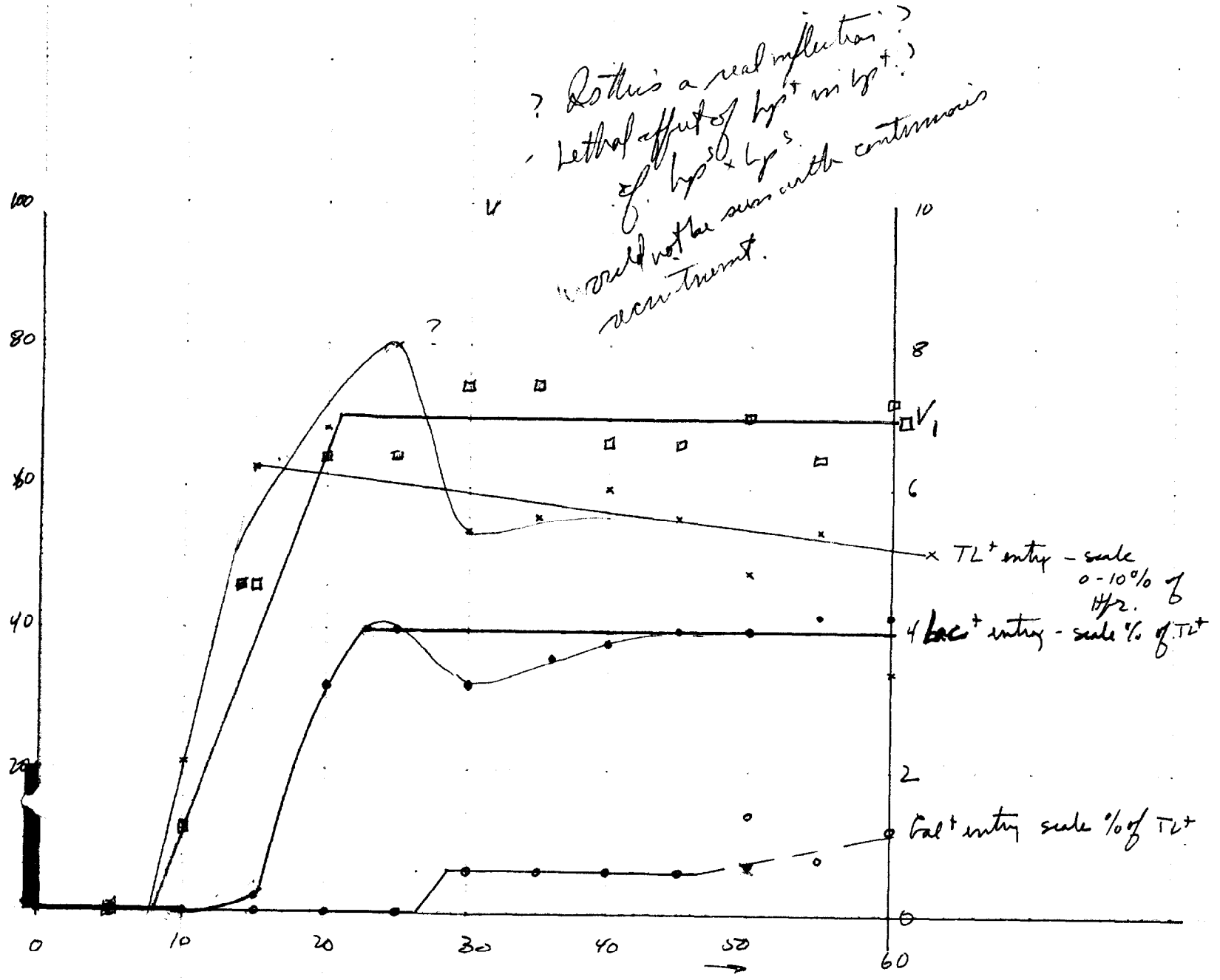
DATE: samples of 50 ml REF: 1401/2

	1	2	3	4	5	6	7	8	9	10
	Time	Tl+ % (Hfr)	T' (=) % (Tl+)	Lac % (Tl+)	Gal (2 Tl+)					
	0	0	–	–	–					
	5	0	–	–	–					
10	10	2.1	12	0	0					
	15	6.3	46	2	0					
	20	6.8	54	32	0					
	25	8.0	64	40	0					
	30	5.4	74	32	6					
20	35	5.6	74	36	6					
	40	6.0	66	38	6					
	45	5.6	66	40	6					
	50	4.8	70	40	14					
	55	5.4	64	42	8					
30	60	3.4	72	42	12					
40										
50										

1401/2

% of Lac, Gal, T_1
based on first samples of
50 colonies each.

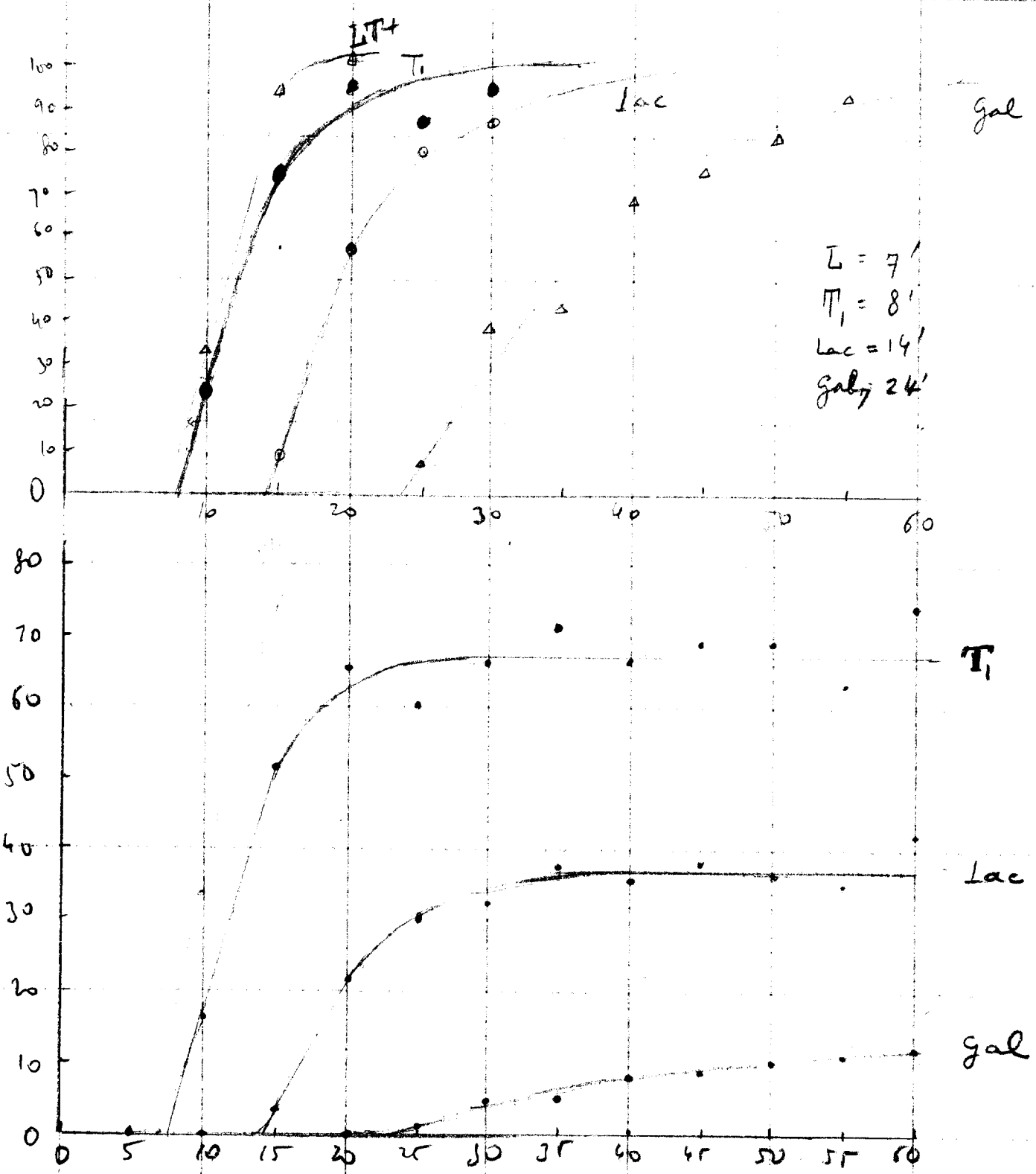


1401-2

DATE: REF:

1 time	2 Lac+ first 50	3 next	4 total	5 Gal+ first 50	6 next	7 total	8 T_1 first	9 next	10 total
0									
5									
10	0/50	0/12	0/62		0/12		6	4/12	10/62
15	1	6/142	7/192		0/142		23	76/142	99/192
20	16	20/92	32/142	0	0/88		32	59/88	91/138
25	20	28/110	48/160	0	1/108	1/108	32	63/108	95/158
30	16	18/56	34/106	3	2/57	5/107	37	33/56	70/106
35	18	19/49	37/99	3	2/49	5/99	37	33/49	70/99
40	19	33/95	52/145	3	9/95	12/145	33	63/95	96/145
45	20	18/50	38/100	3	6/50	9/100	33	36/50	69/100
50	20	12/38	32/88	7	2/38	9/88	35	25/37	60/87
55	21	16/56	37/106	4	8/57	12/107	32	35/56	67/106
60	21			6			37		

Time	% T_1	1-alleles Lac	Gal		% of asymptote T_1	Lac	Gal
0	1 -				–		
5	–						
10	16.1	0	0	10'	23.2	0	0
15	51.6	3.6	0		74.5	9.6	0
20	65.8	21.5	0	20'	95.0	57.5	0
25	60.1	30.0	0.93		86.5	80.2	7.7
30	66.0	32.1	4.67	30'	95.0	86.0	39.0
35	70.8	37.3	5.1			99.9	42.5
40	66.2	35.8	8.3				69.2
45	69.0	38.0	9.0				75.0
50	68.9	36.4	10.2				83.5
55	63.1	34.9	11.2				93
60	74.0	42.0	12				100
		114.4					
Asymp	69.5	37.4	12?				



Total data 1401/2

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19 May 26, 1958 REF: 1401/3

W3060, W3052, 1h cultures -
Spun, resusp. 4x & 2x resp. (♂ less turbid).

Mixture: 1 ml ♂ + 4 ml ♀.
After times: 10', 15', 20', 25', 30', 40'
sampled .2 ml and diluted in 10 ml H_2O (chilled).

Blended, and plated .05 and .05 1/10. (called 1, 2 resp.)

0': Plate recomb. control, .04 ♂ 1/50 and .04 ♀ 1/50.
Also, dilutions 1/10 from such suspensions (Resp. 1, 2).

Media: - D(B, Sm)
1 D(Proc L B, Sm)
2 D(Proc B, Sm).

Controls, .05 ml of dil 1/50 ♂, ♀ on media 1, 2.

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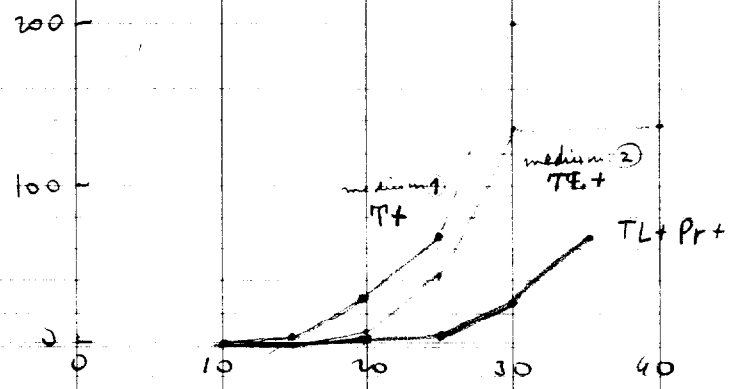
19 May 23rd, 1958 REF: 1401/5.

	Medium 0		Medium 1		Medium 2		
	del 1	del 2	del 1	del 2	del 1	del 2	del 3
10'	0	0	1	0	0	0	
15'	0	0	5	0	1	0	
20'	3	0	31	4	5	3	
25'	6	1	67	9	41	5	
30'	28.	4.		21	134.	17.	
40'	66	9			135	11	1
		del 3:					
0'	0	0	0	0	0	0	

Control, medium 1: 3 4 7 0, colonies
1 "

Media 0. D(B, fm)
1 : D(B, Prol Leu fm)
2 : D(B, Prol fm)



W 3052 is descendant from W 945
3064 W 583

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19

REF: 1401/3

	1	2	3	4	5	6	7	8	9	10
1	Medium (sel)		Gal⁺	Lac⁺	Ara⁺₂					
2		Time								
3										
4										
5	–	20'	0/4	1/4	4/4					
6	(TLPro⁺)									
7		25'	0/8	4/8	8/8					
8										
9		30'	0/32	21/32	31/32					
0										
		40'	8/75	39/50	72/75	L⁺	Pr⁺			
1										
2										
3	1	15'	0/5	0/5	1/5	1/5				
4										
5	(T+)	20'	0/40	0/40	8/40	3/40				
6										
7		25'	0/87	3/87	51/87	40/87	6/87			
8										
9		30'	0/24	4/24	14/24	12/20	4/24			
0										
1	2	15'	0/1	0/1	1/1		0/1			
2										
3	(TL+)	20'	0/9	0/9	1/9		0/9			
4										
5		25	0/51	5/51	49/51		7/51			
6										
7		30'	0/50	14/50	48/50		18/50			
8										
9		40'	2/50	18/50	48/50		22/50			
0										
1										
2										
3										
4										
5										
6										
7										
8										
9										
0										
1										
2										
3										
4										
5										
6										
7										
8										
9										
0										

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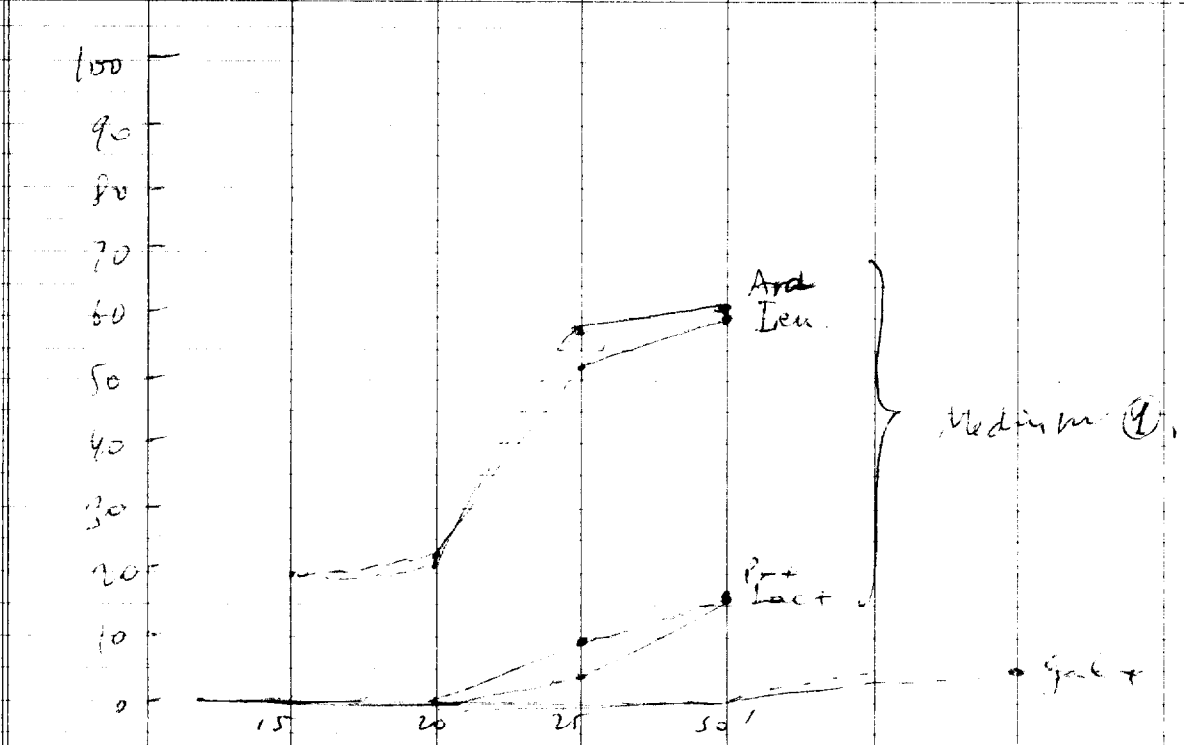
REF: 1401/3



F⁺ = W3052

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19 June 2, 1958 REF: 1401/4

Comparison between W3052, W3064.

ORC cultures of W 3060, W3064, W3052. Spun, resusp. at same conc.

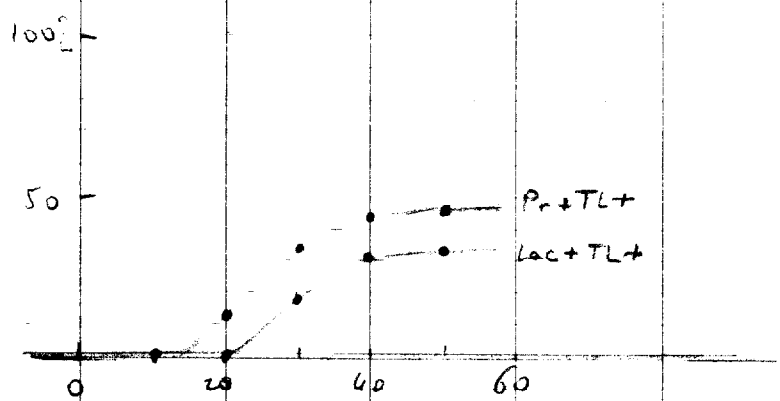
A: 2 ml W3060, + 4 ml W3052
B: " " " W3064
C: " " + 2 ml W3052 + 2 ml W3064.

At 10', 20', 30', 40', 50', 60'
0.01 sample diluted in 10 ml chilled water, blended,
and: 10', 20', 30', 40', 50' → 0.05 plated on 5' Gal/Fructose B,
30', 40', 50', 60' → 1/10 → 0.05 "

	A (all Ara+)* except on Ara−			B (all Ara−)# except on Ara+		Plate counts		
	Gal⁺	Lac⁺	Pr⁺	Gal	Lac	A	B	C
						dil	dil	dil
10	–	–	–	–	–			0
20	0/50	0/50	6/50	0/50	0/50	252	112	82
30				0/50	5/50	33	310	294 4 13
40	0/50	20/50	23/50	0/50	11/50	353	108	111
50	2/50	19/50	23/50	0/50	9/50	47 224	222 7	133 23
60						84 80	18 9	9 14

C

	Ara+			Ara−		Ara+/Ara−
	Gal⁺	Lac⁺	Pr⁺	gal⁺	Lac⁺	
20	0/25	0/25	3/25	0/25	0/25	25/25
30	0/27	5/27	9/27	0/23	3/23	27/23
40	0/23	7/23	10/23	1/27	2/27	23/27
50	1/22	10/22	14/22	0/22	6/22	28/22



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19 June 24 REF: 1401/5

Hfr$_1$ and Hfr$_2$ timing x 3052.

♀ # 3052 T$_1^R$.
♂♂ : 3936 (1895 Az^R), 3948 (3870 Az^R).
ORC, spun, resusp. in Penassay, 1 x conc ♂♂, ½ x conc ♀.

Mating mixtures : 1 ml ♂ : 9 ml ♀ (conc ratio 1:5).

Mating in flasks. Samples of 0.5 ml dil in 4.5 chilled water, blended, → A plated .05 on various media, B diluted 1/10 and plated. B dilutions only for times ≥ 15'.

Times : Hfr$_1$ cross : 0', 5', 10', 15', 20', 25', 30', 35', 40', 50'
Hfr$_2$ " : also 7½' and 13'.

Media : For Hfr$_1$ media 1 Sm Me B, L T
(all D(-)) 2 Sm Me B$_1$
3 Sm Me

For Hfr$_2$ media 4 Sm B, Prol Leu
5 Sm B, Prol
6 Sm B$_1$

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REF: 1401/5

	1	2	3	4	5	6	7	8	9	10
Temp	0	5	7½	10	13	15	20	25		30
1A	105	146		253		0	N.6.10³ *	(N200 large 10³ small)		∞
1B						[100-200 small]?	9 large 320 small	49 large N500 small)		(53 large N500 small)
2A	88	34		52		70 large N1000 small	155	N1000 large		N1000 large
2B						63	13	85		137 large
3A	8	6		9		26	40	18		44
3B						5	2	5		6
4A	245	320	N2000	N10.000	N10.000	N10⁴ *	N10⁴	t.m.t.c		∞
4B						N800 *	N2000 *	"		N2000 large & small
5A	355	172	296	N3500	N8000	N10⁴ *	N10⁴	"		∞
5B						N1000	N1000	"		N2000
6A	58	276	171	165	798	N1300	N6.10³ *	"		∞
6B						143	494	N1200		N2000

* large & small: small in great preponderance.
1A-20': large are of numbers as large as in 2A 20'.

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REF: 1401/5.

	1	2	3	4	5	6	7	8	9	10
	35'	40'	50'	Control	♀					
1A	∞	∞	∞		0,0					
1B	(large ~100 small ~500)	(large 100 small ~100)	(large ~200 small ~)		–					
2A	∞	∞	∞		–					
2B	~600	~600	~600		–					
3A	79	231	~492		0					
3B	7	14	79		–					
4A	∞	∞	∞		0					
4B	~2000	~2000	~2000		–					
5A	∞	∞	∞		0					
5B	~2000	~2000	~2000		–					
6A	∞	∞	∞		0					
6B	~2000	~2000	~2000		–					

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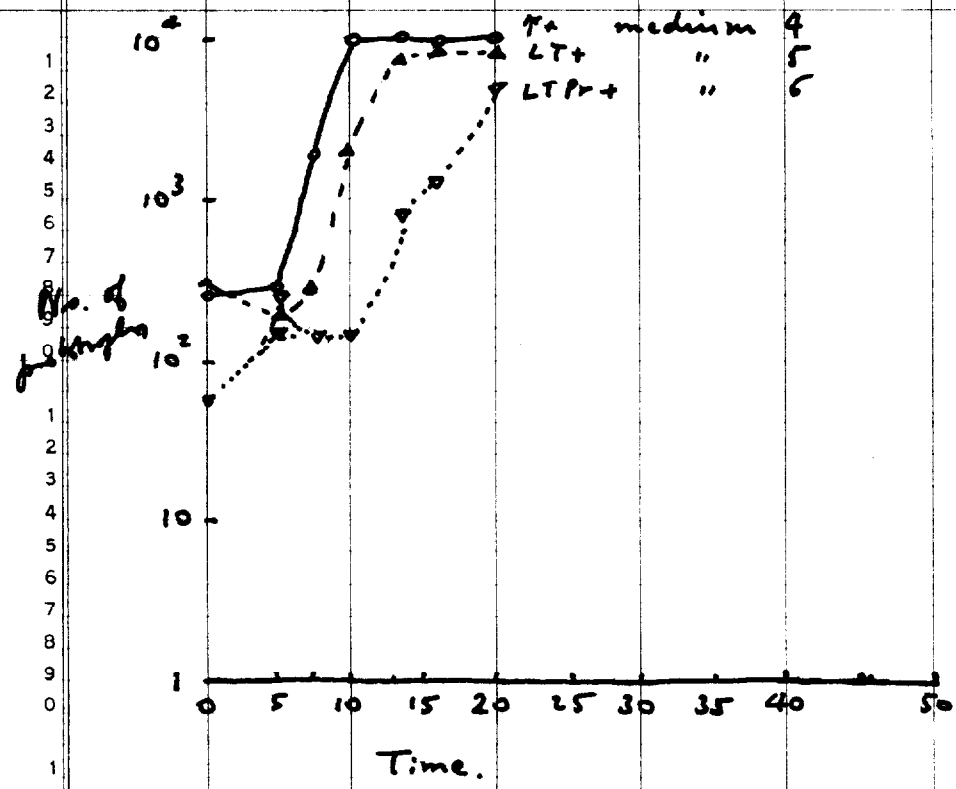
REF: 1401/5



Hfr_2

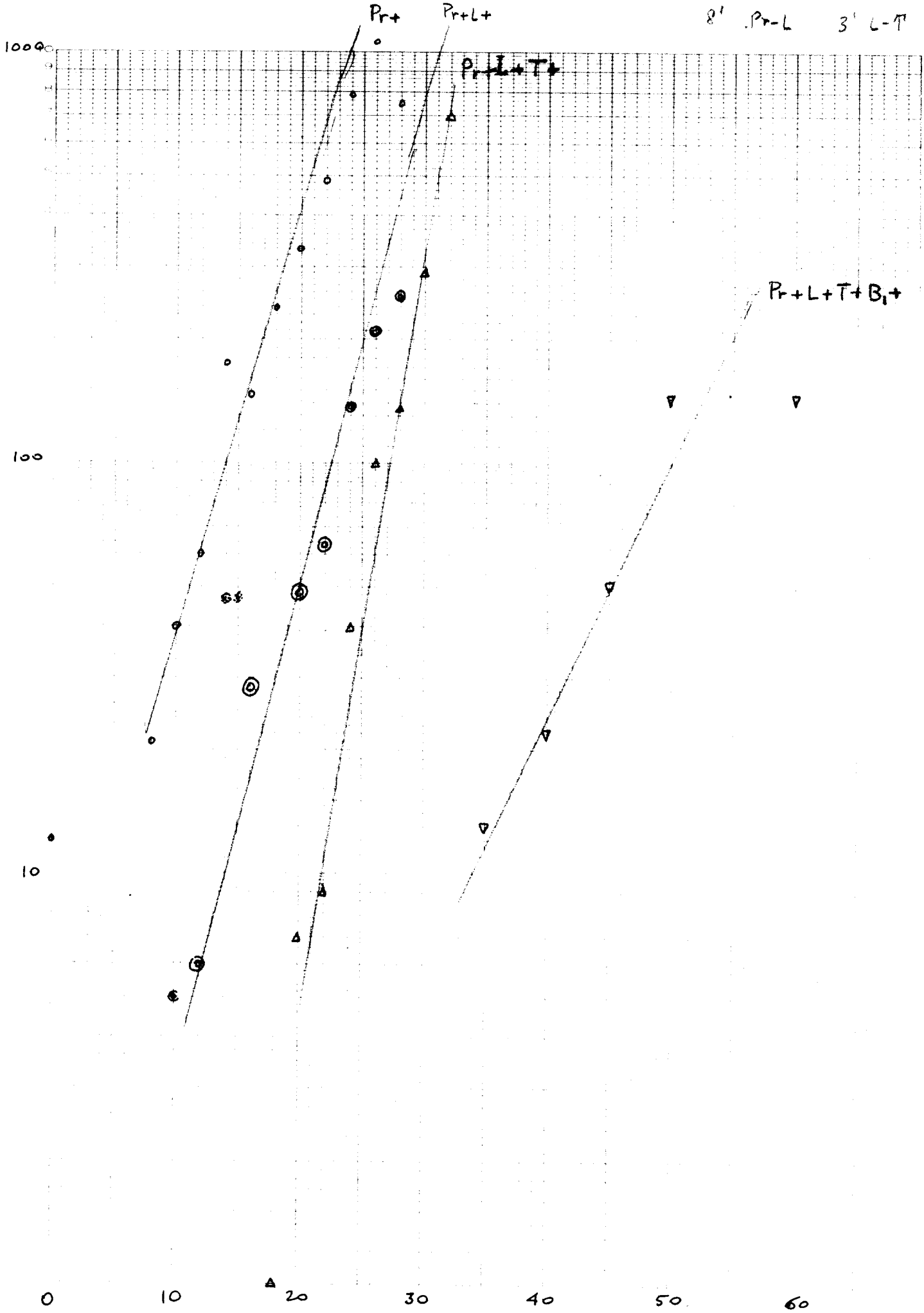
T-L : 2½ minutes 5' T 7½ L

L-Pr : 5' minutes 12½ Pr

Hfr_1 : <u>medium 1</u> Entry of Pr between 10' and 20'.
Repeat !:

<u>medium 2</u> Entry of LT probably between 15 or 20 & 25'. But explore 10-30' again

<u>medium 3</u> Entry of B_1 between 35' and 40' test for Xyl Meth.


Pr+
Pr+L+
8' Pr-L
3' L-T
Pr+L+T+
Pr+L+T+B1+
1000
100
10
0
10
20
30
40
50
60

TIMED MATING, Hfr_1 and Hfr_2

ORC Cultures of W 3936 (Hfr_1), W 3948, (Hfr_2) W 3052.

Spun, resuspended in equal amount of Penassay, and mating mixtures prepared in flasks with 1.5 ml ♂ and 6 ml ♀ and 2 ml fresh broth. Samplings: 0.2 ml at every time, dilution in 9.9 ml chilled water, .04 spread on plated (A) and the same for dilution (B) of 1/10 for Hfr_1 and 1/50 for Hfr_2.

Media:	1.	D-O	+	Sm	B_1	M	L	T	
	2.			Sm	B_1	M		T	for Hfr_1
	3.			Sm	B_1	M			
	4.			Sm		M			
	0.	B Lac		Sm					
	5.	D-O	+	Sm	B_1		L	Prol	for Hfr_2
	6.		+	Sm	B_1			Prol	
	7.		+	Sm	B_1				

DATE: Sat. June 28, 1958 REF: 1401/6

TIMED MATING, Hfr_1 x Hfr_2.

ORC Cultures of W 3936 (Hfr_1), W 3948, (Hfr_2) W 3052. Spun, resuspended in equal amount of Penassay SS, and mating mixtures prepared in flasks with 1.5 ml ♂ + 5 ml ♀ + 2 ml fresh broth. Samplings: 0.2 ml at every time, dilution in 9.9 ml chilled water, .04 spread on plates (A) and the same for dilution (B) of 1/10 for Hfr_1 and 1/50 for Hfr_2.

Media:		M	L	T	
1.	D-O + Sm B_1	Meth	Leu	Threo	sel for R_e
2.	Sm B_1	Meth		Threo	for PL
3.	Sm B_1	Meth			for Hfr_1
4.	Sm	Meth			for PTL
0.	B Lac Sm				for PLS,
5	D-O + Sm B_1		Leu	Pro	for Hfr_2.
6.	+ Sm B_1			Pro	
7.	+ Sm B_1				

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(space for counts)

1401/6

COUNTS

~~Medium 1~~ each column: +, −, ±

July 12 19 58

REF:

1 TIME	2 Ara	3 Ti	4 Az	5 MTL	6 (M) MTB	7 ML B1	8 TL B1	9 MTL B1	10 Σ
			(1) sm B, MLT		Hfr1				
~~[illegible]~~	~~[illegible]~~	~~[illegible]~~	~~[illegible]~~	~~[illegible]~~	~~[illegible]~~	~~[illegible]~~	~~[illegible]~~	~~[illegible]~~	~~[illegible]~~
16	13, 76, 0	76, 10, 3	13, 76, 0	1, 79, 9	9, 79, 1	9, 80, 0	14, 0, 75	10, 0, 79	89
18	15, 52, 0	45, 16, 6	15, 51, 1	1, 54, 12	14, 53, 0	12, 54, 1	15, 0, 52	15, 0, 52	67
20	6, 76, 0	66, 6, 10	12, 69, 1	6, 76, 0	6, 76, 0	6, 76, 0	6, 0, 76	6, 76, 0	82
22	6, 54, 0	48, 11, 1	6, 54, 0	0, 54, 6	6, 54, 0	6, 54, 0	6, 0, 54	5, 0, 55	60
24	6, 59, 1	49, 11, 6	12, 54, 0	1, 63, 2	4, 56, 6	3, 63, 0	3, 0, 63	3, 0, 63	66
26	12, 84, 0	73, 16, 7	17, 79, 0	0, 90, 6	8, 84, 4	6, 90, 0	5, 0, 91	5, 0, 91	96
28	5, 65, 0	55, 8, 7	8, 61, 1	0, 67, 3	4, 64, 2	3, 67, 0	2, 0, 68	3, 0, 67	70

	Ara	Ti	Az	MB1	MB1T	Σ
	~~Medium 2~~		sm B, MT		Hfr1	
20	42, 5, 0	42, 5, 0	43, 3, 40	25, 22, 0	47, 0, 0	47
22	43, 17, 0	3, 6, 51	51, 8, 1	18, 42, 0	60, 0, 0	60
24	71, 25, 4	6, 0, 94	98, 1, 1	37, 63, 0	100, 0, 0	100
26	92, 15, 0	13, 2, 92	104, 3, 0	48, 59, 0	107, 0, 0	107
28	80, 15, 5	4, 96, 0	99, 1, 0	58, 42, 0	100, 0, 0	100

	Xyl	Mtl	B1	M	MB1	Σ
	~~Medium 3~~		sm B, M		Hfr1	
0	0, 3, 0	0, 3, 0	3, 0, 0	1, 2, 0	3, 0, 0	3
10	0, 1, 0	0, 1, 0	1, 0, 0	0, 1, 0	1, 0, 0	1
14	0, 9, 0	0, 9, 0	7, 2, 0	3, 6, 0	9, 0, 0	9
16	0, 1, 0	0, 1, 0	1, 0, 0	0, 1, 0	1, 0, 0	1
18	0, 1, 0	0, 1, 0	1, 0, 0	0, 1, 0	1, 0, 0	1
20	0, 8, 0	0, 8, 0	8, 0, 0	0, 8, 0	8, 0, 0	8
22	0, 13, 0	0, 13, 0	11, 2, 0	0, 13, 0	13, 0, 0	13
24	1, 42, 0	0, 43, 0	41, 2, 0	2, 41, 0	43, 0, 0	43
26	0, 100, 0	0, 100, 0	100, 0, 0	1, 99, 0	100, 0, 0	100
28	0, 99, 0	0, 99, 0	99, 0, 0	0, 99, 0	99, 0, 0	99
30	0, 100, 0	0, 100, 0	100, 0, 0	0, 100, 0	100, 0, 0	100
35	0, 70, 0	0, 70, 0	70, 0, 0	0, 70, 0	70, 0, 0	70

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COUNTS

Each Column: +, −, ±

140116

July 12 1958

REF:

	1	2	3	4	5	6	7	8	9	10
			Medium 4		sm M		Hfr1			
1		Ara	Gal	Lac	T1	Az	M	−(Do)	Σ	
2										
3	20	2, 0, 0	0, 2, 0	1, 1, 0	0, 2, 0	2, 0, 0	2, 0, 0	1, 1, 0	2	
4										
5	30	2, 0, 0	0, 2, 0	2, 0, 0	0, 2, 0	1, 0, 1	2, 0, 0	0, 2, 0	2	
6										
7	35	15, 0, 0	0, 15, 0	13, 2, 0	0, 15, 0	15, 0, 0	15, 0, 0	9, 6, 0	15	
8										
9	40	25, 0, 0	0, 25, 0 ~~23, 2, 0~~	23, 2, 0 ~~0, 25, 0~~	0, 25, 0 ~~25, 0, 0~~	25, 0, 0	25, 0, 0	17, 8, 0	25	
0										
	45	47, 0, 0	0, 47, 0	40, 6, 1	0, 47, 0	47, 0, 0	47, 0, 0	31, 16, 0	47	
1										
2	50	100, 0, 0	0, 100, 0	93, 6, 1	3, 97, 0	100, 0, 0	100, 0, 0	36, 61, 3	100	
3										
4	60	100, 0, 0	0, 100, 0	89, 10, 1	1, 99, 0	100, 0, 0	100, 0, 0	41, 56, 3	100	
5										
6										
7			~~Medium~~ 5		sm B, LP		Hfr2			
8										
9		Gal	Ara	T1	Az	L B1	Pro B1	L P B1	Σ	
0										
	0	0, 1, 0	0, 1, 0	1, 0, 0	0, 1, 0	0, 1, 0	0, 1, 0	1, 0, 0	1	
1										
2	4	2, 5, 0	3, 4, 0	7, 0, 0	3, 4, 0	2, 5, 0	3, 4, 0	7, 0, 0	7	
3										
4	5	0, 16, 0	9, 7, 0	16, 0, 0	6, 10, 0	4, 12, 0	7, 9, 0	16, 0, 0	16	
5										
6	7	0, 10, 0	2, 8, 0	10, 0, 0	2, 8, 0	2, 8, 0	2, 8, 0	10, 0, 0	10	
7										
8	8	0, 6, 0	1, 5, 0	6, 0, 0	1, 5, 0	0, 6, 0	1, 5, 0	6, 0, 0	6	
9										
0	9	0, 3, 0	3, 0, 0	3, 0, 0	3, 0, 0	3, 0, 0	3, 0, 0	3, 0, 0	3	
1	10	0, 82, 0	7, 75, 0	82, 0, 0	3, 79, 0	2, 80, 0	4, 78, 0	82, 0, 0	82	
2										
3	11	0, 7, 0	5, 2, 0	7, 0, 0	4, 3, 0	0, 7, 0	5, 2, 0	7, 0, 0	7	
4										
5	12	0, 99, 0	14, 84, 1	99, 0, 0	5, 94, 0	0, 99, 0	17, 82, 0	99, 0, 0	99	
6										
7	13	0, 98, 0	26, 72, 0	98, 0, 0	15, 83, 0	0, 98, 0	35, 63, 0	98, 0, 0	98	
8										
9	14	0, 20, 0	13, 7, 0	20, 0, 0	8, 12, 0	6, 14, 0	11, 9, 0	19, 1, 0	20	
0										
1										
2										
3										
4										
5										
6										
7										
8										
9										
0										

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COUNTS

Each Column: +, −, ±

1401/6

July 12 19 58

REF:

	1	2	3	4	5	6	7	8	9	10
				Medium 6		sm B, P		Hfr2		
1		Gal	Lac	T1	Az	B1	B1 Rsl	Σ		
3	0	0, 1, 0	0, 1, 0	1, 0, 0	1, 0, 0	0, 1, 0	1, 0, 0	1		
5	5	0, 2, 1	0, 2, 1	3, 0, 0	2, 1, 0	1, 3, 0	3, 0, 0	3		
7	6	0, 1, 0	0, 1, 0	0, 0, 1	1, 0, 0	1, 0, 0	1, 0, 0	1		
9	7	0, 8, 0	3, 5, 0	3, 2, 3	6, 2, 0	5, 3, 0	8, 0, 0	8		
	8	0, 2, 0	0, 2, 0	1, 0, 1	2, 0, 0	0, 2, 0	2, 0, 0	2		
12	9	0, 10, 0	9, 1, 0	0, 2, 8	10, 0, 0	9, 1, 0	10, 0, 0	10		
14	10	0, 8, 0	0, 7, 1	7, 1, 0	3, 5, 0	1, 7, 0	8, 0, 0	8		
16	11	0, 2, 0	2, 0, 0	0, 0, 2	2, 0, 0	2, 0, 0	2, 0, 0	2		
18	12	0, 67, 0	1, 66, 0	56, 6, 4	37, 29, 1	2, 65, 0	67, 0, 0	67		
20	14	0, 30, 0	11, 19, 0	15, 6, 9	27, 3, 0	11, 19, 0	30, 0, 0	30		
22	16	0, 99, 0	1, 98, 0	68, 21, 10	72, 27, 0	6, 93, 0	99, 0, 0	99		
24	18	0, 6, 0	0, 6, 0	5, 1, 0	3, 2, 1	0, 6, 0	6, 0, 0	6		
				Medium 7		sm B1		Hfr2		
		Gal	Lac	T1	Az	Σ				
	0	1, 0, 0	1, 0, 0	1, 0, 0	1, 0, 0	1				
	10	1, 0, 0	1, 0, 0	0, 0, 1	1, 0, 0	1				
12	12	0, 1, 0	0, 1, 0	0, 0, 1	1, 0, 0	1				
14	14	0, 5, 0	5, 0, 0	4, 1, 0	5, 0, 0	5				
16	16	0, 30, 0	8, 22, 0	0, 2, 28	30, 0, 0	30				
18	18	0, 28, 0	7, 21, 0	5, 1, 22	28, 0, 0	28				
20	20	0, 81, 0	23, 57, 1	3, 49, 29	81, 0, 0	81				
22	22	0, 101, 0	41, 59, 1	7, 54, 40	101, 0, 0	101				
24	24	0, 11, 0	5, 6, 0	0, 7, 4	10, 1, 0	11				
26	26	0, 100, 0	57, 43, 0	7, 17, 76	100, 0, 0	100				
30	30	0, 8, 0	4, 4, 0	0, 4, 4	8, 0, 0	8				

DATE: March 24 REF: OPTIM. PULSE

13.15 : Broth cultures 3060, 3064 from water bath, saturated since 2 hrs, inoculated into warmed Penassay 0.75 + 7.5 ml → girello.

14.35 : 0.5 ml. # 3060
5 ml # 3064 } → prewarmed, → girello and also
5 ml broth
samples from parental cultures frozen and iced.

At time:	0'			2'	5'	10'	20'	60'
Dilution with warmed broth (with a 1 ml. pipette to prevent shearing)	0.2 + 0.8 (1/5)	0.1 + 0.9 (1/10)	0.1 + 9.9 (1/100)	0.1 + 9.9 (1/100)	0.1 + 9.9 (1/100)	0.1 + 9.9 (1/100)	0.1 + 9.9 (1/100)	0.1 + 9.9 (1/100)
Incubation time in rotator	60'	60'	60'	58'	55'	50'	40'	0'
Dilution previous to plating, with iced broth	(1/20) 0.5 + 9.5	(1/10) 1.0 + 9.0						
Amount plated on min St B_1	0.1 / 0.02	0.1 / 0.02	0.1 / 0.02	0.1 / 0.02	0.1 / 0.02	0.1 / 0.02	0.1 / 0.02	0.1 / 0.02

Iced parental broths:

#3064 → form 1/20 → .025 min St B_1
#3064 → broth 1/100 → water 1/100 → water 1/10 → .05 B Lac ; 341 ; same

#3060 → broth 1/100 → broth 1/10 → water 1/100 → .05 B Lac. : 17 ; 19
#3060 → form 1/20
broth 1/10 → .025 min St B_1
broth 1/10 → .025 / .025 min St B_1

DATE: March 28, 1957 – Summary REF: 1402 : PULSE

TIME	Gal+/tot.	2	Lac+/tot (including W)	4	T1'+/tot.	6	7	8	9	10
0'	4/31	12.9	4/31 (9/31)	29.0	21/31	68.0				
2'	5/57	8.8	21/57 (28/57)	49.2	39/57	68.4				
5'	12/81	14.7	40/81 (42/81)	52.0	68/81	84.0				
10'	13/79	16.4	33/79 (36/79)	45.5	61/79	77.2				
20'	11/65	16.9	31/65 (31/65)	47.7	46/65	70.8				
60'	5/50	10.0	15/50 (18/50)	36.0	37/50	74.0				
0' 1/5	5/70	7.2	15/69 (20/69)	21.7	48/70	68.6				
0' 1/20	6/52	11.5	19/52 (19/52)	36.5	36/52	69.2				

Conclusions. 5'-10' Pulse is an efficient procedure to raise No. of recombinants.

Dilution of 1/100 at least after pulse if further drainage must be avoided – segregation ratio unaffected, provided dilution is carried out with 1 ml. pipettes. 0.1 ml pipettes untrusted. Interruption on simple dilution in Exp. 1401 probably due to chilling, or plating.

Experiment 1403, not successful because of selection of F+ isolate from Hfr, must be repeated

1402 b

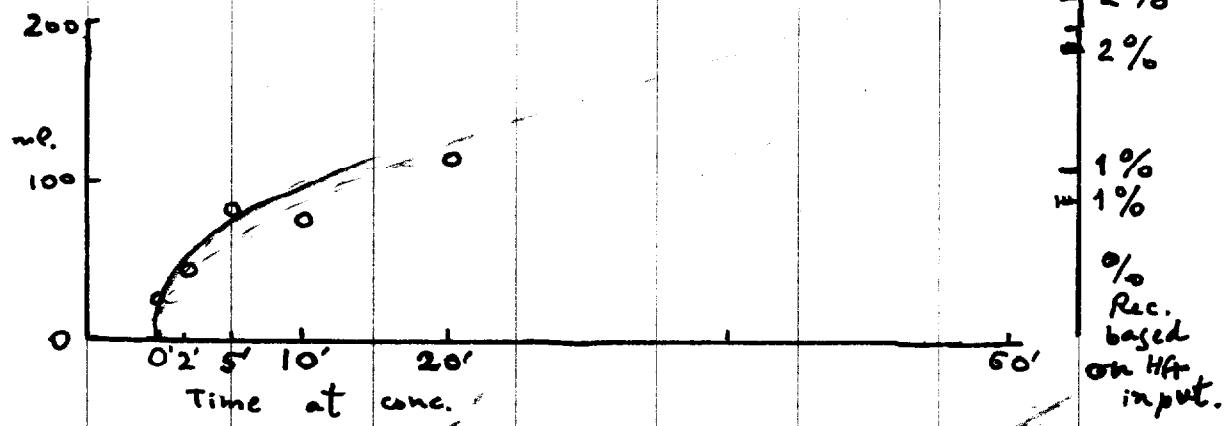
DATE: March 24. REF: PULSE.

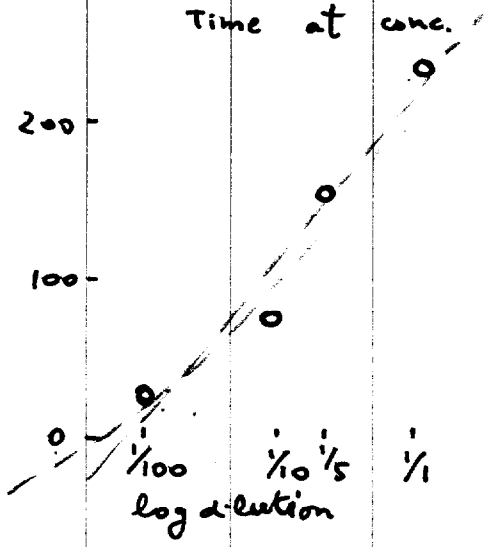
Plate counts.

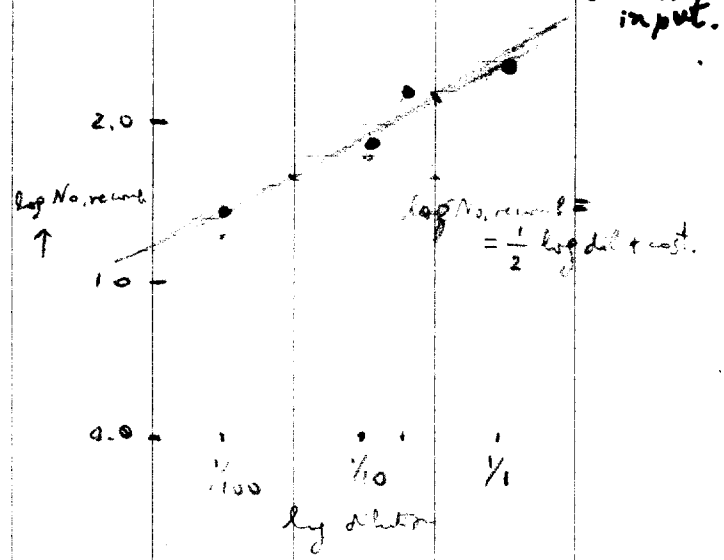
Amount plated	Time	dilution 1/100						Time 0' dil.			
		0'	2'	5'	10'	20'	60'	1/100	1/10	1/5	1/1
0.1		28 +	47	85	77	118	236 ‡	28 +	74	167	236 ‡
0.02		2 +	10	15	13	25	31 ‡	2 +	11	23	31 ‡

+, ‡ : same values, repeated

(Pulse)







MICROSC. COUNTS OF PARENTAL BROTHS:

3060 : 230×10^6/ml

3064 : 1.4×10^9/ml

INPUTS per ml : 3060 $11.5 \cdot 10^6$/ml Ratio Hfr/F- = 1:50

3064 $700 \cdot 10^6$/ml

REF: 1402-2

DATE: 22/4/58.

6 7 8 9 10

3h (1+10) to approx. saturation.
for prototrophy & Hfr-ness.
20 x respectively:

ml in 50 ml flask in water bath
0.05 to 50 ml prewarmed broth.
n plating (Hfr : F- = 1:2)
ne. susp. dil 1:3; exp. repeated
ove.

conc. suspensions dil. 1:3.3; exp.
ctly as above.

and kept at 0° until used.

broth } 0.1 & 0.02 ml
broth } min/StB1.

---

recombination: 1x suspensions
h and plated 0.05 + 0.05

ifficult by smearing (wet plates used)
ounts: 1 colony.

0,1	0,02 ml plated
250	44
400	63
1200	148

Note: counts unreliable

1 2 3 4 5

3060, 3064, cultures rotated
New 3060 isolate tested
Concentrated 10 x, and

10 x Exp : 0.2 ml + 0.2
for 1', then
30' inc. the

3x Exp 10x & 20x u
exactly as ab

1 x Exp. 3x & 6x
repeated exa

All 1:1000 dilutions chilled

10 x : 1/10 in
3 x : 1/3.3 in
1 x plated

For estimation of plate
diluted 1:1000 in brot

Plate counts: made di
Control of plate recombi

Conc 1 x
3 x
10 x

U.W.
MICROBIAL
GENETICS

19

REF:

	1	2	3	4	5	6	7	8	9	10
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