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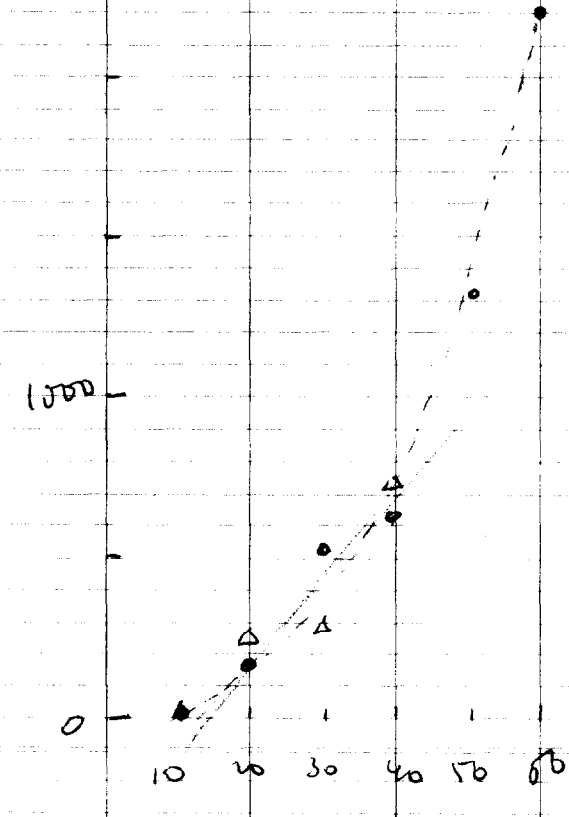
19

REF: 1408/6

	D (fm Δ,)		Sgal	
Hfr 13	A	B	A	B
	.05	.1		
10	22	104	21	
20	173	301	271	
30	522		297	
40	637		725	
50	~320			
60	~2200			



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19 May 19, 1958 REF: 1408/7

Hfr_{13} - Gal

Hfr_{13}, W3908. - 1 ml + 7.5 ml Pen, 1 hr rotation -

Two timed incubs. A. non pulsed in Penassay

B. Pulsed in Penassay, diluted in BGA.

B. 8 ml ♂ conc 2x, + 12 ml ♀ conc. 2x, in flask in waterbath. 2 ml samples taken at 15, 20, 25, 30, 35, 40' — Chilled, blended, plated .05 on 5 Gal Bi+Sm (some with Mg)

A. ♂ & ♀ conc 30 x in chilled Penassay (0.3 ml) then 0.2 of each mixed, incubated ½ 1'30", diluted to warmed BGA 1/100 (.2 to 20 ml).

Samples taken at one	15'	20'	25'	30'	35'	40'
amount	2	2	2	1	1	1
chilled diluent	-	-	1	1	2	3

Blended, plated .05 on same medium as before.

Plate recombinants. ~~.05~~ .01 of 30 x conc. mix + .9 water, of each pair .025 + .025, and .05 + .05 —

Plate counts	0	15	20	25	30	35	40
(A)	1	56	118	174	386	490	589
	2	83	126	176	208	497	541
	3		153	183	291	578	553
				213	317	521	571
		69.5	136	186	300	521	588
	x dilution	69.5	136	279	600	1563	2352

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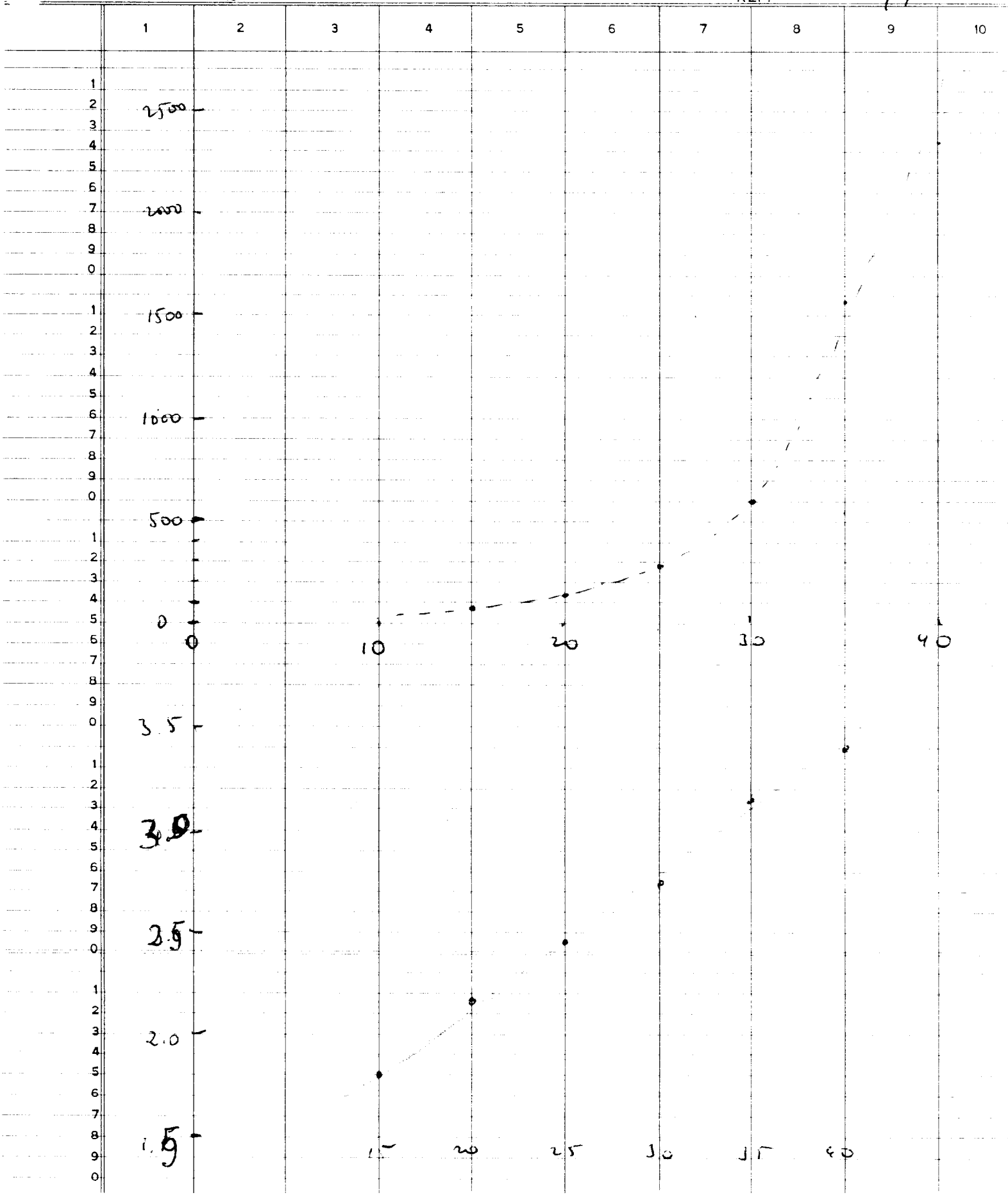
REF: 1408/7



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19 May 21, 1958 REF: 1408/8

Gal timing with HfR_2

W 3870 from frog

W 3908 from recently isolated colony.

1 + 7.5 ml Pen, rotated 1 hr, spun, resuspended to same vol.

Mating mixture 5 ml ♂ + 15 ml ♀ at 37° in flask

Sample 1 ml every time, dilute with 1.5 ml chilled ~~water~~ broth & blend (A). and also further dilution (B).

times	17'	20'	23'	26'	29'	32'	35'	38'	41'
dilution B	1+1	1+2	1+3	1+4	1+5	1+6	1+7	1+8	1+9

(A) + H_2O

Plate .05 on minimum B_1 -

Plate recombination: suspensions diluted 1 + 1.5 water, .025 of each parent on plate.

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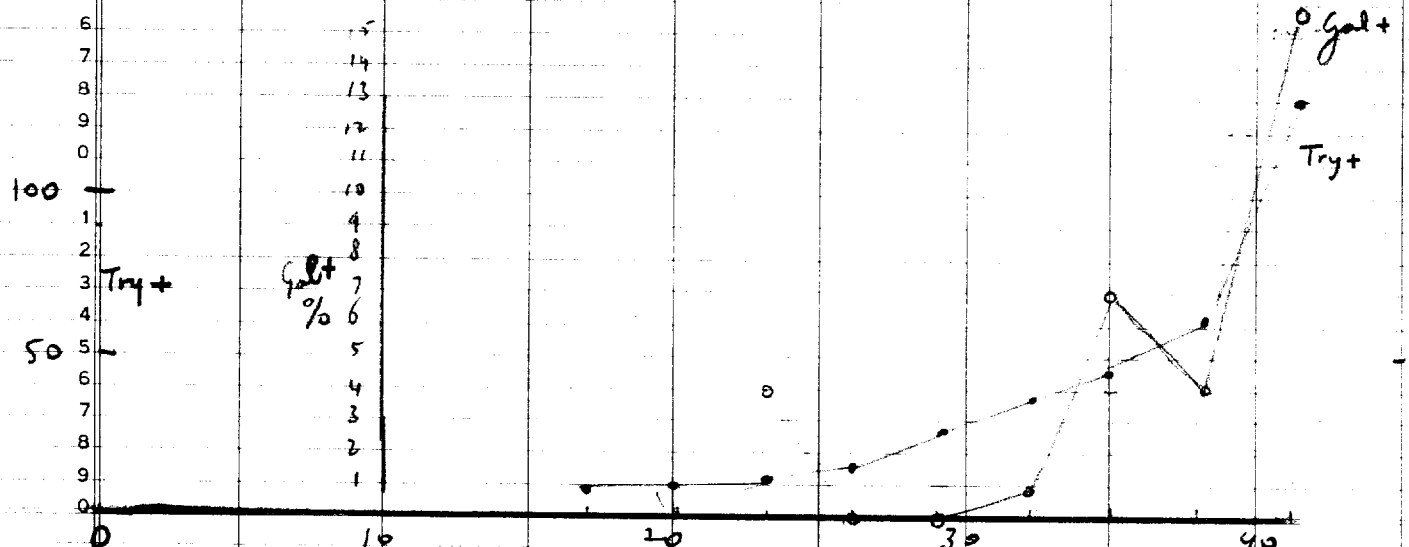
F2

23/5 19 53 REF: 1408/8

D (Sm B,

		A	B	ave	Gal A Gal+	Gal A TOT	Gal A %
17	10, 9	0, 4	9.5	–	–		
20	13, 12, 12	0, 3	10	0	3		
23	8	0, 0	11	1	25	4%	
26	19	1, 0	16.6	0	31	0	
29	31, 26	1, 0	27.2	0	52	0	
32	34, 39	0, 3	36.6	1	110	0.9	
35	39, 62	5	45.2	9	130	6.9	
38	51, 70	0, 1	60.7	5	122	4.1	
41	138	2, 5	130	43	275	15.6	



Two modes of entry ?

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19 May 24, 1958. REF: 1408/9

gal timing with Hfr_2

Identical to 1408/8 except that series B has been limited as follows:

Series B:	44'	47'	50'	
	1+1	1+2	1+3	(A) + H_2O
	1/2	1/3	1/4	dilution.

and timing: 10', 44', 47', 50' have been added

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19 May 26, 1958 — REF: 1408/9

	1	2	3	Counts (5^{40}) 4	5	6	7	8	9	10
1	Time									
2		D							S	
3										
4	0	43, 28,		Dilution 1/2.5: 13 colonies						
5										
6	10	10, 15, 21, 15						20, 26, 27, 19		
7										
8	17	33, 44, 30, 50						19, 33, 81, 81		
9										
0	20	53, 51, 50, 51						33 26 29		
1	23	58, 56, 66, 56						45 42 52 31		
2										
3	26	72, 98, 82, 92						58 60 61 43		
4										
5	29	89, 97, 92,						47 45 45		
6										
7	32	108, 117, 121						55 106 85		
8										
9	35	123, 141, 138, (110)‡						94 138 115		
0										
	38	150 216, 200						152 161 150		
1										
2	41	322, 335, 360, 320						284/47 234/46 290/58 288/36		284/47 | 360/256
3		(*)								
4	44B	253, 278.	('/2)	44A	: 443, 514			A. 386, 411		
5										
6	47B	361, 500		47A				B: 287, 262		
7										
8	50B	580, 620						B 492, 525		
9										
0										
1										
2										
3		‡ agar surface scratched								
4										
5		*) inversion of A and B								
6										
7										
8										
9										
0										
1										
2										
3										
4										
5										
6										
7										
8										
9										
0										

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19

REF:

	1	2	3		4	5	6		7	8	9	10
		D				Sgal						
	Time	Σx	n	dil	$\bar{x} \times dil$	Σx	n	dil	$\bar{x} \times dil$			
8	10	61	4	1	15.25	90	4	1	< 22.50			
10	17	157	4	1	39.25	114	4	1	< 28.50			
12	20	205	4	1	51.25	88	3	1	< 29.33			
13	23	236	4	1	59.00	170	4	1	< 42.50			
15	26	345	4	1	86.25	222	4	1	< 55.50			
17	29	278	3	1	92.67	137	3	1	< 45.66			
19	32	346	3	1	115.33	246	3	1	< 82.00			
20	35	512	4	1	128.00	347	3	1	< 115.67			
22	38	566	3	1	188.67	467	3	1	< 155.67			
24	41	1337	4	1	334.25	1360	5	1	< 272.00			
26	44	531	2	2	531.00	797	2	1	< 398.5			
28	47	861	2	3	1291.5	549	2	3	< 823.5			
30	50	1200	2	4	2400.00	1017	2	4	< 2034			

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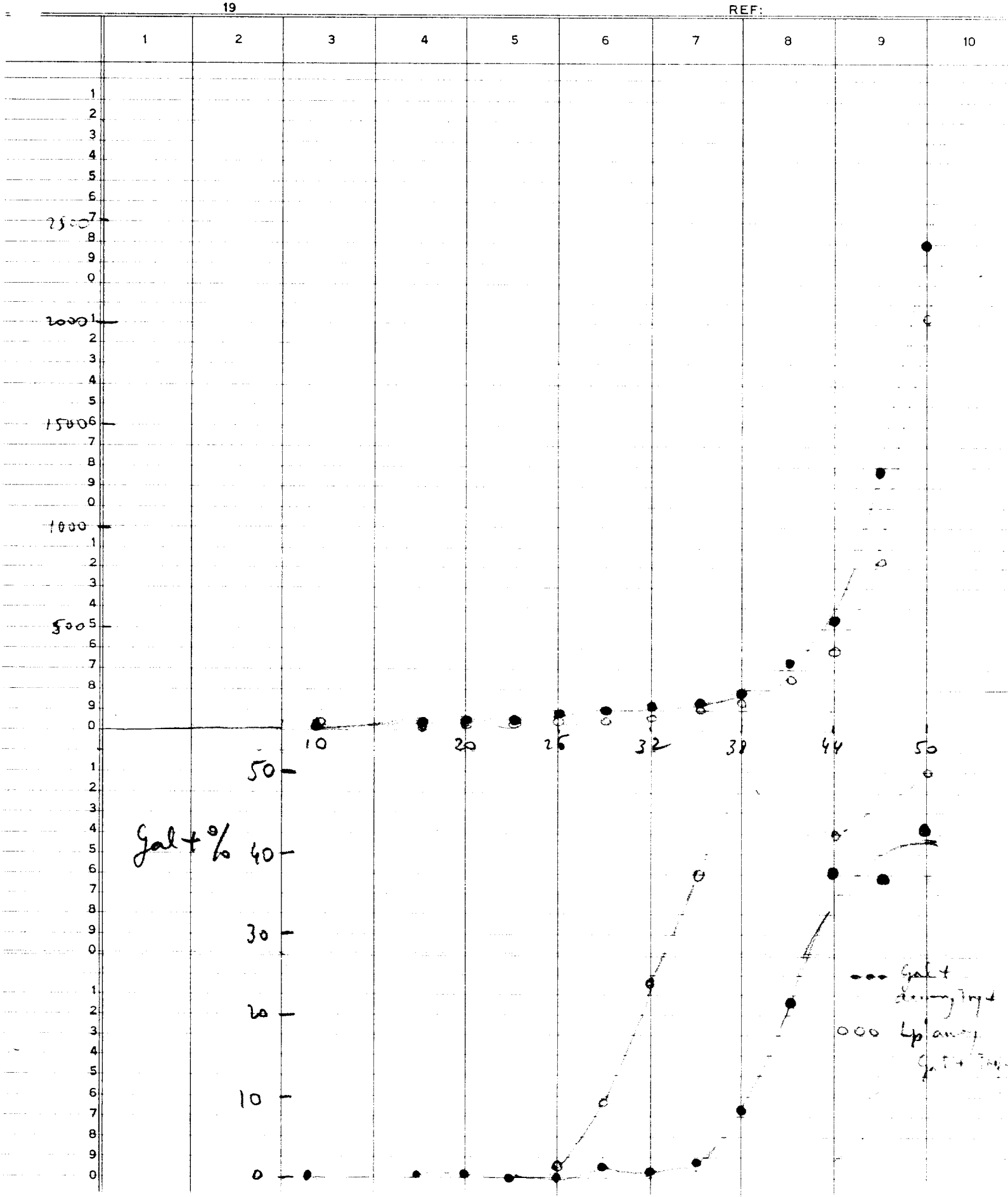
1498-9

19

REF:



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19

REF: 1608/9

	1	2	3	4	5	6	7	8	9	10
1		20	29	32	35	38	41	44	47	50
2										
3										
4	Gal +									
5	Con D med	± -	-	-	1.8	17	17.4	37.9	36.4	43
6										
7										
8	4 hr incub									
9	Gal + on	1.9	9.6	24.1	37.5	50	42.5	50	47.5	50
0	M Trg									
1										
2										
3										
4										
5										
6										
7										
8										
9										
0										
1										
2										
3										
4										
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0										

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1958 May 31st REF: 1408/10.

Gal_2 timing - Hfr_2

W 3870 & W 3959 - (Gal_2 Try) -

Same as 1408/8 and 1408/9 -

1h15' cultures (1 ml + 7.5 ml) - W 3870 from old broth in frig. - W 3959 recent (Rotated overnight) -

Spun, resuspended to same conc., mixed 5 ml ♂ + 18 ml ♀ in flask -

Sampled 1 ml at times: 23, 26, ... 59. + 1.5 ml distilled water

From 41' onwards also dilutions prepared (B Series).

	41	44	47	50	53	56	59	
Series A	1/2.5	1/2.5	1/2.5	1/2.5	1/2.5	1/2.5	1/2.5	blended → plated undiluted →
B*	2	3	6	12	20	50	100	

*dilution with respect to A

Plating: .05 on { D-0 B, Str
S Gal " "
M Gal " " Try.

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♀ 3959 Gal_2^- Tr^- 4^+

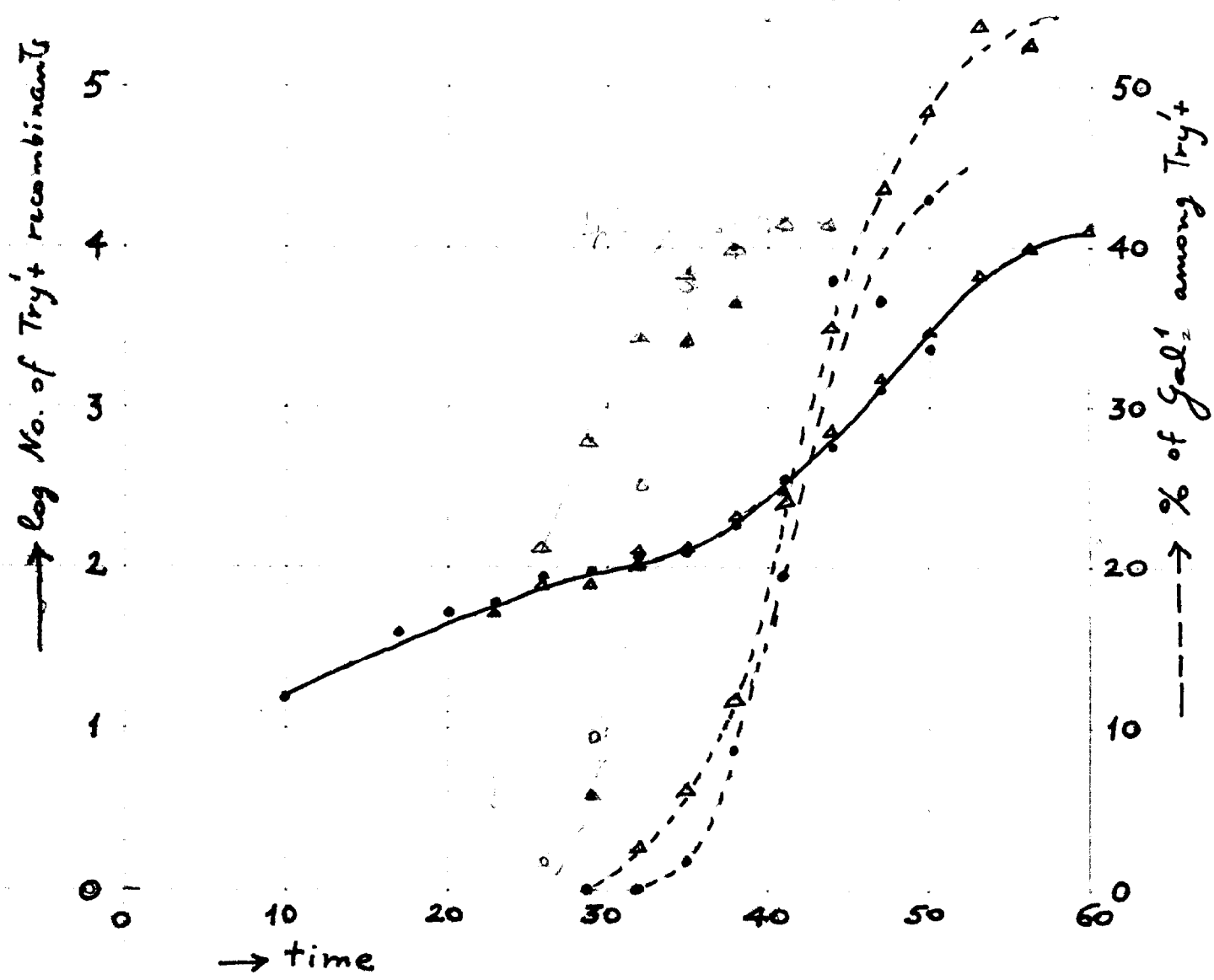
♂ 3870 2^+ Tr^+ 4h

2 June 1958 REF: 1408/10

	1	2	3	4	5	6	7	8	9	10
		D(B,fun)		Average x all	5' Gal fun B, Gal+	5' Gal fun B, tot	Sum	Ave	Gal %	
	0	8,(39)			3					
	23	61, 56, 40		52.3	1	32	32	32	3.12	
	26	62, 96, 79,		79.0	3, 9, 2	83, 90, 57	230	76.7	6.08	
	29	97, 63,		80.0	0	46	46	46	0.	
	32	108, 146		127	4, 3	55, 93	148	74	2.7	
	35	128 (162*), 115		135	2, 9	91, 90	181	90.5	6.1	
	38	230, 220, 165		205.0	24, 11	150, 148	298	149	11.7	
	41 B	139,	x 2	278	27, 19, 36	130, 106, 105	341	227	24.0	
	44 B	273, 217,	x 3	735	57	162	162	486	35.1	
	47 B	255	x 6	1530	88, 95	200, 219	419	1257	43.6	
	50 B	251	x 12	3012	81, 69	162, 147	309	1854	48.5	
	53 B	(333)*, 317	x 20	6500	121, 117	217, 225	442	4420	53.8	
	56 B	164, 236	x 50	10 000	47, 72	98, 130	228	5700	52.1	
	59 B	136,	x 100	13600	92, 72	143, 141	284	14200	57.7	

( ) clumps.

( )* contaminated agar



Expts: 1408/9 •••• Gal_6
1408/10 ▲▲▲▲ Gal_2

Final yield of $\begin{matrix} Gal^+ Try^- \\ Gal^- Try^+ \end{matrix}$ } 3 × 10⁻⁶ of the input

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19 REF: 1408/10

	1	2	3	4	5	6	7	8	9	10
1										
2		M gal Try fm 3/11 counts.				(after 2½ days, incub.)				
3										
4										
5		On the early plates, only small zyg gal+.								
6										
7		" " late " also big colonies observed.								
8										
9										
10	Time	gal+ (small)		(long) only	small (20 hour) colonies					
	23		11 *3						6 X objective 10 fields 6 [illegible]	
1	26		58, 122 *50							
2	29		502,	1		49, 67, 40			33, 43, 39	11, 11, 5
3	32			3, 0		179, 147			198, 162	56, 33
4	35			4 2		345				67, 120
5	38			19, 26						175, 82
6	41 B			39, 33						104, 13
7	44 B			82, 62						52, 79
8	47 B			100 **						77, 113
9	50 B		*	128						49, 60
10	53 B		*	160, 95					146	43 29
11	56 B		*	61, 112					50, 29	14 5
12	59 B		*	105, 90					28, 30	10 8
13			* real 50 taken							
14	0'		12,							
15										
16	** From 47 B onwards counts of "big gal+"s are different because of									
17	different appearance from smaller ones due to overcrowding.									
18	Comparison between various methods of counting small colonies									
19										
20		Total count	20 fields (×12)	10 fields 3x (×18)	10 fields 6x (×36)					
21	time									
22	29	502	624	738	684					
23	32	—	1955	3420	3382					
24	56 B	—	—	855	723					
25	59 B	—	—	552	684					
26										
27										
28										
29										
30										

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19

REF: 1408/10

	1	2	3	4	5	6	7	8	9	10
		(Average of duplicates)								
1										
2	Time	Gal + Strep	Gal + Trp+							
3						Ab Gal Trp+ = value				
4						expected Gal + Trp+ (*)				
5										
6	0	12	-							
7										
8	25	14	-							
9										
10	30	115	-			=				
11	32	702	1			0.4				
12										
13	35	2719	1.5			3.4				
14										
15	35	7055	3			8.2				
16										
17	38	9728	23			24.0				
18										
19	40	18240	39	x 2		66.7				
20										
	44	14934	210	< 3		258				
21										
22	47	43320	600	x 0		667				
23										
24	50	31008	1536	x 12		1460				
25										
26	53	57700	2550	x 20		3497				
27										
28	56	36100	4300	x 50		5210				
29										
30	59	58400	7700	x 100		7847				
31										
32			* Interpolated from: (Trp+) × (50 Gal + average Gal+)							
33										
34										
35										
36										
37										
38										
39										
40										
41										
42										
43										
44										
45										
46										
47										
48										
49										
50										

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19 June 3 19[illegible] REF: 1401/10

	1	2	3	4	5	6	7	8	9	10
		4p⁻ 1R+		4p⁻ Tr t			4p⁻ Tyl + Try-			
1	T1 ax	sensy Strd		sensy Sal+						
2										
3	23	-		33.	5/15		12/50			
4										
5	20	-			3/3	20.0	10/50			
6										
7	29	-				0.0	3/50			
8					5/6					
9	32	-				20.0	10/50			
0										
	37	-			14/15	34.0	17/50			
1										
2	32	1.0 %	1/99		47/50	36.6	15/41			
3										
4	41	3.4 %	3/78		128/138	38.4	19/30			
5										
6	21	3.0 %	4/132		50/81	53.0	14/34			
7										
8	17	2.0 %	2/99		90/92	53.3	16/30			
9										
0	5	2 %	2/96		19/19	59.5	25/30			
1	2	4.2 %	4/90		23/31	74.5	35/47			
2										
3	2	2 %	0/42		10/10	79.2	38/48			
4										
5	2		3/4		81/93	74.0	37/50			
6										
7										
8										
9										
0										
1										
2										
3										
4										
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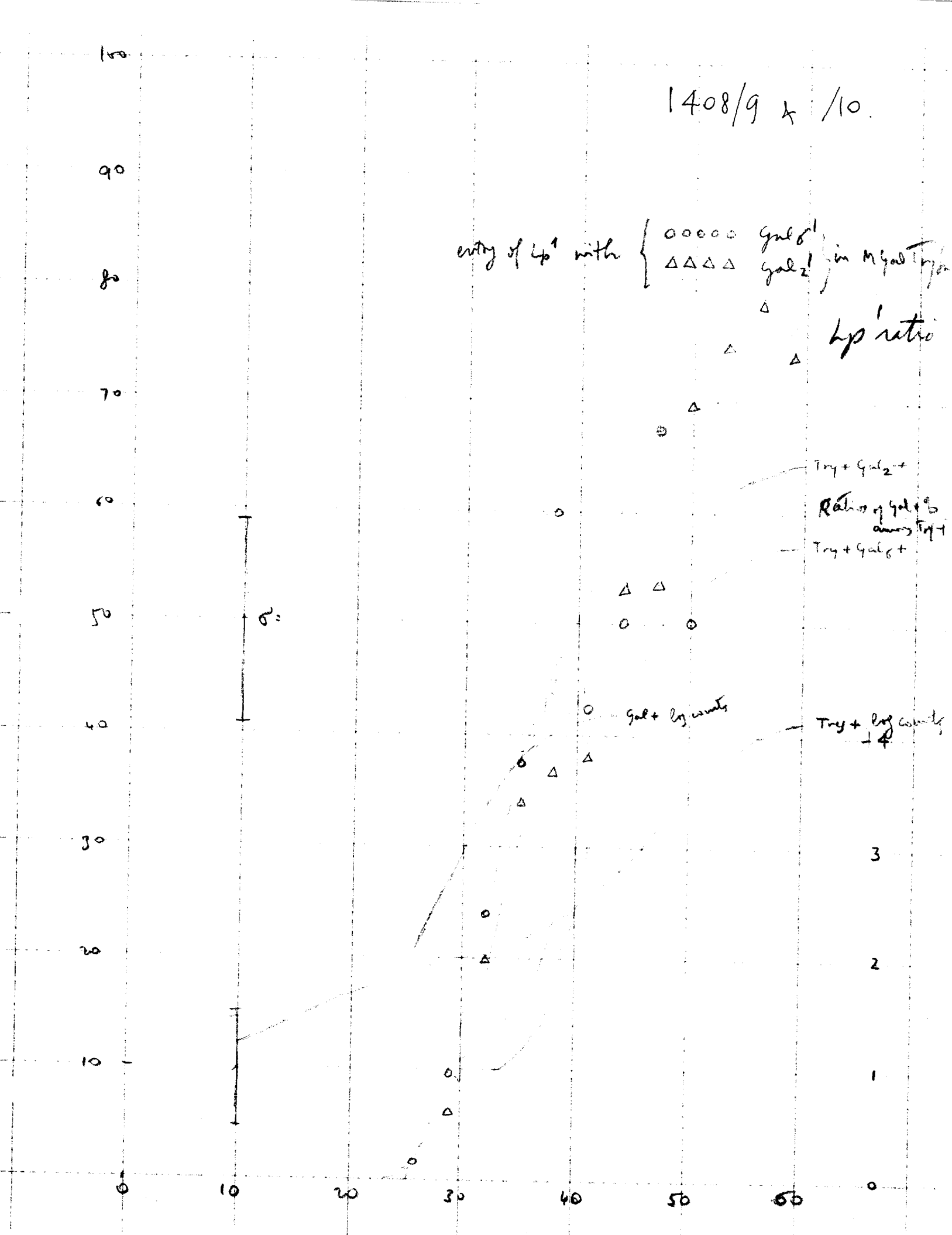
1408/9 & /10.
entry of Lp' with { ○○○○○ Gal δ' / △△△△ Gal₂' } in M Gal Try
Lp' ratio
Try + Gal₂ +
Ratio of Gal + δ among Try +
Try + Gal δ +
σ
Gal + log counts
Try + log counts
3
2
1
0
10
20
30
40
50
60
70
80
90
100

19 June 10, 1958. REF: 1408/11

Hfr, TIMING OF GAL, LP, Try-

ORC W3953, W1895: 1 ml + 7.5 ml Penn 1^h vol.,
spun, resusp. each in 2 ml = 4 × conc.
Mixture: ♂ : ♀ = 1 : 1.

Sampled 1 ml at times: 10', 15', 20', 25', 30', 35', 40', 50'
and diluted 1/2.5 by addition of 1.5 chilled water.
Blended and plated .05 ml. on 3 media.
Sampled .5 ml at times: 60', 70', 80', 90', 100', 120', 150'
and diluted 1/3 by addition of 1 ml chilled water.
Blended and plated.

Media: M Gal Sm Meth Try
S' Gal Sm Meth
D Glu Sm Meth

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19 June 12 1958. REF: 1408/11

Time	S Gal from B, Meth. total count	S Gal from B, Meth. Gal+	D from B,	M Gal from B, Meth Try only Gal+
0	200	1	100, 139	0
10	183	0	310	0
15	213	0	N 320	0
20	172	0	Sm, 233	0
25	178	0	237, 234	0
30	192	1	N400, 209	0
35	269	0	275, 230	1
40	154	1	262, 230	contam 1, 1
50	142	1, 3	284, 140	3, 3
60	144	2, 0	milling	4, 1
70	Sm	0, 4	122, 181	3, 0
80	116, 126	3, 1	Sm, 196	1, 4
90	165	6, 6	191, 198	4, 4
100	138	5, 4	176, 150	7, 0
110	140, 80	7, 6	N350, 208	6, 6
120	140, 143	6, 5	223	2, 3
150	576, 599	68, 80	410	73, 64

From D from B, related on B Gal:

	total	Gal+	tot
0	45, 27	0, 0	0/72
30	51, 59	0, 0	0/110
70	32, 25	1, 1	2/57
90	57, 69	5, 2	7/128
120	48, 32	4, 0	4/80
150	85, 77	13, 11	24/162

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19 June 14, 1958. REF: 1408/12

Hfr timing of gal, Try.

ORC cultures, W 1895, W 3953. 1 ml + 7.5 ml 1% 3/4 nut. Spun, resusp. in chilled broth 2 ml each tube (4x), mixed 6+6 ml (A) and 2+2 ml in another tube (B).

(A): sampled at 0, 30, 60, 90, 120, 130, 140, 150, 160, 170, 180, 210, 240
0.5 ml + 2 ml chilled water, blended, plated .05 on
M Gal Sm Meth Try.
D Sm Meth
S Gal Sm Meth
and also .05 on D Sm B, Meth powered.

(B) blended at 60', 120', 180' and sampled at 120', 150', 180', 210', 240'

0': From mixture in ice bath, dilute 1:5 in water as the others.

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1408-12

1895 ♂ M
3953 ♀ T

19 June 15, 1958. REF: 1408/12

	1	2	3	4	5	6	7	8	9	10
		D fm B_1		Pooled D fm B_1 mate		B4 not B together				
1	Time	non blend	Blended	not Bl	B	Gal+ fm B_1 Gal+	Gal-			
2										
3										
4	0	0, 0		1, 0	1, 0	0				
5										
6	30	7, 6		4, 5			1			
7										
8	60	7, 9		1, 4			1			
9										
0	90	9, 10		6, 4		1	5, 1			
1	120	7, 10	15, 9	5, 4	4, 2	1, 3, 3	4, 7, 10			
2										
3	130	6, 6		0,	3,					
4										
5	140	10, 5	8	1,		0,	3			
6										
7	150	8, 4	6, 8	4,	5, 10, 6					
8										
9	160	15, 15		5, 9						
0										
	170	11, 18		6, 14						
1										
2	180	7, 9	19, 17	2, 13	10, 4	2, 0	7, 3			
3										
4	210	15, 11	9, 11	~~05~~, 18	~~07~~, 7					
5										
6	240	24, 23	11, 29	not made.		4, 1, 2	8?, 7, 14?			
7										
8										
9										
0	W1895	0				0				
1	3953	0				1				
2										
3										
4		Counts too low — Experiment discarded								
5										
6		without further testing.								
7										
8		Lack of late entry due to what?								
9										
0										
1										
2										
3										
4										
5										
6										
7										
8										
9										
0										

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19 June 16 REF: 1408/13

	1	2	3	4	5	6	7	8	9	10
			Gal+	Mgal sm B1		Gal-				
			3-911			3172				
	Time									
	0		1			1				
	22		1			10				
	24		2			6				
	26		7			16				
	28		10			11				
	30		23			38				
	32		30			59				
	34		23			75				
	36		110			63				
	38		137			69				
	40		153			138				
	42		193			203				
	44		281			72				
	♀		0			1				
	♂		0							

Partial reversion to Gal+ 1/3870

2 Gal+ clones out of 12 on indicator tests

19 June 16, 1958. REF: 1408/14

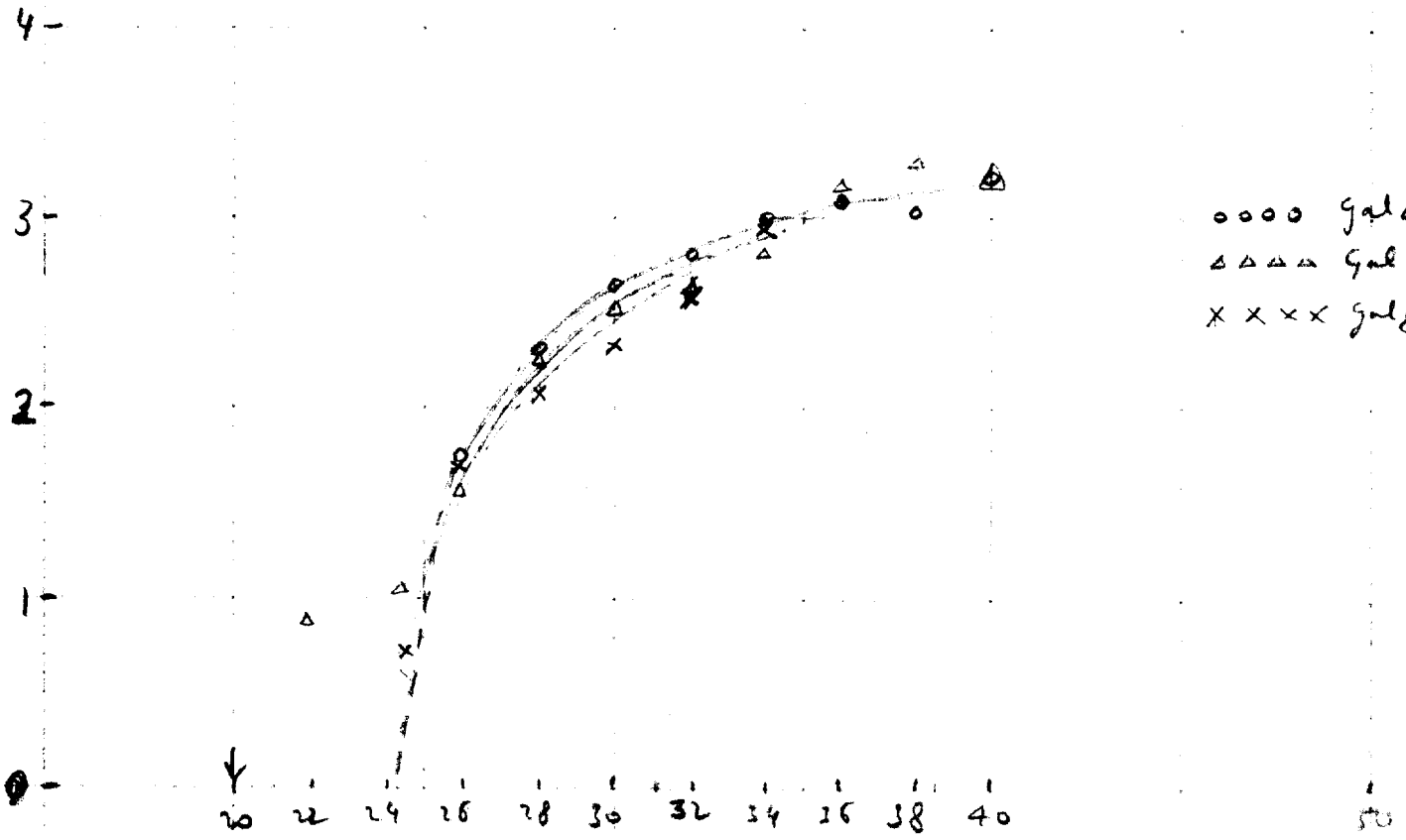
Gal timing, Hfr 2 / Gal 7, 4, 8.

ORC of W 3994, 3997, 3998 φ^R. W3870 from slant
~~Sp~~ 1 ml + 7.5 ml Pen, 1 h 30' rotation. Spun, resusp to same conc.
Mating mixture: 15 ml φ + 5 ml σ in flask.
(prewarmed)
Samples of 1 ml diluted with 1.5 ml of chilled DW, taken
at times: 0, 22, 24, 26, 28, 30, 32, 34, 36, 38, 40, 50, 60'.
Blended and plated, .05 on M Gal B, Sm.

Algo: controls of parents, same conc. as for cross.

Note: 34' for Gal 7, diluted in 3 ml instead of 1.5.

1408/14


4
3
2
1
0
20
22
24
26
28
30
32
34
36
38
40
50
oooo Gal4 (cl. II)
ΔΔΔΔ Gal7 (cl. III)
×××× Gal8 (cl. I)

CISTRONS

I	II	(III)	IV ?
Kin →	Transf. →	Epim.	
Gal$_2$	Gal$_1$		
		Gal$_3$	
	Gal$_4$		
	Gal$_6$		
	Gal$_7$		
Gal$_8$			
		Gal$_9$	

22 22

Lp Gal$_{1,6}$ Gal$_2$ →.

Experiments :

1408/9 : Try Lp Gal$_6$; Gal$_6$ Lp } Gal$_6$ closer to Lp.

1408/10 Try Lp Gal$_2$ Gal$_2$ Lp

Lp Gal$_2$ Gal$_6$?

1408/13 1,2 Gal$_2$ Gal$_1$

1408/14 4,7,8 Gal$_4$ Gal$_9$ Gal$_8$

Lp (Gal$_6$ Gal$_1$)(Gal$_4$ Gal$_7$)// Gal$_8$ Gal$_2$.

1408/15 2,3,4

1408/16 1,2,3,5

1408/17 1,2,8.

1408/18 2,4,6,9

/19 2,5,7,8.