 43, 48, 49, 60

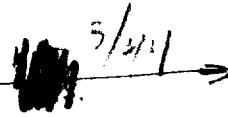
65, 66, 67, 71

1. lac - 22 44 tested.

16, 19, 23 are Var. on lac 115

Monodermis EMS

Monodermis EMP



H.

- 1 5
- 2 6
- 3 7
- 4 8
- 5 12
- 6 35
- 7 43
- 8 48
- 9 49
- 10 60
- 11 65
- 12 66
- 13 67
- 14 71
- L 15 16
- L 16 19
- L 17 23

-
- ?
-
-
-
- V
- V?
-
-
-
-
-
-
- V
- V
- +

- 48h-
-
-
-
-
- V.
-
-
-
-
-
-
-
-
-
-
-

} m EMBlac,

at this reading, - colonies show rather peculiar appearance. - definite centers, but no well-defined sectors.

October 13, 1948.

Test	isolates on Lac, Xygl.		H-
	lac	Xygl. EMSlac-EMS Xygl	
1	Var	-	85
2	Var	-	86
3	Var	-	87
4	Var.	-	88
5	Var.	+ ++	89
6	++ ?	+ ++	90
7	Var.	- ++	91
8	Var.	- -	92
9	++	- -	93
10	Var	- -	94
11	Var	- -	95
12	Var	- -	96
13 Var	Var	- -	97
14 Var	Var	- -	98
15 Var		++ ++	99
16 Var		++ ++	100
17. ++.		++ ++	101

Recheck carefully.

Not heterozygous?

Not heterozygous

Cultures variable on EMS Xygl.

Prick to 7(6) slants from Xygl EMS. Incubate Lac EMS further.

Test 330-2 isolates on all media available.

	Lac	Mal	Sac	Xyl	Arab.	H	
1	+, - V?	-	++	V	++		56
2	++	-	++	++	++		
3	V	-	++	++	++	101	57
4	++; -	-	++	++	++	102	58
5	++	++	++	V	++	103	59
6	+; -	-	++	++	++	104	60
7	V	-	++	V	++	105	61
8	V	-	++	++	++	106	62
9	± V	+	++	V	++	107	63
10	++; -	-	++	(-?)	++	108	64
11				++			65

Study additional isolates from 2 and 11 for study

- 2

-11
 1 ++
 2 ++
 3 +,
 4 +, V, - H65.

Oct. 16, 1948.

Streak out H85-88, 91-97 on Lac EMS. Pick papillae & ~~EMS~~ ^{purify on} EMS ^{Lac⁺ EMS}

	1	2	3	4
1 H85	+	+	+	+
2 86	+	+	+	+
3 87	-	-	-	-
4 88	-	-	-	-
5 91	+	+	+	+
6 92	+	+	+	+
7 93	-	-	-	-
8 94	+	+	+	+
9 96	+	+	+	+
10 97	-	-	-	-

-1 definitely segregating for Lac+/- + 2 (+) 3 (+) 4 (+) *

* semopapilla. Hold 3, 4, 10 for papillation.

EMS:

	1	2	3
1 v. wk +, opp. segn.	+	+	+
2 wk. field +	+	+	+
5 sectorial colo. (V)	(V)	(V)	(V)
6 sectoring weak +	(V)	(V)	(V)
7 (V) *	(V)	(V)	(V)
8 bulls-eye colo same sectoring	++	++	++
9 ++	++	++	++

	4
++ flat colo.	++
(V)	(V)
(V)	(V)
(V)	(V)
++	++

Notes.

- H110
- H111
- H112
- H113
- H114
- H115
- H116.

dark, nice.

And choose * for preservation as T(10)

Compare H93 (→ V) and H96 (→ V) in detail. Streak both out on Lac EMS for further papillae. See 3-15-11 1-2

Oct 5+, 1948.

A). Streak out single variegated colonies and pure colonies to heterologous + homologous media.

An original test plates, 1 colony only was seen on xylose, but 3 lac- colonies seen in H72. Pick these as 333A:1-3 and streak on xylose EMB.

P7.B) Streak 5 var colonies each from Xyl plates H70, H72 to Xyl + Lac

H		Xyl.	Lac
70	A	++ , -	Pure +
	B	+ , - , var.	almost pure +.
	C	- , + , var.	unrecognizable.
	D	" "	almost pure +.
	E	+ , " , var.	" "
72	A	+ , - , var.	Mostly - , +.
	B	+ , - , var.	+ , - , var.
	C	+ , var , -	- , + , var.
	D	+ , " "	+ " "
	E	" " "	" "

Series 70, especially, seems to show loss of lac variability within Xyl segments. Pick var. colonies from Xyl plates to lac + Xyl EMB.

70B-(1-3). not for isolated var. + ...

P7. A). 2, 3 are pure xylose-. (1) Contains preponderantly - but some + or variable. Pick these to Xyl EMB and ^{to} lac EMS. [Not isolated colonies.]

1	+ - ; var on xylose.	Pick to lac EMS and lac EMB.
2	+ -	
3	Pure +	
4	Pure +	
(0)	to EMS. See 333a.	

333A: 1-6.

	Xyl.	Lac
1	±	±
2	±	±
3	±	±
4	±	±
5	±	±
6	±	±

No partial seg-
negation here

accidentally, not pure - when picked.

333A0 is a plate of EMS Lac streaked ultimately from a ~~lac~~ "lac" colony of H72. About 50% are lac-. Test them on Xyl EMS. Keep on Lac EMS.

c). Streak out H72 on 45 Xyl + Lac to look for - colonies.

P10

1-3	lac - ?
4-8	Xyl - ?

	lac	Xyl.	
1	-	-	(1)
2	-	-	(2)
3	-	-	(3)
4	-	-	(4)
5	-	-	(5)
6	-	-	(6)
7	±	±	(7)
8	-	-	(8)

False to lac + Xyl EMS for papillae, except (7).
 No papillae on Xyl. Also H72: no papillae.

P18.

Papillae tested on EMS Lac.

	1	2	3	4
1	++	++	++	
2	++	++	++	
3	++	++	++	
4	++	++	++	
5	++	+	++	
6	+	++		

These lac- prototrophs are monogenic for lac-

333a.

P10. B)

70 B'	1	++;	-	-+V
	2	++;	-	-;+;V
	3	+++		-;+
c'	1	++	-	-+V
	2	++	V	-+V
	3	++	-	-+V
	4	++(-)		-+V
E'	1.	++	ming	Var.
72. Ax	1.	-;	V	-
	2.	+	-; V	-
B _L	1			+ - V
	2			+ - V
	3			+ - V
	4			+ - V
B _x	1	+ - V		
	2	+ - V		
	3	+ - V		
	4	+ - V		
C _L	1	V	- +	
	2	V	- +	
C _x	1	-	V	
	2	-	V	
	3	-	V	
	4	-	V	
DL	1	++		
	2	++		
D _x	1	- +		
	2	- +		
E _L	1	++		
	2	++		
E _x	1	- V		
	2	- V		

Except for HT2 DL, and doubtfully for series 70) the segregation of lac and Xyl is strictly correlated. Pick colonies and mass of DL to check on segregation.

D ₁	1	-	
	2	-	
	3	+	
	4	-	
	5	+	
	6	-	
	7	-	
	8	-	
(0)		±	
D ₂	1	-	
	2	+	
	3	-	
	4	-	
	5	+	
	6	±	
	7	-	
	8	-	
(0)		±	

should all be pylose +.
Note alteration from typical behavior of HT2.

→ 333B1 + 2.

Pick var. colonies from ~~DL~~ D₁₃ and D₂₆ and
 a) test nutrition.
 b) streak on EMS Lac.
 c) s.o. on EMBlac + Xyl to verify of HT2.
 (1 : Variegated, +, - both on lac and Xyl
 (2 : " " " "

The error was based on the use of SalEMB as Xyl EMB.
 No partial segregation here!

October 12, 1948.

- | | | |
|---------------|------------|--------------------------------|
| ① W108 x W466 | Mostly - ! | From Lac EMS to Lac |
| ② W327 x W466 | Mostly + ! | From Lac EMS to Mak |
| ③ W252 x W466 | Mostly - | 100+ pedes. Lac EMS to Lac |
| ④ W108 x W478 | Mostly - | Lac EMS. to Lac |

① 24 tested. 10, 12, 18, 73, 3, 4. = 1-6

② 48 tests. No heterozygotes noted.

③ 79, 64, 82, 49, 52, (97), (94), (96), 2. 7-15. *had lighter appearance on EMS Lac*

④ 20; (others?) 16.

Retests:	vac	blue		
1	Var	++	M-100.	
2	?	V?		W108
3	V	++	M-101	x
4	V	++	M-102	W466
5	++	++		
6	V	V?	W103	
<hr/>				
7	V	++	104	} W252 x W466
8	V	++	105	
9	++	++		
10	++	++		
11	V	++	106	
12	V	++	107	
13	++	++		
14	V	V?	108	
15	V	V?	109	
16	?	++?		

Retest column from 16. None segregating.

10/16+/1948.

A) Grow H72 in Y2 broth overnight to allow segregation, and plate on lac; Xyl EMPB.

Counted by N. Z. + calculated.

	+	-	Var.	Σ
Xylose a.	20	274	6	
b.	25	345	8	
c.	16	196	5	
	61	815	19	895.

$\chi^2_4 = 0.15$
 $p = .99!$

$\therefore + = \frac{815}{61} = 13.3 : 1 = \alpha$

lactose

29	228	9	266
15	178	4	197
32	248	10	290
76	654	23	753

$\chi^2_4 = 3.17$ $p = .53.$

$\therefore + = \frac{654}{76} = 7.5 : 1 = \beta.$

This gives linkages as Xyl - Pf = ~~25~~ 7.0

Lac - Pf = ~~10~~ 11.8

B). Take - and + colonies and test on heterologous medium.

	lac-	lac+	Σ
Xyl-	109	16	125
Xyl+	64	0	64
	38		

lac - Pf = $\frac{16}{125} = \frac{16}{125} = 16.6$
 Interference?

	Xyl-	Xyl+
lac-	104	7
lac+	182	1

Xyl - Pf = $\frac{7}{111} = \frac{7}{111} = 6.3$

+ colonies from 336a retested on both media.

- 1-16 " Xyl - Lact⁺
- 17-23 Lac - Xyl⁺
- 24 Lact⁺ Xyl⁺.

	EMB	Xyl	Lac
1	-	-	+
2	-	-	+
3	-	-	+
4	-	-	+
5	-	-	+
6	-	-	+
7	-	-	+
8	-	-	+
9	-	-	+
10	-	-	+
11	-	-	+
12	-	-	+
13	-	-	+
14	-	-	+
15	-	-	+
16	-	-	+
17	-	-	+
18	+	-	-
19	+	-	-
20	+	-	-
21	+	-	-
22	+	-	-
23	+	-	-
24	+	+	-

(Lact⁺ -).

6 24 -

	Xyl	Lac
1	+	+
2	-	+
3	-	-
4	+	-
5	-	+
6	+	-
7	+	-
8	+	-

not segregated for either lac or Xyl (S.O.) test isolates.
 and a mixture of
 Xyl⁺ Lac⁻ and Xyl⁻ Lac⁺.
 Sure --- (4) was also
 found, the culture may have been
 a mosaic.

	X	L
9	-	+
10	+	-
11	+	-
12	-	+
13	+	-
14	+	-
15	-	+
16	-	+

+	Df	+
xyl		Lac
-	x	y

xyl - lac -	$(1-x)(1-y)$
xyl + lac -	$x(1-y)$
xyl - lac +	$y(1-x)$
xyl + lac +	xy

①. Interference: In A, $\frac{x-l+}{x-l-}$ should = $\frac{x+l+}{x+l-}$. $\chi^2 =$
 Expectations in some columns are < 5 .

②. Linkage. Use only single crossover data.

$$\text{lac} - \frac{x_{yl-}}{x_{yl+}} = \frac{1-x}{x} = \frac{1}{x} - 1. \quad (2b).$$

$$x = \frac{1}{r_b + 1}$$

$$336. r_b = 17$$

$$x = .055$$

$$3369. r_b = \frac{104}{7} =$$

$$x = .077.$$

$$\text{mean: } r_b = \frac{111}{36}$$

$$x = .061$$

χ^2

34	2	36
104 ✓	7	111
138	9	147

$$X_{yl-} - \frac{Lac-}{Lact+} = r_a.$$

336: $r_a = 33/6$

$r_b = 109/16$

$\bar{y} = 15.4$

$\bar{y} = 13.4$

$\bar{y} = 12.8$

108.	17	16	125
109	5	6	39
3334			
142	22	164	

$$\chi^2 = \frac{1}{5} + \frac{1}{17} + \frac{1}{34} + \frac{1}{108} =$$

- .01
- .20
- .06
- .03
- .30

$p = .0660$

Summed data 336...

Lac-	X _{yl-}	X _{yl+}
	34	2
	104	7
	<u>138.</u>	9

X_{yl-} Lac- Lact+

X _{yl-}	33	6	
	109	16	
	<u>142</u>	22	174

336 a. Random plotting. Defend from absence of X+L+ class.

X-L-	1328
X-L+	167
X+L-	122
	<u>1606.</u>

gives $x = 7.7$
 $y = 11.2$

Oct. 15, 1948.

W583x58-161.

low yields: Abandon exp.

1) EMS lac

October 19, 1948.

Repeat.

ca 30:1 - : +

EMS Xyl B ₁	Σ	+	-	EMS Xyl:	Σ	+	Σ	+
	32	2			54	4	201	2
	136	1			339	1	120	2
ca 3% +	41	1		ca 1% +	147	3	162	1
	41	1			277	3	178	1
	31	3			96	0		
	28	1			199	1	2218	23
	309	9	300		170	3		
					92	1		
					183	1		

2) EMS Lac B₁. Colonies picked indiscriminately to homogeneous medium.

Classified by presumptive 1st original score + B₁ in plates:

1. Xyl + B₁
2. - B₁
3. Xyl + 0
4. Xyl - 0
5. lac - 0
6. + 0
7. - B₁
8. + B₁.

This experiment unsuccessful on two counts

- ① Tests were not decisive, most suspensions found being apparently mixtures.
- ② Confusion of classes.

Group	Lac	Mal	Gal	Xyl	Arab
1	+	-	+	+	+
2	+	+	+	+	+
3	-	+	+	+	+
4	-	-	-	+	-
"Xyl +" T(0)	-	+	-	+	+
	-	+	-	+	+
	-	-	-	+	+
	-	+	-	+	+
	+	+	+	+	+
	+	+	+	+	+
	+	+	+	+	+
	+	+	+	+	+
	+	+	+	+	+
	+	+	+	+	+
	+	+	+	+	+
	+	+	+	+	+
"Lac -" T(0)	+	-	-	-	+
	+	-	+	-	+
	+	-	+	-	+
VI Lac + T(0)	+	-	+	-	+
	+	-	+	-	+
	+	-	+	-	+
	+	-	+	-	+
	+	-	+	-	+
	+	+	+	+	+

8.

8a

	lac	Mal	Gal	Xyl	Arab
1	+	+	+	+	+
2	-	-	-	-	-
3	+	-	-	-	-
4	·	·	·	·	·
5	-	-	+	+	+
6	-	-	-	-	-
7	·	·	·	·	·
8	-	-	-	-	-
9	·	·	·	·	·
10	-	-	-	-	-
11	+	+	+	+	+
12	+	+	+	+	+
13	·	·	·	·	·
14	+	-	+	+	+
15	-	-	+	+	+
16	+	+	+	+	+
17	+	+	+	+	+
18	+	+	+	+	+
19	+	+	+	+	+
20	+	+	+	+	+
21	+	+	+	+	+
22	+	+	+	+	+
23	+	+	+	+	+
24	+	+	+	+	+
25	+	+	+	+	+
26	+	+	+	+	+
27	+	+	+	+	+
28	+	+	+	+	+
29	+	+	+	+	+
30	+	+	+	+	+

	lac	Mal	Gal	Xyl	Arab
1	+	-	+	+	+
2	+	+	+	+	+
3	·	·	·	·	·
4	·	·	·	·	·
5	+	+	+	+	+
6	+	+	+	+	+
7	+	-	+	+	+
8	·	·	·	·	·
9	+	-	+	+	+
10	+	-	+	+	+
11	+	-	+	+	+
12	+	-	+	+	+
13	·	·	·	·	·
14	+	-	+	+	+
15	+	-	+	+	+
16	+	-	+	+	+
17	+	-	+	+	+
18	+	-	+	+	+
19	+	-	+	+	+
20	+	-	+	+	+
21	+	-	+	+	+
22	+	-	+	+	+
23	+	-	+	+	+
24	+	-	+	+	+
25	+	-	+	+	+
26	+	-	+	+	+
27	+	-	+	+	+
28	+	-	+	+	+
29	+	-	+	+	+
30	+	-	+	+	+

October 18, 1948.

Inoculate H72 fairly heavily into T(0) + T(B₁). Shaker.P19. No growth A20. Heavy growth in T(B₁); none in T(0).Is H72 B₁ -
" viable?

①. Streak out H72 on LacEMB, EMS, EMS'.

②. Plate out T(B₁) tube on LacEMS'; XylEMS'.P21. ①. On LacEMB: almost all lac- (2. Do. on XyloseEMB.
(i.e. most of the stock culture is segregated.).
A few + noted on EMS.②. 2 plates on LacEMB. 140 colonies. All lac-
EMS' - too small to readA22. - only noted on all plates, ~~EMS~~, EMS' lac + Xyl'A22. Pick single + colonies of H72 from ~~EMS~~ EMS Lac to T(0) tubes to -
a) resuscitate H72 and b) continue exp. Streak out on LacEMB from
T(0) suspension. Use #6.

LacEMB. (OK).

1 ✓
2 ✓
3 ✓
4 ✓
5 ✓
6 ✓

See 348.

Segregation of Mal, Gal, Ar.

Oct 21, 1948.

~~W478~~ W478 & W583 on Mal, Gal, Ar EMS.

Low yields!!

101-120 Gal+

121-123 Arabinose.

1-100 ~~Mal~~ Maltose.

} test on EMB Galactose + Arabinose

test on EMB Maltose.

D.G.

100 colonies picked from Mal, not readily scored. Only 39 Mal+
 Rechecks: 16, 25, 31, 50, 59, 87, 95, 99.

20 Gal+ colonies: All Gal+ Arab+. No heterozygotes.

3 Ar+. 2 Ar+ Gal+. 1 Gal- Ar+, -? Rechecks 121.
 1-8 on Mal EMS 9 on Ar EMS.

1	16	++	++
2	25	++	++
3	31		odd.
4	50	++	++
5	59	} no +s.	
6	87		
7	95	++	++
8	99	++	++
9	121	++	++

2 cdo from EMS tested.

Oct. 23, 1948

Squad H72 grown on T(0) (see 348) on EMS Lac + Xyl and
expose for 5-15 sec. of 348 for control.

Exp. n. 4. Centids inviable !!

October 23, 1948.

Grow H72 on T(10) — see 344. — dilute 10^{-7} and
plate on EMB; EMS lac; Xyl for — colonies.

n.g. Culture inviable.

Verifications

October 23, 1948.

See 345.

streak out streaks on media indicated.

- | | | |
|------|---|-------|
| | EMS Xyl. EMFLac | |
| H93 | swell + v. small + cols. | |
| H96. | n.g. | |
| H58 | n.g. | |
| 60 | n.g. | |
| 62 | + - cols.
EMS Ar(B ₁) | |
| H 85 | + cols? | |
| 86 | n.g. | |
| 88 | 1 - col. | |
| 93 | v. small + cols on EMF | |
| 94 | numerous + and - cols.
EMB. EMS Xyl. | |
| 95 | a few + and - cols. | Mal - |
| 96. | n.g. | |

93 → Xyl EMB, Lac EMB, Lac EMS.

~~Lac V~~ Xyl V; Ar - "Lac -"

85 Xyl EMB, Lac EMB, Ar EMB.

94 " "

Xyl V
Lac - (slow??)

62 Lac EMB; Ar EMB

Lac V Ar +

H52 Strk.

Lac V OK

H1 S.

OK

Resuscitation and presentation of 11 cases
Rechecked.

	EMF/Sec	EMD/Sec	EMB/Sec	EMBA/Sec	
H725	V				
H72n	mostly -				
H93			V	-	
H62	V, +, -			++	
H85	(V) ^{slow} +, -		het +; - ?		+ character here?
H85					
H22	++ , - (probably V)				
H94	-		+ weak? +		
H95	+ , - V			-	
H88			-	-	Significant? several months as EM's diff.
H70	++ , - +				
H11	V, +, - ++		++ , -		
H52	(+), -, (V)				
H71	(V), ++, - -				

Oct 24, 1948

W478 x W583

Ar, Mal + Gel EMS.

Arabinose: 24 + colonies. All ++

Saccharose: 28 + colonies All ++.

~~#33~~

Maltose: 50+ " All++ Check 4, 5, 18, 19, 43 (N2)

0.005% T₂
 N.A.

test on Lac & Mal
 (300)

58-161 Isln (-)

Oct 29. Inoculated to Perm. assay.

Oct 30 Irradiated 7 secs on Q.M.B. glu. plates

Oct 31 16 colonies picked seemingly glu (-)

checked out on C.M.B. glu.

Nov 1 four (4) apparently glu (-) streaked again on Q.M.B. glu

Nov 2 2 glu (-). checked on T₁. Both T₁

4 Shus

Nov 3	Tested on:	"N"	"Z"	"N"	"Z"
	Glu	neg	neg	pos	pos
	LAC	neg	neg	pos	neg
	MAL	neg	neg	neg	neg
	FRU	neg	neg	pos	pos
	MANNose	±	±	pos	pos
	RHAM	neg	neg	±	±
	Arab	neg	neg	pos	pos
	Gul	neg	neg	pos	±
	AgE	---	---	pos	pos
	Tell	neg	neg	neg	neg

15 Shus.

Nov. 8 Plated on K gluconate
 "N" "Z" 15 Shus
 ± slow slow ±