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W3703 → W6-
(WG4 Hfr.)

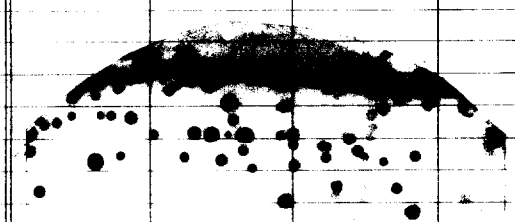
Ref: F of WG4 does not infect to K-12 F.

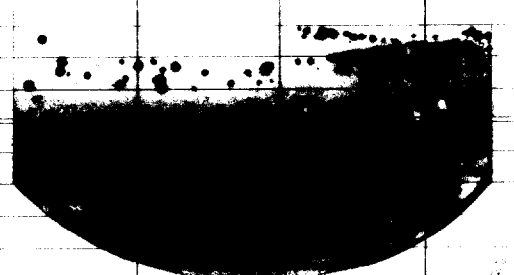
24/V. ; 1959

L Try Lac Mal Sr.

Method : W3703 1ml + W6- 0.1ml + 5ml phasing.
↓
Incubate 24hrs. at 37°C.
↓
Spread on MLac + M
↓
Replica plate it on MGal seeded 2979 on it.

Result : No F^+, and no Hfr was observed.

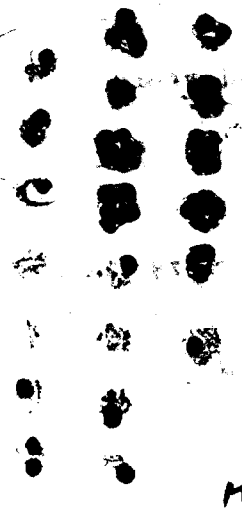




MLac + Meth.

MGal
X2979

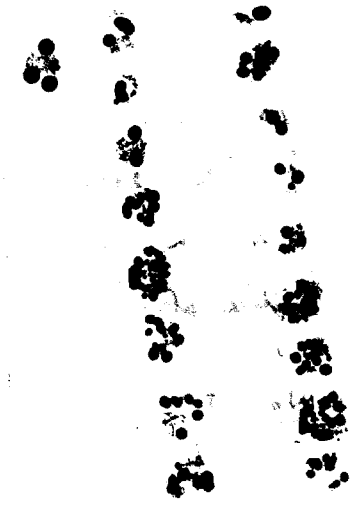
5 -x Wb-



MLacB1

x3828

C -x Wb-



MLacB1

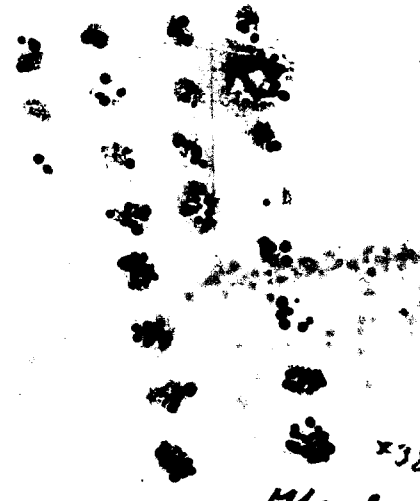
x3828

2 -x Wb-

x3828

MLacB1

4 -x Wb-



x3828

MLacB1

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76 a

~~Infectivity~~ Infectability of F^R to F_8. purpose: Does F^R keep F_8 for a while? Temporally F_8^+.

28/4 ; 1959 REF: P.44 P.45 P25.

Method : cultural age : Overnight culture. (passage 5 ml.) Experimental condition for F_8 infection:

Ratio of mix : 1 : 1. Shaked : 3 hrs. at 37°C on rotator.

Principle

$MGal_4 F_8 \times MF^R S^R$.
W4534 $\times$ $Gal_4^- S^R$.

(Spot them : Untreated.
↓
Add Sm. 400 γ/ml. Sm
↓
Keep them at 37°C for ½ hr. : treated by Sm.
spot them.

	1	2	3	4	5	6	7	8	9	10
								Shaked 3 hrs.		
	3220 F^- $MGal_4$	4534 F_8 $MGal_4$	4526 F^R MS^R	~~3086 F^- MS^R~~	4534 × 4526 F^R	4534 × 3086 F^-	3220 × 4526	3220 × 3086		
recombination 3994 ($F^-Gal_4S^R$)	—	—	—		++	++	—	—		
3086 ($F^-M^-S^R$)	—	—	—		—	—	—	—		
Infectivity 3086 + 3994	—	++	—		++	++	—	—		

Next step ① purify 4534 → 4526 & 4536 → 3086 on Blue Sm. and test sex-compatibility by replica-method. (see next page. P76b)

on MGal and on MGal Sm.

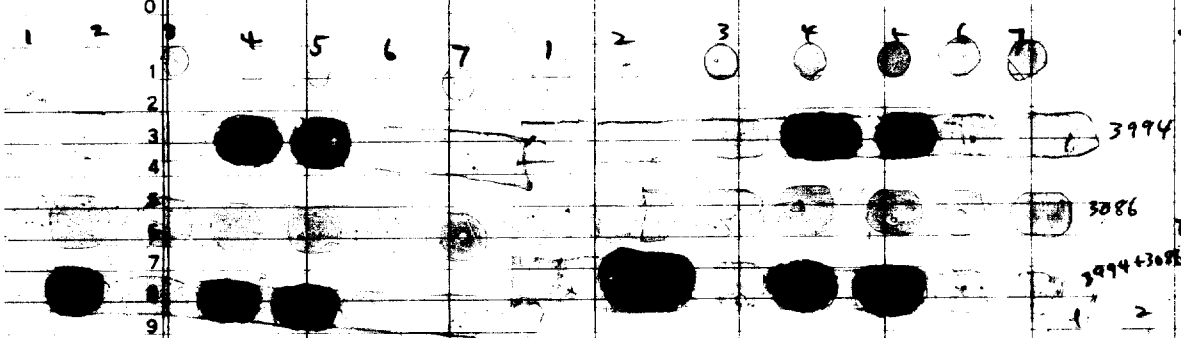
After treated by Sm.

on MGal
on MGal Sm

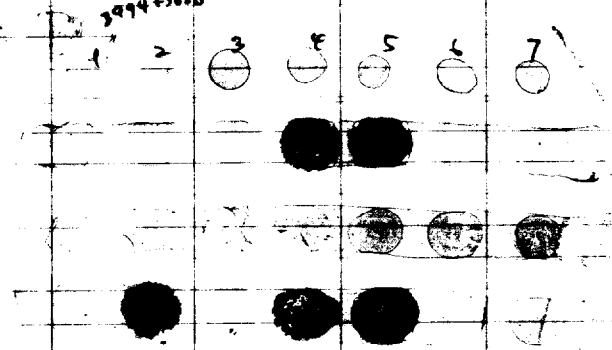
Untreated.

untreated on MGal Sm.

Sm.
37°C 30 min. incubate. after addition of Sm.



treated on MGal Sm.



on MGal.

2 possibilities: 1. Sm-treated W4534 still can transfer F under the presence of Sm.

2. F^R can carries F_8 for a while. and possesses F_8^+ character for a while. But it cannot continue those traits.

Continued from former page

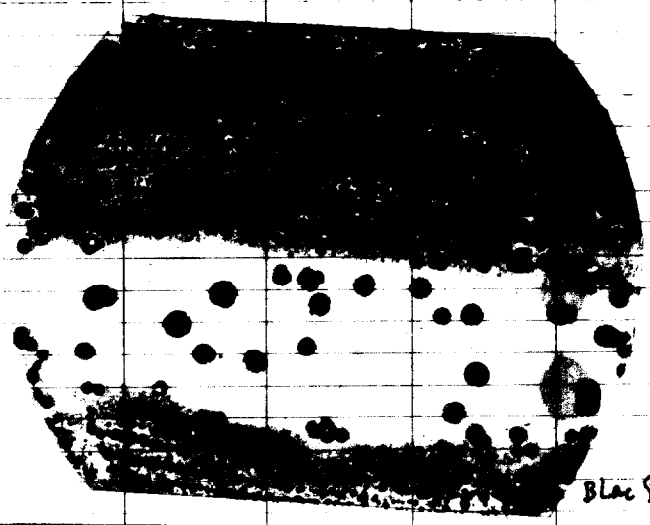
3D/4 ; 1959.

REF: cf. p76a: (Next step)

Method: ① #4 and #5 (4534 → x3086) were streaked on Blac Gm.
(4534 → x4526)
② Replica plate on Mlac B_1 seeded W3828, and see futile colony is there or not.

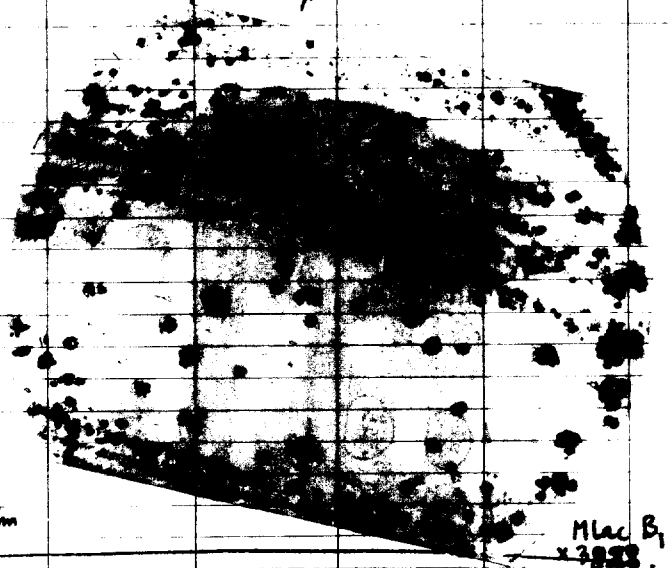
Control. 4534 → x 3086

Master plate



Blac Gm

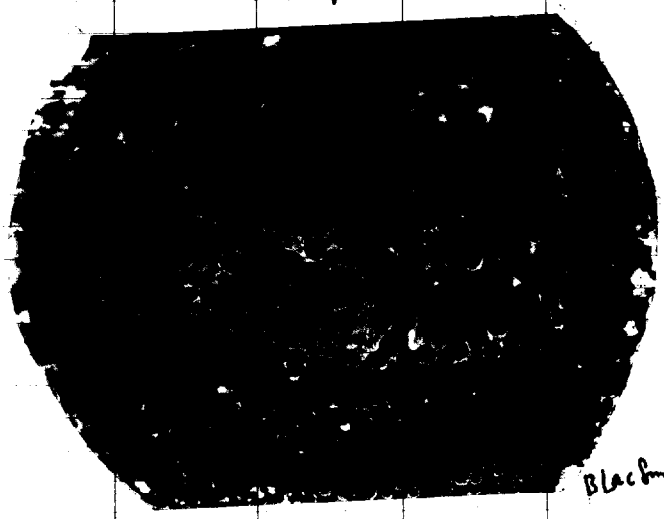
Replica plate.



Mlac B_1
x3828.

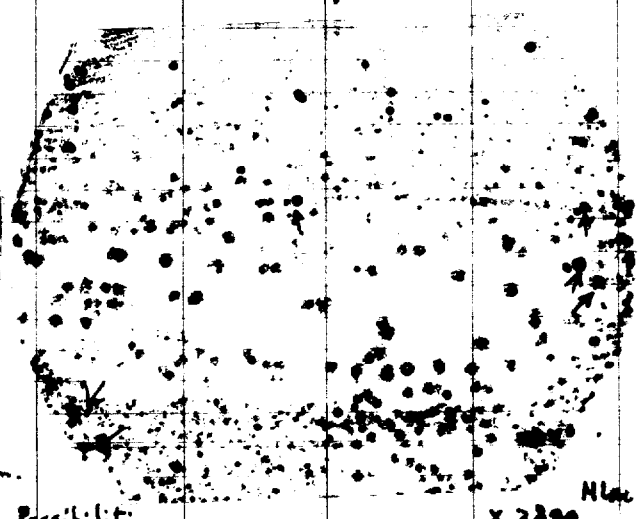
4534 → x 4526

Master plate



Blac Gm.

Replica plate.



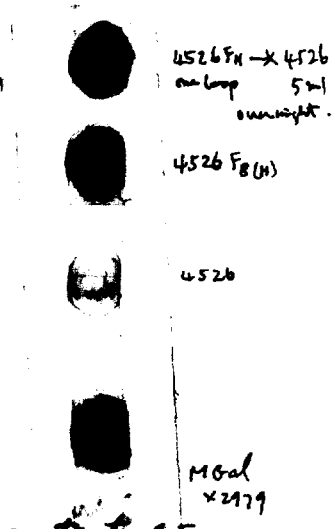
Mlac B_1
x 3828

Next step:
① Repeat this experiment with control.
② If it is host range mutant of F_8, it must be infective to F^- and F^R.
And also H1 for Gal unlike Hfr_1.
(See back page). This is H1 for Gal.

Possibilities:
① Is it mutant of F_8 (Host range mutant)
② Spontaneous (Reversion) mutant of F^R to Hfr_1.
(This seems unlike, because there is no recombination in No. 3; see p76a)
③ Recombination between F^R and F_8. and get Hfr_1 after recombination.

Can infect to F^R.

F_8 + +
+ + F_R



Rough estimation of Host range hypothesis.

① Mix W4526 F_{8H}. → × W4526
one loopful 5 ml pheassy.
pheassy broth culture overnight grown culture.

② Make spot test. on MGal. ×2979.

If. black spot was obtained, it may be Host-range mutant of F_8.

Result and conclusion: 4526 F_H → × 4526 does not become black spot. Infection of $F_{(H)}$ is still very low to 4526 or not infective to F_R.

#2 —x W6—
#4 —x W6—
#5 —x W6—
C —x W6—

AO #2
AO #4
AO #5
AO #5.
AO -C

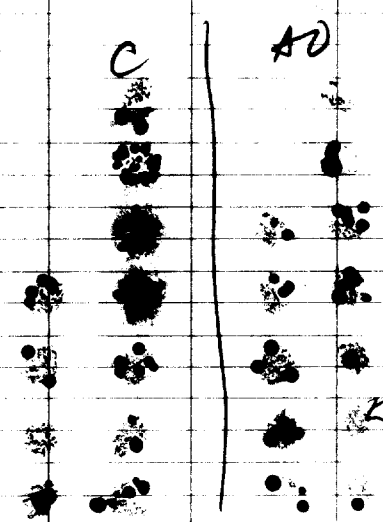
27/4 ;1959 REF: cf. 74.

Experimental conditions: 0.1 = 0.1 : 5ml
grown overnight at 37°C. Pen

0.1ml, AO 30γ/ml, 9 pen.

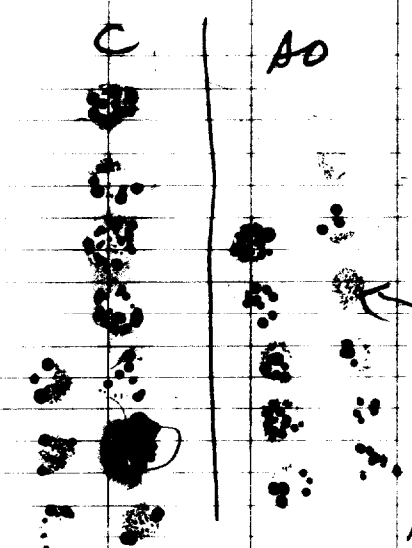
Result: F of
1. #2, #4, #5, C, are infective to F⁻, and gives low fertile, probably F⁺. (See back page).
2. F of #2, #4, #5, C, are sensitive to acridine treatment.

#5.



MlacB, X3826

F1 —x 3086



MlacB, X3828

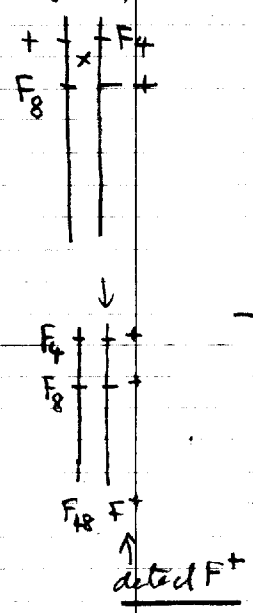
unsuccessful result.

Detection of recombinants from double F^+ strain.

$F_4 \times F_8$

1/VI ; 1959 #2 was used. Cf REF P42, P50.

Principle.



point : Look for F^+ carefully. assumption : F_8 and F_4 is not allelic.

Method 1. Seed #2 on Blac Sm. ($\times 10^{-2} \times 10^{-2}$. 0.1 ml) + ($10^{-2} \times 10^{-2} \times 10^{-1}$. 0.1 ml) ×2.
2. Replica plate on MGal Seaded 2979. on it.
3. Look for Co colonies. 2985

# of colonies tested	# of colonies steril (on MGal ×2979	Hfr colonies
451	1	450
437	0	437
415	0	415
395	0	395
510	0	510
372	0	372
376	0	376
333	0	333
422	0	422
533	1	532
Σ 4044	2	4042

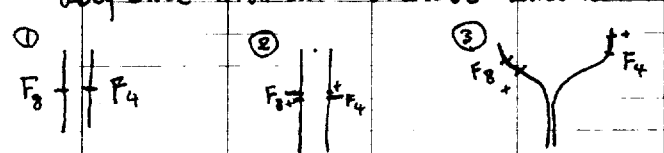
check Gal marker F^+ or F^-

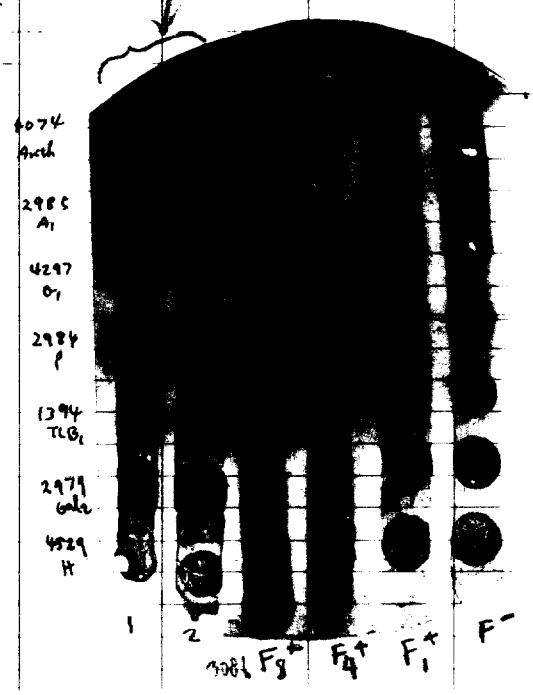
These two colonies are F^- (See bellow).

Possibility : 1. spontaneous reversion of F_8^+ or F_4^+ into F^+ (or F^-)

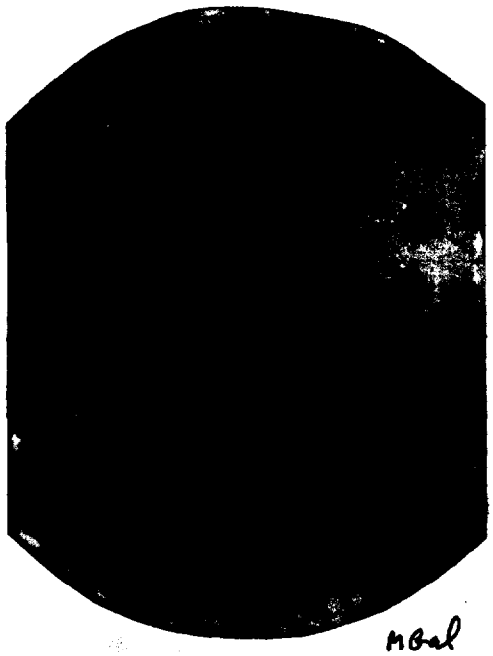
Result: F^+ was not observed (negative result was obtained)

Conclusion : ① F_4 and F_8 locus are allelic, and cannot recombine, or ② they are very close with each others or ③ does not make synapsis.





An example: Replica plating method gives clear spotes in this case.



MGal
x2979

m Blac Sm

Isolate Hfr_8 from W3208. unsuccessful result.

1/VI ~ ; 1959 REF:

Purpose : It is necessary to get Hfr_8.

Method : 1. Take W3208 from stab culture, and purify it on Blac.

2. Cross-brush the 10 colonies. (x 2979 on MGal)

3. pick Hfr and | treat by AO. and see it is resistant to AO or not. infect F to 3086 and see is it infective or not. Use F_8 as control.

Method for infection:

① WbF_8, or W3208 Hfr_8 : 3086 : Penassy broth
1 ml 0.1 ml 1 ml.

② incubate it over night at 32°C.

③ Purify on Blac Pm.

④ cross on MGal x 2979.

1/10

x 2979 on MGal.

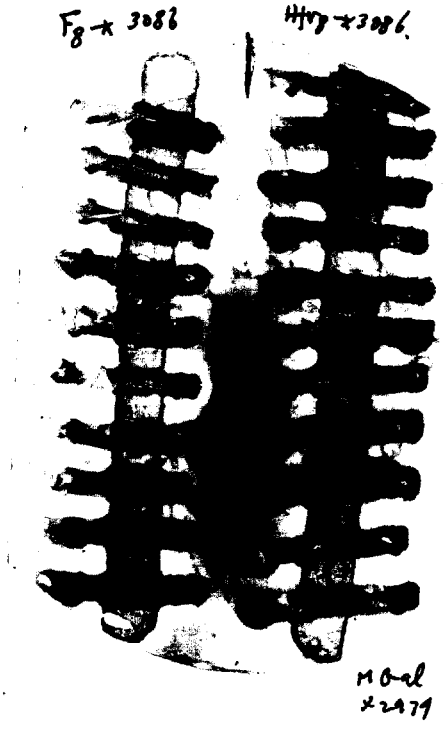
Result : This strain is infective to F^-. ∴ F'_8 (not Hfr_8)

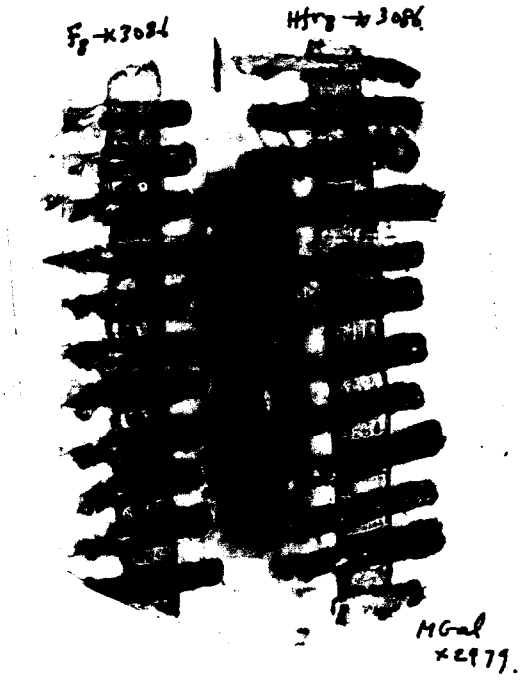
Ratio of infection : $Hfr_8 \times F^-_{3086}$ $15/23 \times 100 = 65.2$ (%) ← (see back page)

control. $F_8 \times 3086$ $17/21 \times 100 = 81.0$ (%)

Next step :
Treat W3208 with AO, and look for Hfr. or use lyophilized culture for reisolation (no mutation) of Hfr_8.

Comparison of infectivity of F_8 to F^-





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GENETICS

W4568

81

Characterization of Host range mutant of P_8.

4526 $F_{8.H}$.

4/11 ; 1959 REF: cf. P76a,b.

Purpose : Confirm this is "Host range mutant" of F_8, and this trait is heritable character determined by itself

1. Infect F_{8H}. to W3735 ($\varphi^S F^R$ M), and isolate W3735 F_{8H}.

· Mix 1 : 1 in pancreas (1ml : 1ml : 1ml)
4526 F_{8H} 3735 Ren.

· Purify it on BGal and replica plate it onto BLac Sm. and see which coloring is W3735. or W4526.

· Pick W3735 (S^S) and cross brush with W2979. on MGal

Purpose : put more markers into F^R mutant to differentiate between F^R mutants.

2. Isolate Gal_6^- from ~~W3735~~ W4526

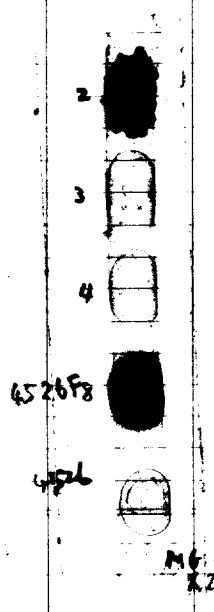
① HFT-6 ⟶ W4526
0.2ml 1ml overnight culture. Keep this one hour.

② spread it on B-O. and incubate it overnight.

③

3. Infectivity to F^-. 3735 F_{8H}^+ ⟶ 3086. ; Method : standard method was

Result: Infective (very efficient) to F^-. 100 %. (21/21)



3828 F_3 → Y10.

4/11 ;1959 REF:

1st trial.

Method : 1. Mix them 1 : 1.

2. Incubate it overnight.

3. Streak it on Blac, and pick Lac^+. Test Xyl. transfer. ×2979. on MXyl + B_1

Result : Strong competition was observed. 3828 has more selective advantage over Y10. Unsuccessful all F^-. try again;

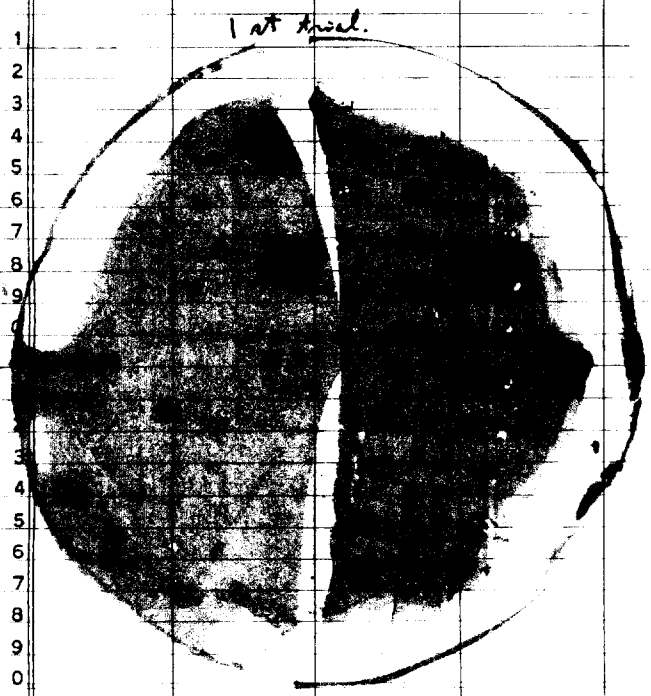
2nd trial:

Method : 1. Mix them 3828 Y10 2 : 1 (1ml 0.5ml), incubate it overnight. Ca. 20hrs.

2. purify it on Blac. ~~(and MLac + TLB)~~

3. Cross brush Lac^+ (Y10) against 3086 on MLac B_1. ~~[illegible]~~ test at least 100 colonies!

1st trial.

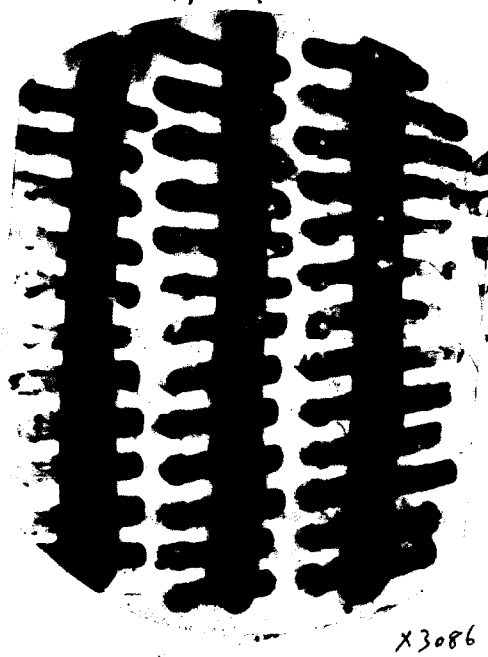


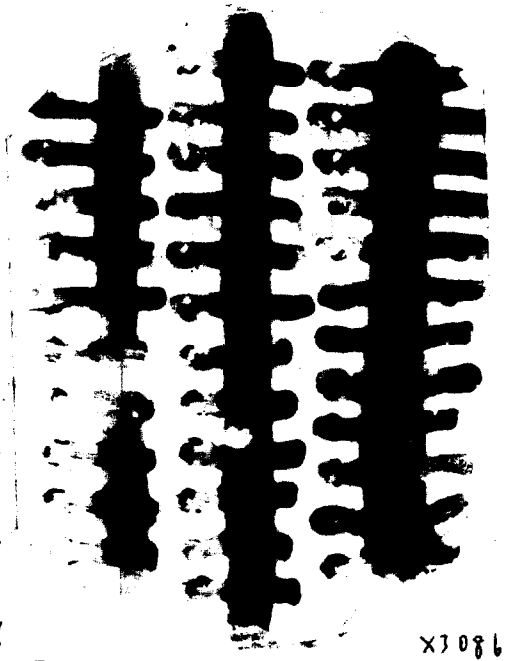
Ratio : (See back page).

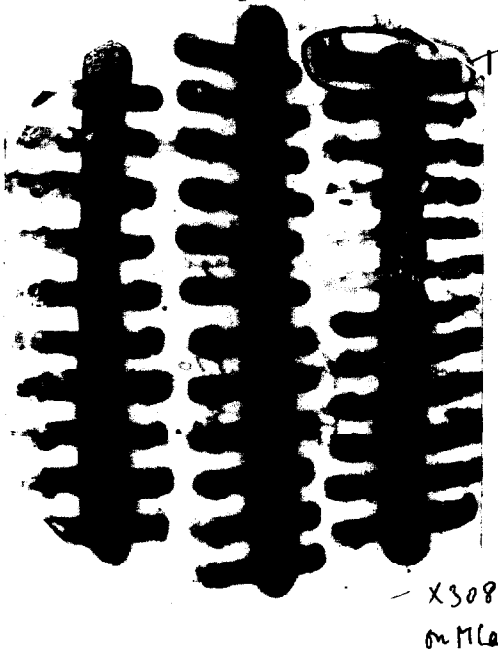
Hfr/S. 0/139.

No F_3 were transfered to Y10.

3828 F_3 only gave F_1 out F_3.



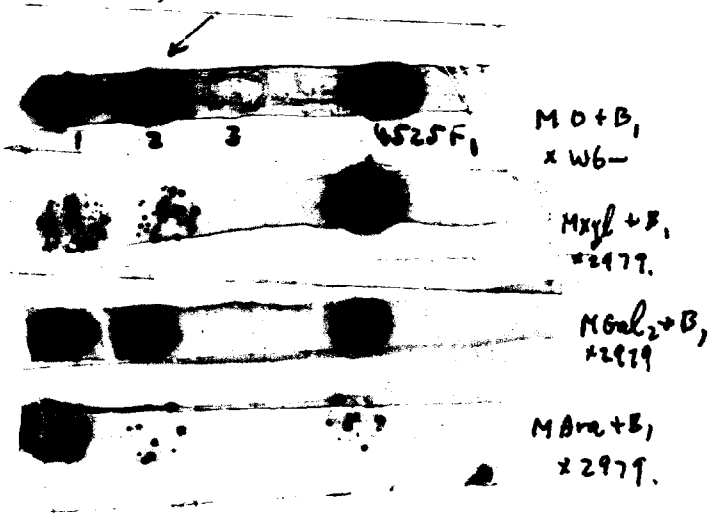




Synthrophy.

Retest. 1, 2, 3. by spot test.

Result : they are not F_3^+. ~~[illegible]~~ only F^+. #3 is F^-



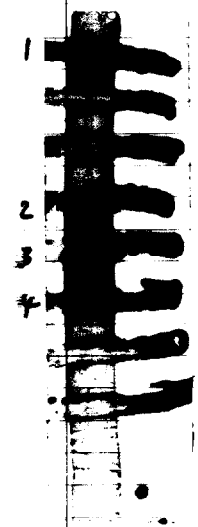
3828 F_3 —x W6-

6/11 ; 1959. REF:

Method 1. Mix them 1:1
2. Incubate it overnight, at 37°C
3. purify it on Blac and Test Lac^+ on sex-compatibility.
x2979 on Mxyl.

Result.
W6 ♂ was obtained after mixed culture with 3828 F_3 and W6-.
Is it $O_3^{\uparrow}$ or F^+?

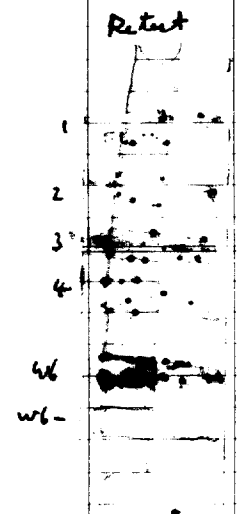
3828 F_3 —x W6-



x2979 on Mxyl.

4/8
=50%

Retest



x2979 on Mxyl

W4576 (F_4 A_1^- Gal_6^-)

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P5

Infect F_4' and F_4'' to W4528.
(selected 10 times for infectivity) $F^-A_1^-Gal_6^-$

8/VI ; 1959 REF:

Purpose: Comparison of the infectivity of F_4 mutants.

1st comparison: Result.

	F_4^+/total (%)	
W6 F_4 → W4528	0/34 (0)	Select on BGal. Compare rate of infection.
W6 F_4'' → W4528	4/35 (11.4)	Test for H1 for Hgl (on MHgl)

(See back page)

2nd trials of infection of F_4 (non-selected F_4) to W4576 was not successful.
0/32 : 0% rate of infection: all F^-.

2nd comparison

Spot W4528 F_4'' and W4528 F_4 on W4293 $F^-A_1^-$ and W3996 $F^-Gal_6S^R$

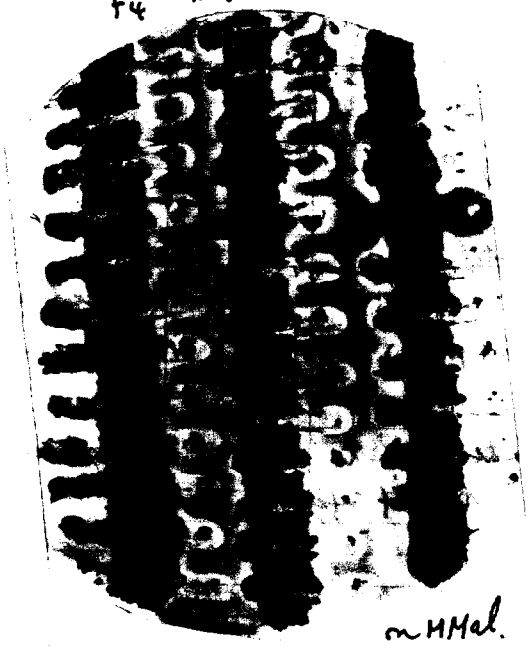
7/IV. Method for selection of more infective F_4.

1st trials. W6 F_4 one loopful (overnight culture) + W6 F^- 5 ml (ca. 10^8 cells/ml).
↓
incubate overnight.
one loopful : + W6 F^- 5 ml (ca. 10^8 cells/ml).
↓
Repeat this process.

This selection was done for 10 times: W6 F_4''.

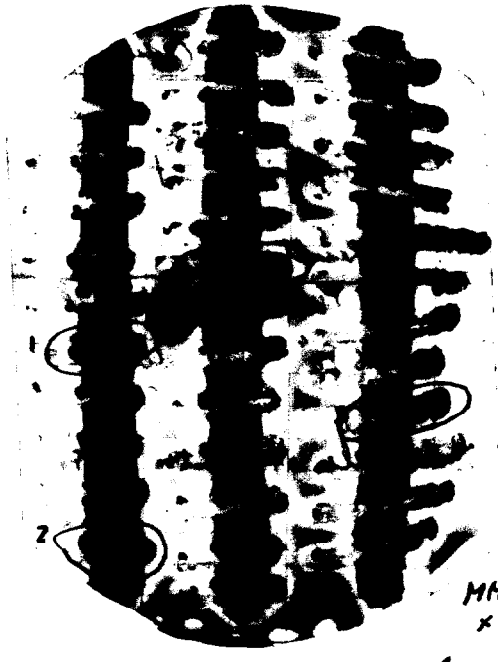
2nd trial: From 8/VI's experiment, the infectivity of F_4'' looks still not enough. Try selection again using W4528.

F4 — × 4528



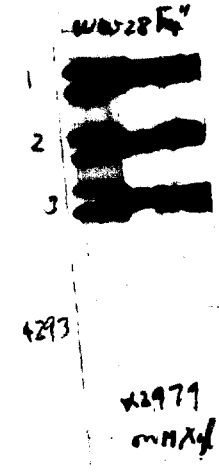
on MMal.
×2979.

F4" — × 4528



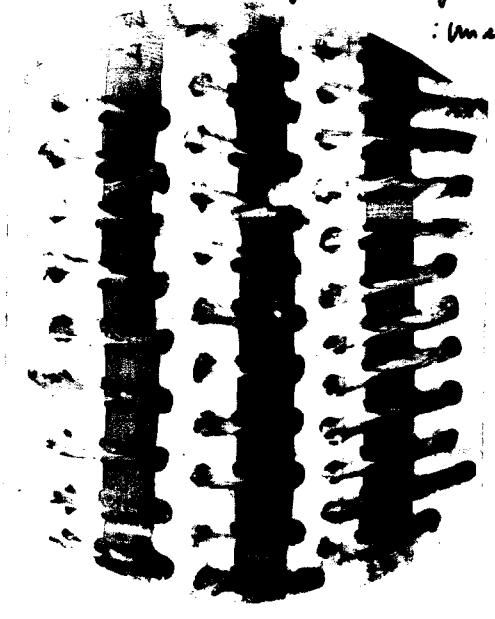
MMal
×2979.

Retest of infected F4"



Mal, marker is reversible.
Even though, F4" can be told
from highly probability

2nd trials of infection of F4 to ω4528
: unsuccessful :
May be some reason



Treat W4552 (3086 F_5)
with AO, and reinfect F_1 to it.
1959, Test the possibility of σ_3 type F'. REF:

Purpose and Principle:

3086 F_5 ~~and 3086 F_6~~ transfer their F to F^-. But the infected cell shows low fertility after infection. Then, what factor was left behind at the time of F transfer. If a factor which determines Hfr character cooperated with F_1 was left behind after treatment of those F'^+ strains, reinfection of F_1 to F^- obtained after treatment of the F'^+ may gives arise Hfr or F' mutant with high frequency.

Hfr

Method of treatment: Usual method. conc of AO. 30 γ. Pen. 5 ml. overnight at 37°C.

Rfd. F_1

Cure from F_5 with AO.
Result: Rate of F^- 1/11 = Ca. 1 % (See back page.)

split off.

Infect F_1 to 3086 F_5^-: W6 → × 3086 F_5^- Use usual method for infection.

1st run was not successful. × 2979 on MXyl.
Result: Rate of infection: (see back page)
0/66 = 0 %

F'^+

Why?
2nd run was successful, and get F^+. Rate of infection:
try again. 23/58 = 39.7 % (See bellow)
Exptl. condition: 5 ml pen. + 1 ml W6 + 0.1 ml 3086 F_5^-.

Conclusion: F_1 → × 3086 F_5^- gives plain F^+, not F_5^+.

This Hypothesis looks incompatible to the result. (3086 F_5^- × W6) gives F^+.

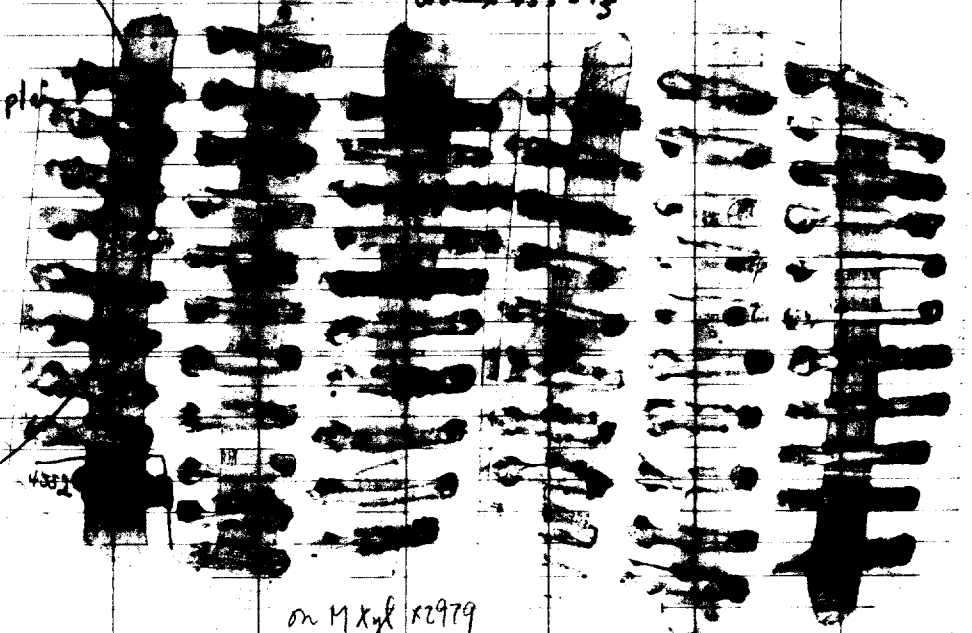
removable by AO.

W6 → × 4552 F_5^-

plain

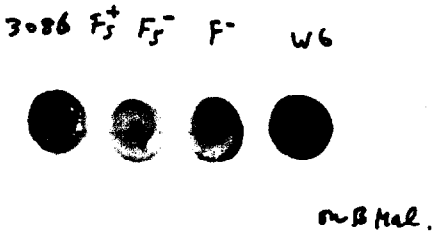
σ_3 type F.
not simple F^-

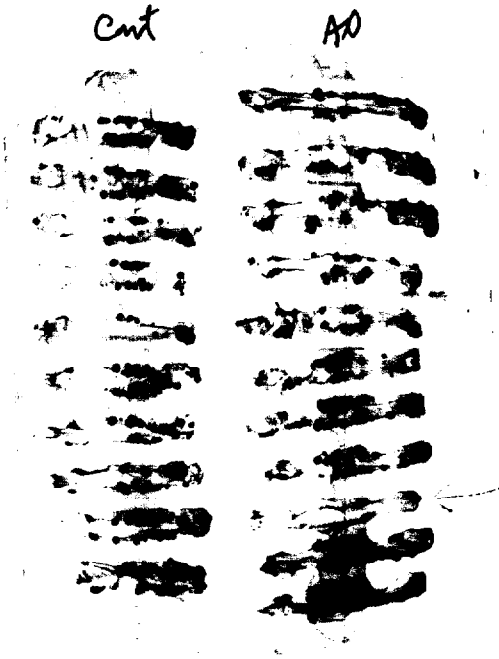
Reinfection of F_1



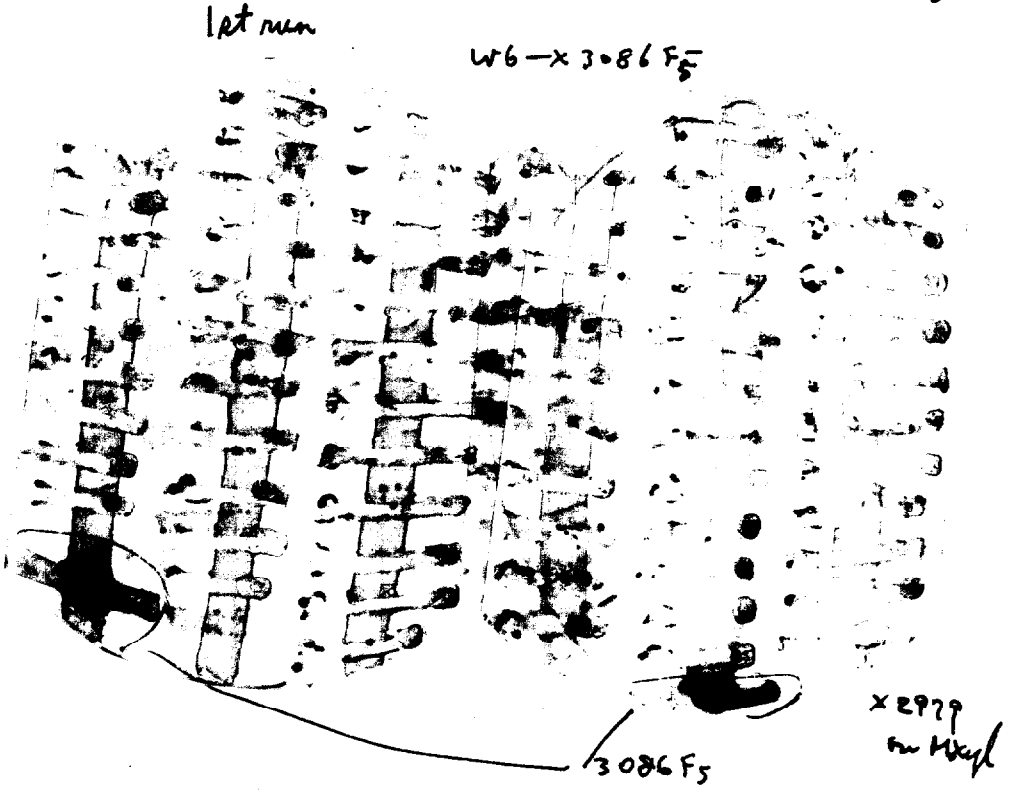
① cure from F_5.

3086 F_5





② Infect F_1 to 3086 F_5^-



Unsuccessful. compair infectivity of F_3' of W3642F_1^+ to F^-

Take control (W6) F^+ M Mal$_5$ Gal$_2$ Lac$_1$

15/VI ; 1959. REF:

Exp. design. : W3642 F_1 ⟶ 3086 Select on BGal Sm.

W6 (control) ⟶ 3086 Replicaplate then on MXyl. ×2979.

Method:

Result:

	Exp. W3642 F_1 ⟶ 3086			Control. W6 ⟶ 3086		
	# of colonies	# of Hfr$_3$ colonies (%) (select on MXyl.) 2979)		# of colonies	# of Hfr$_3$ colonies (%) (select on MXyl ×2979)	
1	618	0	(0)	249	0	(0)
2	661	0	(0)	208	0	(0)
3	700	0	(0)	245	0	(0)
	Σ 3642 ⟶ 3086	0	(0)	Σ W6 ⟶ 3086		

Further experiment:

Use 3833 for F receptor.

Agglutination of F′ strains.

18/4 ; 1959 REF: Peter says he also observed this phenomena.

1 2 3 4 5 6 7 8 9 10

○ Starting point: W4293 F_4 showed agglutination. but F^- was not
(A; Gal_6^-) (X^+ Sugar$^-$ S^R)

young culture : 2hrs. shows no distinct difference between them.

Experiment:

1. Inoculate

Hfr_2 F_2 , Hfr_4 , F_4 , Hfr_5 , F_5 ; F^- . ; F^+

4321 , ~~W3321~~ $W6F_2$, W4321 , $W6F_4$, 4536 , $3086F_5$, W6-3086; W6 , W3086F^+

and incubate them for overnight. All culture was obtained from stock collection in cold room.

In overnight culture, agglutination was not observed.

2. Inoculate 0.2 ml of the overnight culture into 5 ml. phassey broth. and shake it on rotator at 37°c for 2hrs. 10:00 AM — 12:AM.

Result : Agglutination was not observed in Hfr, F′, F^+ and F^-. after 2 hrs, 4hrs. incubation.

Conclusion: Agglutination of F'^+ cells are sometimes observed, but not always does it. Presumably, it is influenced by unknown experimental conditions sensitively.

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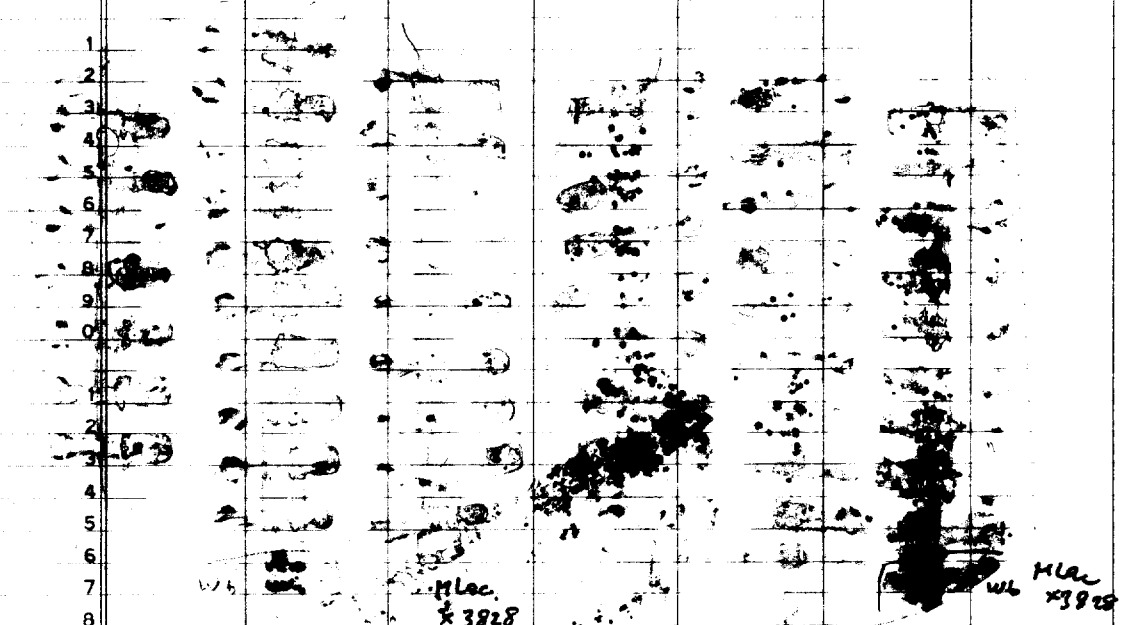
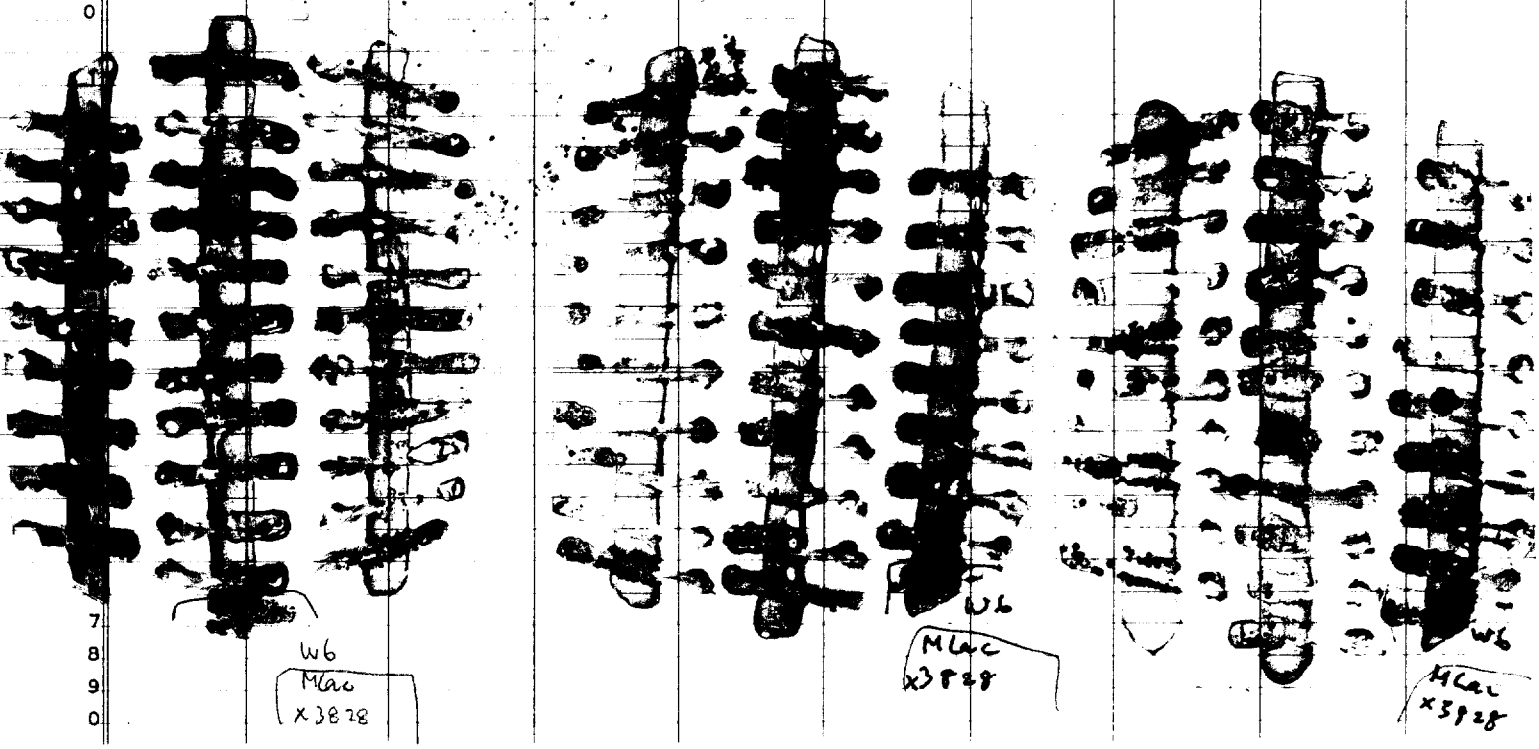
W4526 F_8^-, (Infectability to F_1)

19 REF: cf. P 89

	1	2	3	4	5	6	7	8	9	10
							Ratio.		Mixed culture.	
1		Principle :								
2		Control {	W6 → x W4526 (F^R)				0/26		Overnight:	
3			W6 → x W3086				22/28		W6 1 ml. & 0.1 ml.	
4									Penassy 37°C.	
5									Purify on BLac Sm.	
6			W6 → x W4526 F_8^- (1)				10/31			
7			W6 → x W4526 F_8^- (2)				10/29			
8			W6 → x W4526 F_8^- (3)				11/29			
9										
0		W6 → x F^R control.			W6 → x 3086					





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2nd trial on

W4583

: Take all F_1 apart from (3828 F_3^+)

Isolation of ♀$_3$ from F_3^+.

95

5/4 ; 1959

REF: If it does work, this may be apply to other F^+!

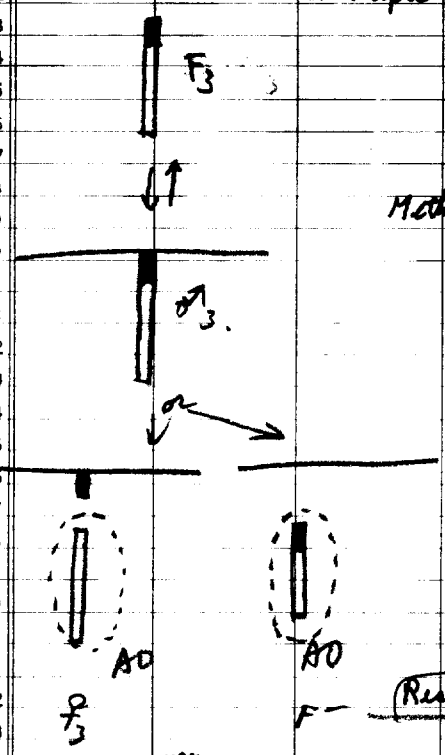
Apossibility.

Method for detection of ♀$_3$ among many F^- colonies.

Principle : F^+ M^- Lac$^-$: W1816.

F_1 ♀$_3$ M^+ Lac$^-$: W3828 F_3^- (arised from W3828 F_3 by AO treatment)

M^+ F^- M^- : W3086



Method :

① treat W3828 F_3 with AO (30γ/ml penassy broth; 5 ml, treated for 48 hrs)

② Seed it on BLac. ~~Smear Blac~~.

③ ~~Replica plate it on MLac Seeded W3086~~

Cross-brush the treated W3828 F_3 against ~~and MLac seeded W3086~~ W3086 on MLac.

Results: ♀ = 51 / total 92 = 55.4% See back page.

④ Inoculate those ~~male~~ female clones into penassy.

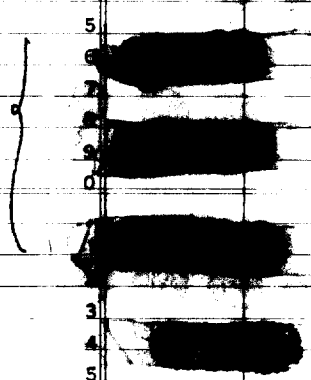
⑤ Make spot test as P.82. using W3086, W1816. on MLac. Use control of ♀$_3$: W3876 ; F^- W3828.

Result: All of the females are all ♀$_3$, not F^-! : See back page.

most of them may become plain F^- but some of them may becomes ♀$_3$ type

not very clear cut, but still higher than F^- (control)

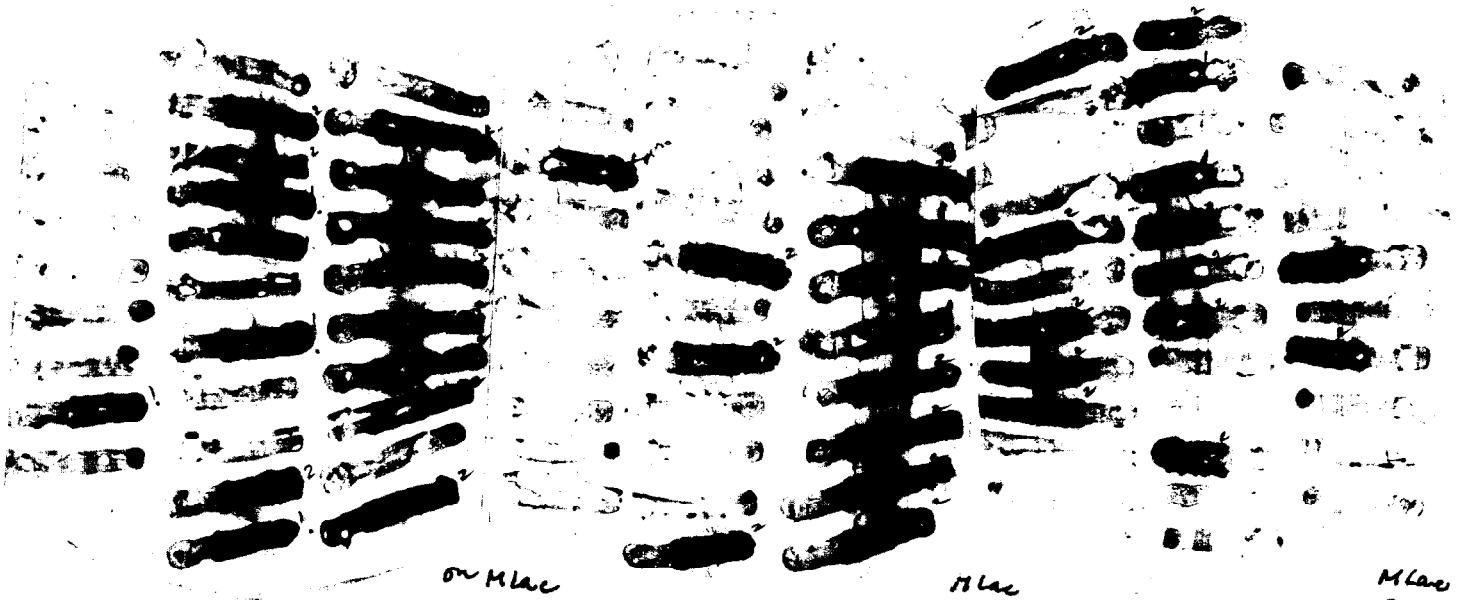
W6 W6−



W3828 F_3^- #1	H1	+ 3086
W3828 F_3^- #10	H1	+ 3086
W3876 : ♀$_3$ control	H1	+ 3086
W3833 : F^- control.		+ 3086

on MLac

AO-treated (W3828 F_3) W458B

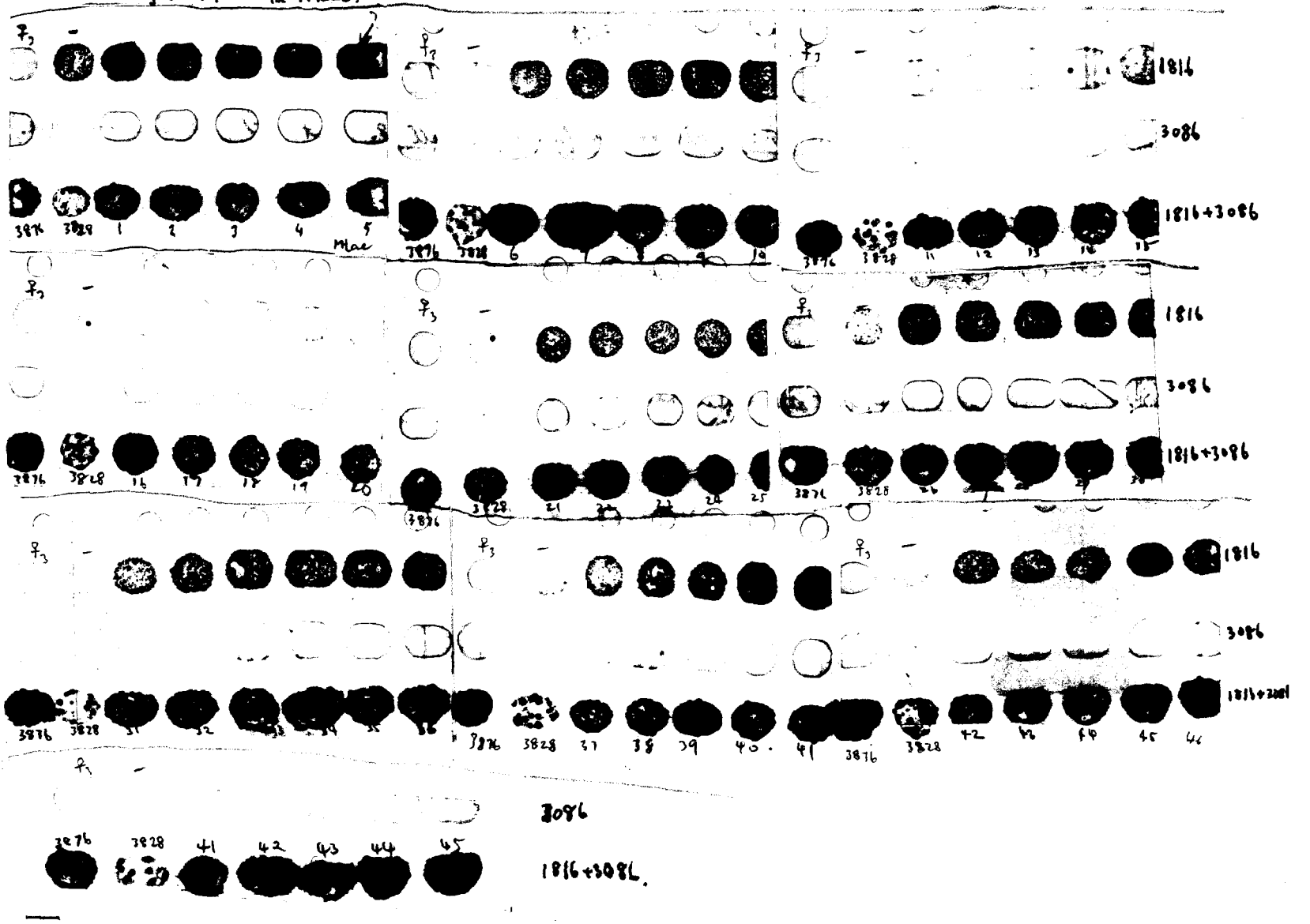


on MLac × 3086 | MLac 3086 | MLac 3086

State I (HI for Lac) : 10
State II (Lo for Lac) : 32
♀ : 51

State I : 9	State II : 32	F^- : 50

Test on ♀₃ or F^- : on MLac.



Cross $F_2 \times F_4$

select Ara_1 (H1 for F_4) and Ara_2 (H1 for F_2) on M Ara.

20/VI – 23/VI 1959

REF:

1. Purify W6 F_2, W6 F_4, W3086. on B Lac.

2. Make overnight culture of them.

3. Add 0.2 ml of the culture into pen (5 ml) and incubate 4 hrs.

4. Mix W6 F_2, W6 F_4, W3086. and shake it for 2 hrs. at 37°C.
   1 ml 1 ml 0.1 ml.

   ① W6 F_2 × W6 F_4 → × 3086 4:30 ~ 6:30 PM.

   ② W6 F_2 → × 3086

   ③ W6 F_4 → × 3086

5. Seed it on B Lac Sm. and incubate them.
   dilution: $10^{-2} \times 10^{-2}$ 0.1 ml/plate.

6. Replica on the M Ara seeded W2979 ($Ara_2 F^-$) and W4550 ($Ara_1 F^-$) [10^8 cells 0.3 ml/plate]

Result: Fertility of W6 F_4 on transfer of Ara_1 is very low as Peter said; therefore it is very hard to tell is it F^+ or not, however, rate of infection under this condition was confirmed.

Control F_2 → × W3086

total no. of colonies.	F_2^+	(%)
341	230	(67.5)
325	198	(61.0)
394	258	(65.5)
346	208	(60.2)
322	194	(60.3)

Σ

Control F_4 → × W3086

269	152.	(56.5)
155	89	(57.5)
260	132	(50.4)
323	182	(56.4)
304	189	(62.2)

Σ

F_4, F_2 → × W3086

	H1		Lo	
444	77 (F_2)	(17.7)	54 F_4	(12.2)
304	72 (F_2)	(23.7)	37 F_4	(12.3)
344	66	(19.4)	37	(11.1)
239	45	(18.8)	36	(15.0)
305	48	(15.7)	37	(12.1)

Σ

Effect of U.V. to infectivity of F'

23/VI — 25/VI ; 1959 REF:

Experimental conditions: 1. Purify all mutants on B Gal. before use.
2. Use young culture : 4hr at 37°C on rotator. (overnight culture 0.2ml / 5ml pen. → incubate for 4hrs)

Principle :

W4534 (M⁻ Gal$_4^-$ Lp^s F$_8$)
W3637 (M⁻ S^R Lp^s F⁻)
X
W3104 (Gal$_4$ Lp^s F⁻)

Exp 1.

Time of irradiation:

	F$_8$ infection	0.1ml. Colony count. #/plate
0	++	— ~~[illegible]~~
5		—
10	++	too much
15	+	ca. 10^4/plate

Method:
Survival of F$_8$. : 0.1ml W4534 UV + (1ml : W3637 / 1ml : W3104) → incubate it overnight. → spot on MGal.
survival of all: 0.1ml /plate.

Exp 2. Qualitative test

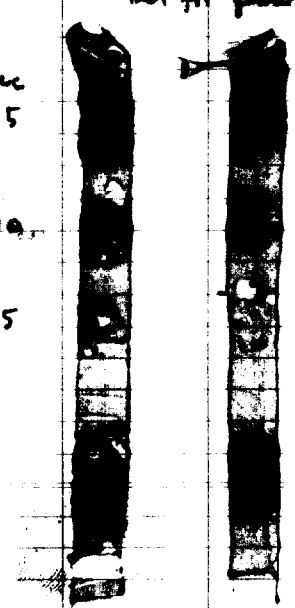
Test for ~~[illegible]~~ Infection.

survival test.

Sec.
5
10
15
0

on BGal.

sec
5
10
15
0



on MGal

0
10
15

on MGal.

Method: ① 1ml : W 4534. broth culture : ca. 5cm diameter : 4534 UV. → X ⊕ 3637 + 3104
② U.V.-irradiated additionally.
③ Spotted on BGal., MGal.

Infection of F' by Killed cell.
Sm. EtOH, $CHCl_3$, CCl_4.

28/21 ; , 1959 REF:

Principle :

F_8 :	4534	F_8	M Gal_4	Cp^s
	3637	F^-	M	Cp^s
	3104	F^-	Gal_4	Cp^s
F_4''	~~4528~~ F_4''	F_4''	A_1 Gal_6	Cp^s
	3106	F^-	Gal_6	Cp^s
	2985	F^-	A_1^-	Cp^+

Method : ① Treat w 4534, and w 4528 F_4'' with various agents.

1 ml of F'-donor pneumon overnight grown culture

Agent used:

- one drop $CHCl_3$
- CCl_4
- 0.1 ml Sm (x100) (usually use 1 mg/ml : Sm 0.1 ml / 100 ml)
- 1 ml of Et OH (95%)

} keep it for 15 min. at room temp. (1 hr.)

② Expel. the treated agent. by centrifugation. or bubbling air.

$CHCl_3$ & CCl_4 is adsorbed into plastic Petri's disk, therefore it is easily removable by this procedure.

Et: OH is removed by centrifugation (once).

Sm is removed by centrifugation (twice).

Centrifuge → discard the supernate.
Add 5 ml pen.
↓
centrifuge → discard the supernate
Add 1 ml pen. and suspend it.

Result : Streptomycin looks good. Almost no decrease.

Next step: Check various concentration of Sm, and various time of treatment.

W4534 (M Gal$_4$ Cp^s F$_8$)

W4534

untreated control

EtoH

CCl_4

$CHCl_3$

Sm

x100

on BLac

3637 + 3104

3104

3637

blank.

untreated control

EtoH

Sm.

$CHCl_3$

CCl_4

W4534 MGal

W4528

W 4528 (A$_1$ Gal$_6$ F$_4^{II}$)

Untreated control

EtoH

CCl_4

$CHCl_3$

Sm.

x100

BLac

3106 + 2985

3106

W2985 blank

C

EtoH

Sm.

$CHCl_3$

CCl_4

W 4528.

on MGal

Confirmation: Infection of F_8 to F^- by Streptomycin-killed cell.
(24 hrs treatment) at 37°C.

28/VI ;1959 REF: 1mg/ml Sm.

Principle:

W4534 M Gal4 F_8
W3086 M F^- S^R Mal1
W3994 Gal4 F^- S^R

Method:
① Add 0.2ml of Sm to Overnight culture of W4534 (ca 10^8 cells/ml), 2ml, Pleasey. 10 times concn
Control: non-addition of Sm. 2ml.
② Incubate it overnight. at 37°C.
③ Wash twice with broth. (2ml) 5min: centrifugation.
④ Spot it on MGal streaked W3086 + W3994. on it.

5ml overnight culture
2ml 2ml
Sm 0.2ml
Incubate them for overnight at 37°C.
Test their infectivity by spot-test.

Survival test by spotting on Blac.

Infectivity of F_8

W4534

treated by Sm (overnight) ×10: 24hr
untreated
W6- {
on Blac.

Control.
3220
4534 untreated control
4534 Treated with Sm. ×10 for usual use. 24hrs.

3086 M
3994 Gal4
3086 + 3994
on MGal

Conclusion: Sm-killed cells still have some F^- infecting ability to F^-, but it decreases into about < 1 ~ 0.1%.

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Transferring ability of F'+ to infect F' to F- after killing by Sm.
(Short time treatment:)

28/VI ~ 30/VI ; 1959

REF:

Method

Conc. of Sm:
200U ×200 : 0.2 ml Sm soln.: ~~1000 U/ml~~ 2 mg / 1 ml
100U. ×100 : 0.1 ml Sm. soln.: " 2 mg / 1 ml.

Time : 1/2 hr. & 2 hr.
1/2 ; 3:15 ~ 3:30
2 : 3:15 ~ 5:15

System :

W3994 F⁻ Gal₄ S^R
+
W3086 F⁻ M S^R) × F_8 — W4534 F

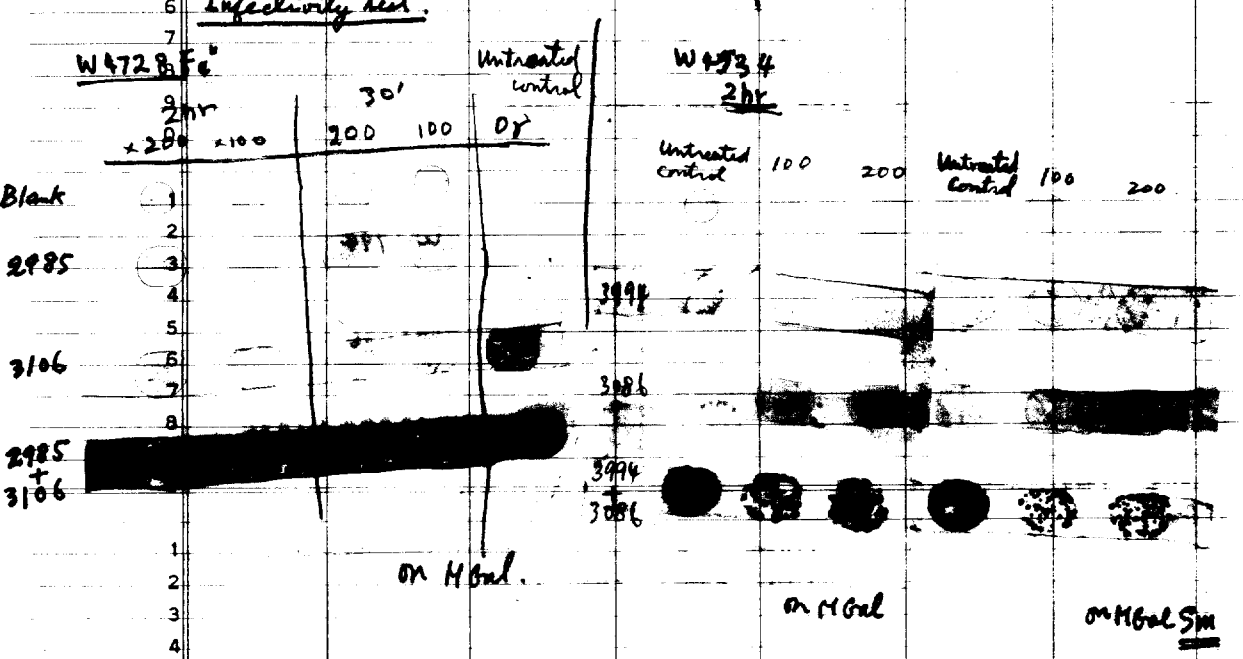
W2985 F⁻ A₁⁻
+
W3106 F⁻ Gal₆) × F — W4576

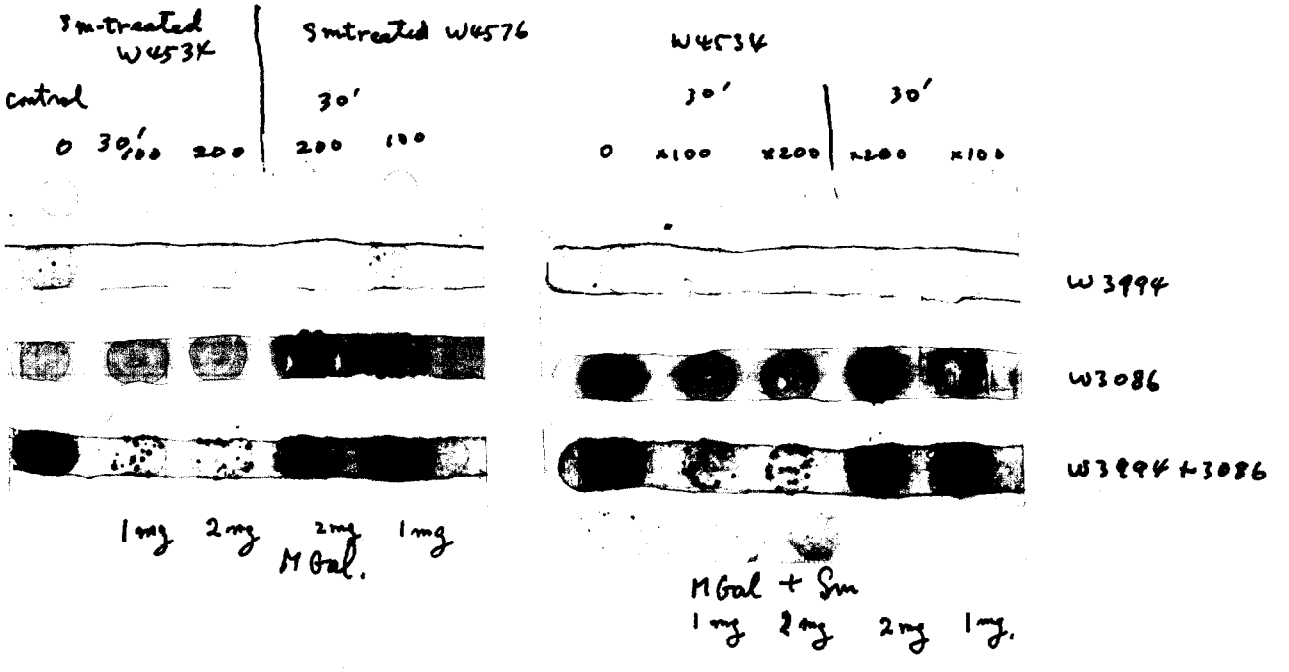
Method :
1. Add Sm 200U & 100U. /ml. separatly.
2. Incubate them for 1/2 hr. & 2 hr.

Survival test : 0.1 ml / plate : after washing : Same sample used as F donor.

	100U-30 min,	200U—30 min.	100U ···· 2hr.	200U ···· 2hr.
W4728 (F₄")	3	0	0	0
	0	0	0	0
W4534	0	0	1	0
	0	0	0	0

Infectivity test.




Sm-treated W453K
Sm treated W4576
W453K
control
30'
30'
30'
0 30' 100 200
200 100
0 ×100 ×200
×200 ×100
W3994
W3086
W3994+3086
1mg 2mg 2mg 1mg
MGal.
MGal + Sm
1mg 2mg 2mg 1mg.