Hfr, F+. miscellany.

June 20, 1952.

Resume Hfr studies. Slant of W1895 showed a yellow contaminant. Restrick on EMB Lac. Colony #1 was extreme rough. (continued Hfr) Keep as W2041. Retain a "smooth" culture as W1895. Both cultures test equally Hfr.

Attempt to resuscitate diploid cultures.

777-series mostly inviable.

81051. OK. Mtl v. Recorded as Lac- Mal- Preserve as H-311
✓ Mtl v Lac-.

H245 - n.g. first attempt gave all Lac-

6/25. A W1678 × 1607 2 trials No Lac+ SR colonies.
B 1956

C W1918 × " 2 trials. No Lac+ SR
D. 1956

Rare papillae in thick streaks: A, B, C, D
see 954

6/26. W1590 × W1940 EMB Lac. No + colonies noted. 1?

E Hold for papillae. 1? See 952.

F H310 × W1895

G. H311 × W1922.
(Het Lac = Mal $\bar{x}$) See 955

H267/SM. 7 Lac v : all prototrophic. 1236 are Mal- 4,5 Mal + ✓ 7 Mal v

See 953. Save #2: H312

Hfr Lac+ X Hft Lac- 952

June 27, 1952.

W1940 x W1590. Mix in Penassay ca 4 hours, streak out on EMB Lac.
24 hours: opp all Lac-, some colonies have a dense center.
A. ① streaked out, gives a few Lac+, apparently not var, but with var. appearance on EMB Mal- ② Lac+ is pure Mal-.

6/29 48 hours: ca 1% Lac S colonies, and typically lac+ at colony intersections.
B. Picked and streak EMB Lac. (8)

C x W1956. Similar to B, somewhat fewer Lac+. (4)

6/30: Lac+ appear pure, Held for further development. ~~3~~ 3 colonies in B restreak EMB Lac.

no change
some begin to papillate

1 Lac v } Mal-
2 Lac v }
3 Lac + }

Restreak 1, 2 Test nutrition. both TL
1 (BM): - (TLB) + Try B, only.
2 (BM): + (TLB) +++

Use #1 for further tests. Both are mostly rather weak Lac v+.

↓ H 316

Hfr x diploid (H312)

June 27, 1952.

H312 isolated by sm selection from H267 as Mal- lac$\underline{v}$ prototroph.

W1895 x H312 on EMS $\underline{Mal}$ for Mal+ prototroph.

7/30 4 hours. streak on EMS Mal. (v H312 parent. ca 10%v)

Mal+ only as papillae.

---

overnight: streak on EMS Mal.
~~Mal+ only as~~ after 3 days, numerous papillae noted.

7/6 8 streaked out directly. 1 clearly Mal$\underline{v}$. (H318)

Restreaks on EMS Mal → Mal+, Mal- colonies.

Restreaks 4 of these to EMS; EMB Mal. also lac$\underline{v}$ +/+-

---

7/9. 40 tests 2 Mal$\underline{v}$. Restreak EMB Mal, EMS Mal.

#1 ✓ (H319) #2? ↓ Mal+ Lac-. Restreak single EMS Mal col.

~~#1~~

H319 on EMB Mal gives $\underline{\text{almost exclusively}}$ Mal- and Mal$\underline{v}$ colonies. The latter restreaked do the same. on EMB lac, mostly weak+ lac+ and Lac-, occasional lac+. Triploid? or variety of Mal+, lac+ segregants?

July 16, 1952.

H318 = W1895 x H312 (H267 Mal - s^R).

8 segregants: 4 Mal+ MH+ all lac-

	S+	D(0)	(BM)	(TLB1)	S	D(0)	(BM)	(TLB1)
1	S	−	−	+	R	+	+	+
2	S	−	−	+	R	+	+	+
3	S	−	−	+	R	+	+	+
4	R	÷	−	+	R	+	+	+

∴ Mal+ are TLB1− Mal− are prot. (also seen on EMS Mal see 953)

Note crossover Mal+/mutr.

H310 ~~H310~~ x W1895
~~H311~~

~~955~~
~~956~~
955

June 26.. 1952.

H310 = non-disjoined (W1895 x W1177) TLB, - Mal-S^R Lac$\underline{v}$ Xyl-
8 + Prototrophs picked from EMS Lac. (pred. Lac+ and Mal+).
~~Most appear Lac$\underline{v}$. Spot on EMB Lac, streak EMB Mal, Mtl (or Xyl).~~
4 from EMS Mal.

8 Lac: all pure Lac+ 4 Mal: 3 pure+, 1 Mal$\underline{v}$! H313
check on EMB Mal / sm. check on EMB Lac.
↓ 4 Mal- S^R
2 Mal+ S^R
2 Mal+ S^S

Prot → S^v Mal^v Lac^+ Mtl^v
Dissolate on EMS Mal;
Purify Mal+, Mal- for
nutritional and fr test.

24 Mal+ streaked out EMB Mal: No likely Mal$\underline{v}$.
✓ vs. EMB Mal / sm. ✓
all Mal+: 4 S^R 18 S^S

Pick 8 Mal$\underline{v}$ from H313 to purify Mal+, Mal-.

40 Mal+ → no clearcut Mal$\underline{v}$. Restreak 4 on EMB Mal, Lac; check vs sm.
not $\underline{v}$ on either
↙ others were Mal+ S^S (40)
B) 1 seemed to be Mal+ S^S / Mal- S^R Restreak: appears ~~Mal~~ (2) Lac+! Retest
vs SM: single colonies and mass.

Repick single colonies No Mal$\underline{v}$ left.

H313 segregant: prepare for outcrosses 955

July 5, 1952. 7 pairs:

Mal - Xyl - Mtl - completely linked. All auxotroph, Lac +

	A - - - S	Nutr: D(M)	D(TLB,)	B + + + S	D(M)	D(TLB,)
1	R	-	+	S	-	+
2	S	-	+	S	-	-
3	R	-	+	S	-	-
4	S	-	+	S	-	+
5	R	-	+	S	+	-
				S	-	-
7	R	-	+	S	-	-

These types are therefore seen:

				Repeat	Test EMS Lac, Mal 1607	1177
A	Lac+	MXM-	S^s (TLB,)-	A2, 4	✓ +++ Mal+ >	
	2	MXM-	S^R TLB,-	A1, 3, 5, 7	✓ ++ Mal+ >	x3: ++++ x5P.161: ++
	3	MXM+ S^s	M-	B5		++++ -> ?
	4	MXM+ S^s	TLB,-	B1, B4	+++ Lac+ <	
	5	MXM+ S^s	(MTL)? ✓	B2, 3, 6, 7.		16781

B5 would appear to be ≐ W1895] #3 is certainly Hfr ≐ W1895.
Also test types 1, 2, 4, 5 for Hfr] #1, 2, 4 are either F+ or Hfr.

7/8. Also mix W1607 + 1, 3, 4 4 hours. Streak EMB Lac SM. (for recomb + F+ transduct)

B. 1 Scored poorly ?
3 Scored poorly
4 ca 2% +
on repeat, 1, 3, 4 gave 1-5% Lac+ S^R.

C. Repeat 1956
1345: 4PM-9PM in broth
all gave 1-5% Lac+ S^R
∴ Hfr.

D) Reisolate Lac- from
B1, 3, 4, 2.

Plate x W1956: all F- ✓
∴ 1, 3, 4 are confirmed Hfr, non transducible

#2 is only one in doubt.
It reacts with intensity of F+ (non Hfr) with W1607.

Is #2 Hfr? If so, test H310 ✓
Lysed colony noted in plating of B2 (W1607) on EMB Lac → λ^s (acc EML).

July 13, 1952 ff.

H310 (mass culture) x nD(o)

7/16. 58-161 +++ ∴ H310 is presumably Hfr!
W1607 ++++

58-161 x 1956 ++± and W1895 x H310 is not especially significant.

1607 x 1956 −
1802 x 1956 −

(cf. 955D).

7/16. Test Lac+, Lac− from H310. ~~cf~~ compare with from haploid W1895 x W1956.

1 E.R.L. 7/10/52 Single cell - zygote syn. Lac+ SR #7 TL
2 " " " " Lac− SR #7 BM ✓ W1607
(3 Lac− SR #11 , TL
4 +

5 H310 Lac+ (TL ✓)
6 " Lac− (TL ✓)

~~what does this mean?~~ ✓

All gave 0 prototrophs x W ~~[illegible]~~ 1607!, #2 x 1956
Repeat H310, Lac+ and Lac− x W1607; #2 x W1956.
(F+) F− probably F− but cont.

Repeat H310 and Lac−

H310 x 1607 ++++
H310 Lac− x 1607 − ~~[illegible]~~
" x 58-161 +

∴ at least two segregants of H310 are not Hfr.
H310 itself appears to be.
Try crossing H310 x F−! Streak out H310 x W1607 on EMB, EMS Mal
= ~~955E~~ 964

7/21/52

a. 8 Lac+ S^R from W1895 x W1956. 5 were also associated with Lac- S^R. Check nutrition: 2 Lac+ were B_1 -; remainder were T L B_1 -. Test these 6 Lac+ and 5 Lac- for F status.

1-8 = Lac + 11-18 Lac -. x W1607.

	x 1607	
1	-	++ x 58-161.
3	-	
4	-	
6	-	
7	-	
8	+++ → ~~two~~ (3) 2 types of colonies on Lac	→ A Lac ++ → ~~MH+ and -~~
		→ B, C Lac+ weaker sl. mucoid

all appear to be compatible ♀ F-.

Restreak #8; The others appear to be F -.

b. Isolate Lac -, + segregants from individual H310.

Use early after checking purity.

Lac+		Lac-	
21	-	31	-
22	/		
23	-	33	
24	-	34	
25	-		
26	-		
27	-		
28	-		
29	-	39	
30	-	♀ 40	

all were incompatible x W1607.

---

955 F 8A = MH- but MH+ papillae and one + colony noted!

Restreak. Test MH+, and MH- W2068

MH- papillae after several days. ♂. W1956 itself?

~~H310~~ x ~~W1895~~ 956 ~~956~~ 956
H311 1922

June 26, 1952

H-311 = Het diploid: Lac- Mal- MHlv. prot. EMS Lac. Pick 8 Lac+ → Lacv.
A) Prot. mostly Lac-, occasional Lac+. (May be revisions of H-311?)
8 picked: all are Mal- S^s MHlv (exc. #1, #8 MH-). Restrick these on EMB Lac. (prob. not diploid) (Should select for Mal+ prototrophs)
✓ : these two are Lac+ Mal- MH- not diploid.

? Are the Lacv revisions of H311 or recombinants with W1895?

Restrick from EMS Lac (ctg. Lacv) to EMS Lac., Mal. B)
This strick is ca 30% Lacv but very few if any Mal+ on EMB.

C) Reni EMS Mal.
~~Almost all Mal-. 2?? Mal+ prototrophs. Restr to EMB Mal.~~
4/5 show a +v rx on EMB Mal. Possibly Mal+ Lacv? Restrick on Lac, Mal: all 4 are Lacv Mal+v, as surmised.
These must be results of H311 x W1922, and presumably hemizygous for Mal. Het diploids may not bypass elimination!
R. test additional

Test #2A for Mal hemizygosity: 8 Mal EMS papilla → Mal+ on EMB. Recheck on EMB Lac: all were Lac- → Lac-
7/10: 3 addl.
~~4 Lac+, 4 Lac- segregants isol. All were MH-! See 956A.~~

D. 40 Lac+ tested: 23 were clearly Lacv.
Spot on EMS Lac, EMB Mal: all 23 are Mal-. Do not save

7/15/52 Segregate (H320) A2. 4 Lac+, 4 Lac− (all Mtl− on first test) (Hfr × Het diploid Mtl v)

	Lac	D(o)	D(BM)	D(TLB)
1	+	−	−	+
2	+	−	−	+
3	+	−	−	+
4	+	−	−	+
5	−	−	+	−
6	−	−	+	−
7	−	−	−	+
8	−	−	−	+

where are Mtl+ ?

Pick on EMBMtl.

3 types: #1, #5, #8.
TL− M− TL−

All should be S^s Mal−.

[Test for Hfr by Lac S cross (exc. #5 and 8 are Lac−)]

H320: Select for reversions on EMS Mal. Pick ~~88~~ 48 papillae to EMB Lac.

1 was still Lac v → Lac v Mal+ ∴ H320 is hemizygous Mal−.

λ / W1827

957

~~June~~ July 1, 1952.

lysis = +

Seed plates of:

Spot	W1827B	W1485
λ	+	+
K12	+	+
1485	−	−
1827ABC	−	−
B	−	−
518	−	−

no background plaquing

Cross-strains on EMB Suc.

	B	λ	1485
1827A	−	−	−
" B	−	+	−
" C	−	+	−
1827B+λ	+? plaquey	(B: + !!*)	+ pl.
K12	− ?	−	+
1827B+K12	−	+ , +	+

*prob. used 1827B.

No evidence that 1827 (= 1827B) is lysogenic vs. 1485 or B.
Reduce B/λ ✓ B is λ^B.

7/2. ~~B/seed plate:~~

~~1 λ~~
~~✓ (λ+1827B plaques)~~
~~3 (λ) + (1827)~~
~~4 1827~~

Plate ca 100 particles of λ on
A) 1827 → 100 plaques, clear,
B) 1827+B (B) no 100 turbid plaques
Pick plaques A, B to
1485, 518, B. None grew on B
all others equally + on 1485, 518.
No gross evidence of phage modification.

EML now reports that W1055 (Use 1027?) was responsible for the modification.

H245 x W1922

958

July 2, 1952

7/3. 40 Mal+ colonies streaked EMB Mal. Mostly Mal+ or +v
8 are likely Mal v. Restreak EMB Lac, Mal; EMS Mal.

2 3 6 7 8 are Mal+/- v?

1 2 6 7 8 Lac+/- prob. Lac v. 3 are -+
4 pure -

1, 6, 7 certainly Lac v 6 certainly Mal v.

Retest from EMS, single colonies: 1, 2, 3, (4), (5), 6.
Remember

1-5 are Mal v Lac v # 6 Mal v Lac+

At first glance #2 appears Lac+/- , 1, 3, 4, 5 Lac +-/-+
Reprinify Lac+/-. H321

H295 secondary. ....
H324-325

June 26 ff. 1952.

Plate H295 from D(lac) on EMB Gal. Replica to Lac. Pick apparent Gal-lac$\underline{v}$ and test for λ...

1 Gal-lac$\underline{v}$ secondary isolated. It is apparently still $\lambda p^+/\lambda p^s$. = H317

Later proved to be Gal$\underline{v}$ still, although with slower expression of Gal+.

Difference between H295 - H317 obscure. May be primarily a shift from segregation ratio +>− to −>+. Lysid colonies (Gal-) are very prominent.

H324-325.
June 9, 1953. Plate out from D(lac) to EMB lac, Repl. to Lac, Mal, Gal. 3 plates each. >95% lacv(+). ca 150/plate

possible lacv Mal-

1. H325
2. H325

lacv Gal-

3. H324

See 1056

4. Galv lac- H324
5. Malv Gal-lac- H324

H316 x W1895

July 4, 1952.

EMS Mal.

v. high yields. ca 20% Mal+. Picks smallest colonies, restreak EMB Mal

16 tests: 3 likely Mal v. hold to pool with further tests.

+ 40 tests } . No Mal v noted !

Some possibly Mal+ v but no Mal- segregants seen in estimate.

F- via sonic disruption

7/7/52.

1 plateful W1895 (A); W1678 (B) in ca 15 ml. H_2O.
Subjected to Raytheon machine at full power, 15 min.
Then filter (14 lb Mandler).

4:30 Inoc 1 ml filtrate (± 1 ml W1607) in 5 ml Penassay.

9A8: Controls clear. Plate Exp. with W1177 on D(0) or EMB Lac.

		× W1956	on EMB Gal.
	A = 1607 + 1895 fil.	0	15
	B = 1607 + 1678 fil.	0 0	7
48h.	C = 1607	0	13
	D = 1607 + 10^9 λ (K-12 UV)		315 (confirm Mt Morse).
	E 58-161 × 1956	$+10^2$	

∴ Ultrasonates lack both F+ and Gal+ transduction activity

Streak out papillae from C (8), D (12) to confirm Gal variegation; test possibility of F transfer coincidentally.
"D1" was a very dark papillae. C papillae do not show such clear fermentation rx generally.
E (v) are areas between papillae for control use re F+ transduction.

In addition to streaking-out, cross-brush fushly pulled papillae / sm EMB Gal; all unif. SR
spot on D(0) - all auxotr.
Inoc pooled samples to Penassay to prepare pulm. F+ test.

C 1-8 D 11-20
D 9-10

8C: all appear to give pure +

12 D: at least 9 show Gal v, ~~some~~ possibly all.

Repurify from single (selected) colonies.

Pooled Gal+ (C, D) and Gal- (E) remained F- (× 1956)

7/14/52

Restr single colonies.

C (spont rev.) 8: all pure Gal+

D. (should be 19/20 Galduced). 12 tests:

1	+	−	7	+	−
2	+	−	8	+	only
3	+	−	9	+	only
4	+	−	10	+	only
5	+	−	11	+	−
6	+	−	12	+	−

∴ 4.5 | 3.5
8 | 0 | 8
6.6 3 | 5.4 9 | 12
11 | 9 | 20

is comparison. ~~No serious doubt of effect.~~

For further study, Restreak single colonies of # "1" for acquired stability, and 8, 9, 10 for present status.

7/15. In restreaks, #8 showed 1 Gal− ? (Recheck). 9, 10 were entirely +.
but restreak single colonies → all Gal+

A. (but background has poss − ?)

7/15 #1 showed 3 +, − 1 "pure +". Restreak

7/16. All colonies were +, −. Restreak + and v → 7/17 all +, −, v colonies.

RESUME: (W1607+ λ(K−12))

9 → Gal v ; 3 → Gal +

Purification: Gal v ↓↓↓↓↓↓↓↓ all v ... ; Gal + ↓↓↓ all + ↓↓↓ all +

7/15 all v

7/16 all v

7/17. all v

↓ 12

7/18 all v

↓ 8

7/19 all v

Instability of Gal v seems to be persistent. No truly stable Gal+ noted!, if retested by further streaking.

Save as stants: 1v, 8, 9, 10+.

Restreak to look for + from v

8 spont. reversions from Gal− from #1 were all stable + for 2 transfers

July 18, 1952.

5 plates DNZ Lac harvested to ca 20 ml. Ultrasonic (10,030 cps)
2 minutes. K12, W1895 (H)

A) Survival ratio (measured 7/21/52). Plate 1 ml 10^{-8}. { Initial counts
B) untreated treated – estimate density at ca 5% (subj. est.) { ca. 10^{11}/ml
treated cells ca 10^{8}/ml

Filter through 14# Mandlers: Highly opalescent filtrates.

#1: K12 2: 1895 . test sterility 1 ml samples.
add 1956.

Sterility affirmed in 4 day incubation.

Reinoculate samples to fresh broth for F tests.

(#1956 – see 963) Test × W1607: Both F – !

∴ sonates inactive re F

A 1895 / 5 ml Primassay + 0.1, .2, .4, .6, 1.0 ml of 0.1% BT
B 1956 " 0.1 5% Tetr. (blue tetrazolium)
blue coloration by BT, test inhibition of small marks

ca 7/15-16. ERL pulled a single long cell from 1895 x 1956. This divided to 3 cells → microcolonies.
Only parental combinations were recorded, however. (no detectable crossovers or plasmogamy). Save
-A1 (1895 type) -B1 (1956 type).

F+ transduction via λ

963

Lysate # EML 769-9

A	1ml	in 5ml Penassay		① clear ② turbid!
B	.5	" "	+ W1956	
C	.2		"	
D	.1			
E	B	1/5	"	
F	D	1/5		
G	0		"	

1st testing at ca 6 hours.
2'd " at ~~30~~ 24.".

Lysate may have a "dormant" contamination. Streak A on Gal, NA.
① → Mal+ Gal+ (very likely the W750 Gal+ used to prepare lysate)
∴ Experiment above is confounded. Do not pursue.
Ethis crossing suspensions of treated W1956 showed some Mal+ on EMB Mal: #9 (750+) and #10. (1439)

Tentative conclusion: F transduction is due to "dormant contamination".

4P20: Inoc 1ml lysate #10 in 5ml broth
P 10A21: clear
~~[illegible]~~ ".

Repeat c̄ # 10.
1. 1ml 1/5 sterility test 48h. CLEAR. Add 1956
2. 1ml + W1956 F-transduction test
3. Inoc test of 4P20 c̄ W1956

1956 culture may be cont.

2 and 3 were both F- (x 1607).

✓ #1 : F-

July 21/1952.

See 955E. EMB, EMS/Mal.

Repeat cross EMS lac 7/29.

8/1 24 lac+ → EMBlac.

8/3 Repurify 6 possible lacv.

	Lac	Mal	EMBlac	
1	V	+ V		Mal+
2	+?	+		
3	−	−	N.G.	
4	V	−		Mal−
5	V	?		Mal+.
6	+?	−		

Save for hemizygosity tests on Mal− lacv.

H322

H310 Revertants! EMBMal.

#1, 2 Mal+ lac− no test.

6 addnl Mal+ lac− " "

see 1057

Phage tests on W... 965

Sleeves

7/15/52. Check phages: (small vial stocks)

	T1	T2r	T3k	T4	T5	T6	T7	λ-2	Bacty. X
B	✓	+	±	±	✓	✓	✓	—	
K12	✓	+ v.sm.pl.	—	+	✓	✓	✓	✓	

T2 T3 T4 should be received.

					K12	B	K12	B
T2	1.	T2:	6/8/48	Nozicks? ✓ ✓	R	S	S	S
	2	T2r	12.3.46	Lu? (contam.)	R	S	S±	S
	3	T2k	6/8/48	Nou?	R	S	S	S
	4	T2r	6/8/48	K12	R	S	S±	S
	5	T2r	7/17/48		R	1 plaque	S±	S
	~~6~~	~~T2r~~	~~7~~		R	S		

T2 should be further grown on B. EOP probably reduced.

					K12	B
T3	1	CSH 10/23/51	✓	✓	R	S
	2	7/30/51 (13)	✓		R	S
	3	9/24/51			R_s (typ. pl.)	S
	4	T3C 11/2/51			R	S
	5	T3K 7-17/48			R	1 plaque
	6	T3K 7-18/48.			R	S
	7	T3 6/8/48	A.N.		R	S
	8	T3K	A N		S	S

K-12 is clearly resistant to T3 proper. What is T3K? Test various stocks for history. Possible host-modifications?

T4	1	6/8/48	A.N.		S	S
	2	"	(11.12.)	✓	S	S
	3	7/18/48.			S±	S
	4	7/17		✓	S	S

HT5" A.N. 6/8/48. what phage? : not T1, 3?4? or mod?

	T5 AN	T5 ✓
~~B~~	S	S
~~K12~~	S	S
B/1,t	S	S
B/4	S	S
B/347	S	R
W1955	S	R
W1956	S	R
W1957	S	S
W1979	S	R

Our present T5 stock seems ok. Identity of T5 from Nozicks?!

---

cf. T3 - T3K. B, B/3,4,7; K-12, 58-161, W1678, W1590, W1485, W1918, 1956, 1802

	T3 S	T3K	plaque.
B	S	S	
B/347	R	R	
K12	R	S	
58-161, 1678, ~~1590~~	S	S	
1590, 1485	Plaques	S	
1485	S	S	
1918 1956 1802			

Note: T3K ≠ T3H. Plaques on 1485!

T3 stock is evidently inactive on

K-12 ....

It shows a limited e.op. on W1485

full active on W1918 (Y10.... acc'g Novick).

T3K was selected by novick (on K12?).

9/15... T3, T3K spotted on E coli B, 1918.

Repulsed and tested:

	T3/B	T3/1918	T3K/B	T3K/1918
B	+++	+++	~~B~~ +++	+++
K	−	±	—	± (1 pl.)
1918	+++	+++	++	+++

Question of possible host modification should be studied more explicitly. T3K originally gave +++ on K as well as B.

For testing WG series, use T3K initially; then T3 for T3K ^{S} types.

Prepare Salmonella FA. 966

9/22/52.

(A) Stock "PLT22/LT2" 10^{10} 3/52 found quite moldy.

(B) 10AM. In 100 ml Pinassay Incubate is aeration

1	1 ml LT2	+	1 ml stock PLT22
2	"		.1 "
3	"		.01 "
4	"		1 ml SW435 (LA22)

2PM. Heat shock #2, #4 60° 1 hour. Sediment. Treat supernatant i $CHCl_3$.

Test .1 ml "4" / SW435 EMB Gal

(C) Gal+ transduction

9/21 Ca .1 ml SW435 ± .1 ml stock FA LA22/LT2.

9/23 SW435 1 papilla

SW435 + FA ca 30 Gal+ papillae → Streak out 16

FA sterile 0

1st plating: apparently chan Gal+, −

2d "(+): all apparently pure +. Repick a few lightest (incl 1 ? −): 3d plating: no unstable (over)

(D). Assay FA #2 .1 ml on SW435/Gal : papillae 5

D(0) : colonies. 6

Throw out FA: sterile

(E). 1 ml each SW603 (para B0 ca 10^{10}/ml); 966-4

→ tracks and swarms small loopful

(F) SW603 5 ml/petri dish UV 9/26 (48+ hours)

No tracks / no swarms!

0 −

20 − swarm: isolate

40 −

60 −

120 −

(G) Assay 4: 800×10^8 phage/LT2;

∴ phage/FA = ca 10^8!

Gal+ papillae/ml / SW435

61/.1 ml = 610

Note — SW552 "Group D Rough" - not transmitted by PLT22/2.

966 C 16 tests of SW435 transinduced Gal +
all stable

9/29/52. Second run: SW 435 + .1 ml 966 B 4. (see G)
Pick 8 papillae (smallest or apparently mottled)
↓
↓
all stable cf. 9/23.

---

24 tests total

UV-induced O → H 966F

9/27/52

In previous run, 20 seconds gave a swarm. Repeat on a larger scale.

b — = F1

9/27 2 plates SW603 / 10 ml.

Irradiate 5 ml aliquot 25 seconds. (F2) 5 ml — (F1) 0

Concentrate to ca .5 ml Plate .02 ml / plate

9/29 UV-0. 5 plates: 3 show swarms (late develop; no tracks)

Moz to Pen. is purifying = F2, 3, 4. = all b

UV-25 secs 5 plates 0 swarms 0 tracks

Recap. (late letter to Evening)

① WG 5,6,7,8 found to ~~be~~ grow poorly on D(o)

② Indole-negative citrate positive (presumably slow)

aerogenes or intermed. (cellobiose - not gummy)

③ Crossability tests (= 776-34 = W1395)

a). C : 0 × >300/plate but all paren. (certainly not definitive!)

b)

④ Auxotroph hunt. Grew poorly on D(o), apparently resistant to penicillin. Should be repeated with D(succ)

A. Repeat SRP (interm growth)

			Mal	Lac
9/24	K-12 × 2058	+++	+, −	
1	wg6 × 2058	5	all +	
2	wg6 −	5	all +	all +
3	wg6 × 1956	9	"	
4	wg6 × 1970	1	+	
5	wg6 × 1817	9	all +	

no suggestion of futility!

B. Penicillin run. Ca. 3×10⁸/ml. 3:15 - 930

		Succ	penic.	Turbidity
9/26	1	+	−	+++
	2	−	−	+++
	3	+	100	++
	4	+	300	+
	5	+	500	+
	6	+	1000	+
	7	−	500	+

} plate: ca 300/10⁻² ml

→ But poor growth when replicated to D(o) + spread succinate 1? mutant

(over)

~~SRR~~ + SRP test do not support putativity of wg 6

Similarly wg 7 never gave a clear response.

ca 9/26 Try crosses:

W1987 x 1817
1978 x 1987
1978
1987
1978 x 1956
1978 x 2058
1978 x 1817
1987 x 2058
1987 x 1956

} on D(o)
all barren

~~Obs~~ Observations on recombinants Hfr X
TCN expts.

9/23/52.

TCN IV. 23 Lac s on EMB Lac. All tested also had Lac + S^R, ∴ recomb.

Confirst striking, all appeared to give pure +, − (no nondisjunctions).

9/26 Restreak + to look for possible persistent Lac s (like H3 10) all pure Lac +

9/28 VIII TCN. Pick Lac s colonies from EMB Lac various times to EMB Mal.

		Mal −	Mal +
1895 x 1956 Penassay	45 min	3	0
	90 "	19	0
	120 "	19	1 *
	180	14	0
	240	7	1 *

Replicate to EMB Lac sm to verify. Restreak * to verify whether they are actual zygotes or trivial conjunctions.

→ on streaking and replicating, very few Lac + S^R.

∴ these two are probably trivial sectored colonies.

FA from other Salmonella.

sensitivity tests and productions

~~188~~ 970

9/26/52.

1. Test sensitivity of var. stocks by cross-brush of broth cultures on EMB plates

	PLT22/2	PLT7/7
LT2	S	S
SW 435	R	S
SW 603	2 plaques	R
stanley	sens. sR	R
SW 553	S	S
eastbourne	R	R
Boyd 4	S	S
" 11	S	S
typhi H901	S !	R?
LT 7	S	S
coli 2	R	R
coli 3	R	R
Boyd 4	S	S
Boyd 11	S	S
altendorf	S	R
typhi Bo	R	R
typhi H901	S lytic	S lytic!

	Phage (continuum)	FA (SW 603)	
~~1A~~	1A	—	—
~~1B~~	1B	—	—
~~2A~~	2A	+++	+++
~~3~~	2B	+++	+++
	7B	+++	
	8B	+++	

} Thomsett

PLT22			
LT 1	S±	12	S
3	S	15	S
4	S	typhi V,F,	R
5	S		
6	S		
8	S		
9	S		
10	S		

Note host specificity of PLT22/2 on pair B as previously remarked. PLT22 remains the phage of choice. Grow in broth and on plates on

	①	②	③	④
	stanley	SW 553 (dublin)	altendorf	and H901
agar = A	B d 1,2	D gp	B c 7	D d
broth = B				

	⑥	⑦	⑧
	enteritidis	abony	sandiego
	D gm	B b enx	B eh enz 15

		22	7
6	enteritidis	S	1 pl.?
8	San diego	S	S
7	abony	● - few pl	R
	kentucky	R	R
	derby	R	R
	montevideo	R	R
	coli 5	R	R
	coli 1	R	R
	539	R	R
	~~typhi 901~~		

	PLT22	7	
SW 609	R	R	spont b
618	S + few pl.	R	transind b
623	2 plaques	1 plaque	transind i (PLT22/2)
H 901	S	faint S	
SW 664	R	R	
SW 623	1 plaque	R	

Although H901 is readily lysed by PLT22, no satisfactory preps. have been obtained via liquid culture. Adapted plaques should be used. Ditto for stanley.

B g - derby carries carrier

D b - marinum

D e - eastbourne

= Prepare PLT22/603 609 618 623 stanley & single plaques from PLT22/2. SW 603 and derivatives are evidently poorly sens. to original PLT22/2

phage - lysogenicity tests on

stanley LT4 / plaque. PLT22++

H901

eastbourne LT10 several, (LT1, LT3) probably severe lysog.
PLT22 very slight

typhi 60

Boyd 1104 eastbourne++) but filtrate acted on neither

A B C D

Layer plating test for lysogenicity: did not work too well owing to overgrowth.

Plaques:

	1	2	3	4	5
A	stanley	[illegible]	[illegible]	[illegible]	
B	LT1				5
C	LT6				10
D	PLT22+	+	+	+	

FH / Salm. misc. 970

B
d 1,2 (1) STANLEY, typhi ...
Repeated efforts to grow PLT22 on stanley, typhi H901 or 60 have failed, despite plaquing on agar.

~~(4) S. typhi H901~~

(5) heidelberg 5a LT-7 5b LT-22 n.g. Repeat: ok. ... See 971D5 ...
PLT22 / heidelberg

? eastbourne
coli 1-5
london dub by montevideo

SW694 — resistant to PLT22, PLT7

not FA against SW666.

Motility transductions 971 A

9/29/52.

A SW603 / motility agar

B SW435 / " + typhimurium serum 1:500 (~~[illegible]~~) (OK).

C styphi H901 / d-antiserum 1:500 ~~[illegible]~~ (inadequate); 1:200

A: see SW666

1 + "FA 970-1A, B". no tracks no swarms (FA: no phage)

2 FA 970-2A, B numerous tracks and swarms. Streak out and

Keep non-b as 971A-1,2 ~~+ SW663, 662~~ ✓✓

b 971A-3 test single colonies: Search 2A, 2A' < 2A: 1 b+++ ~~[illegible]~~ / 2A': 1,2,3,5 b:+++ later shown

Check non-b on available sera: b, i, d, e, 1,5; 1,7, 1,2 Inagglutinable GP but motility OK under scope

9/29. 7. Swarms + tracks. 2 plates Test à b, e.

a } 16 isolates all b. = 971A4

b } = 971A5

2/29 8. swarms + tracks. 2 plates.

a 7 isolates all b

b 8 6 are b 2 are ? (#5, 8.) A7; A8, ~~[illegible]~~ SW664 abony / sandrijo

possibly IX XII enx - ?

react c̄ enx 1:100 abony rx mostly b, very faint enx

1:10 reacted c̄ #1, etc. (nonspec ?)

	b	e
abony	+	+
sandrijo	~~[illegible]~~	+
para B	+++	

e serum?

Repeat 7 for non-b

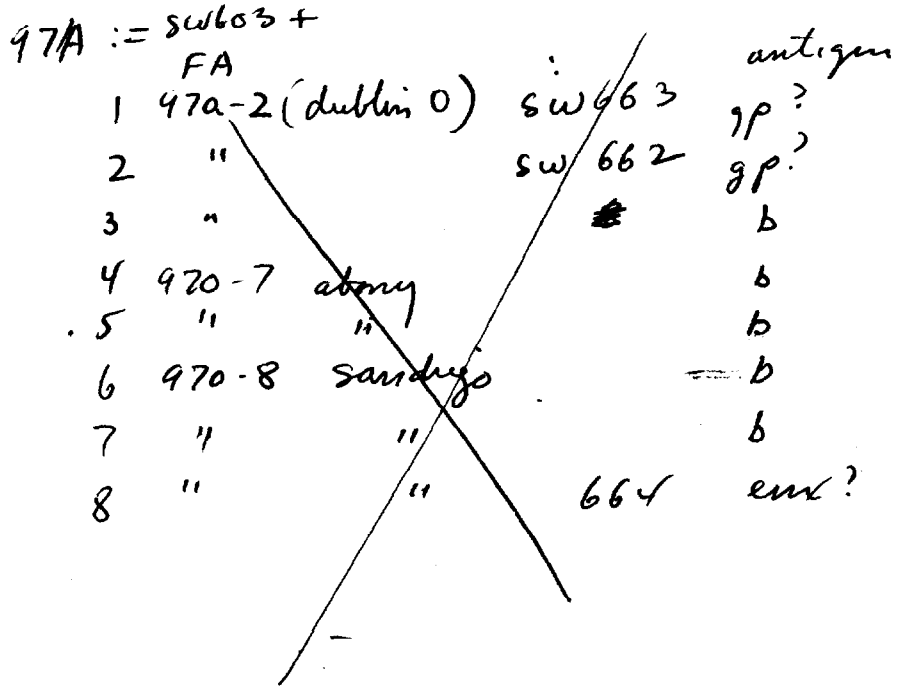
3. few tracks; Swarm

6. " "

Notes: SW603, A1, A2 are strong Xyl+ SW553 is v. weak ±.

see over for stock tab.

97A := sw603 +


FA
1 97a-2 (dublin 0)
2 "
3 "
4 970-7 atony
5 "
6 970-8 sandiego
7 "
8 "
"
"
sw663
sw662
664
antigen
gp?
gp?
b
b
b
b
b
enx?

971A

Recap.
10/2/52

A SW603 [= para B/O)
B SW435 + tymor serum
C S. typhi H901 / d serum 1:200
D SW666 = SW603 Gal-

A. 1. FA 970-1 Inactive

(dublin) D gp ✓ 2 970-2 2 plates, 5 colonies each. 1/5 : b 4/5 : b
others presumably gp to be sent to Chamblee to verify. Save 2 nas-b
as SW-663,662 = 971A1, A2 resp. ~~Save~~ 1 b as 971A3.

altendorf B c;1,9 3 970-3 1 plate: several fearks and swarms. Streak out and
test single colonies. 10/10 b

4 970-4 H901. No satisf FA to date

B r;1,2,3 5 S. heidelberg

enteritidis D gm ✓ 6 970-6 1 plate: as 3. 6/11 b, save 5 nas b
[illegible]
[illegible]. see D6

10/2/52

971A

abony B b enx

7. 1st test: 2 plates many T and S (tracks + swarms). 16 isolates all b.
parent culture is largely in b phase [anti b serum would be invaluable).
2d plating 10/2/52 4 plates. 8/8 single colonies: b. save 2 b as 971A-4 and 5
Inoc #1 (strong b) #7 (weak b) and mass into b agar. Also abony to isolate enx phase.

10/4. Both 1 and 7 were blocked on b agar, gave 2-3 swarms = 971A7-1B, -7B.
[abony swarmed out directly (single cols. should be compared).]
[also cf. sport rev.]

	1	7
B: orig.	b	b

3 swarms ea. not b, 12, enx ... ??

T.O. all extraneous cultures (probably z_{33})

8 2 plates many T+S. 7/7 all b 6/8 b 2/8 reacted c enx at 1:100
This serum aggl. para B at 1:10. Rx not very strong but confirmed microscopically. ~~Save~~ 2 b as 971A-6, 7. 1 presumptive "c" as 971A8 = SW664 (✓ ch: —)

San diego B ch enx 15

Antigen transductions to typhimurium 971B
SW 435

Sept 29 - 1952

B 2 9/30 2 plates no buds 10/3 2 buds replant to i, 1, 2
(dublin)
D gp
#1 did not grow out well on i. #2 maggl. i, b, 1,2 (trace rx i each). Presumably gp. Phasicity?
not Salm on EMB
(Gal-) SW674

B 7 9/30 4 plates no buds PM 1 swarm. Crew poorly. probably contaminant
abony
B b enx 10/3 2 buds. Pick to broth #1: b #2: magg: b, 1,2, enx, i
isolate as SW672. Plant on b
contam (not Salm).

B 8 9/30 4 plates no buds 10/3 no buds or swarms
san diego
B eb en 2,5
1 plate: filled i growth Contam?

B-0 Immobilization OK on 1:200, 1:400 1:800 H-tymus antiserum
In 1:500, para B grew well. However, buds are growing out very poorly!

Antigen transductions to S. typhi (H901) 971C

Sept 29, 1952

C - 0 9/30 H901 not completely suppressed: dense spreading growth may obscure some swarms.

C - 2 9/30 1 plate no buds PM 1 bud-swarm → maggluit b, d, i
D gp
Isolate as SW-~~665~~ 667

C - 7 9/30 2 plates 5 buds 1-5
B b enx
all 5 are b (abony parent pred. this phase)
Isolate as SW 670

C - 8 9/30 2 plates no buds PM: 2 buds? ✓ → swarms
B ch enz15 both react with enx (d?) Purify and isolate as SW 668-9

C - 15 (abony 2) cf. C7. 10/25 - v. faint turbidity away from moc. Pick + test

C - PLT22 1 plate 2 (or 3?) buds. → 8 swarms later
B i 123
3 isolates: all 3 are i-, not ~~b~~, d; 1,2 ~~I~~ same as 971C22

no trails associated with the buds. Moc buds to fresh plates, broths

c5 (anti D inadequate) but no kudsor swarm

transductions to paratyphi B
SW-666

971D

~~#~~ 10/1/52

D7: many swarms pooled: all Gal- on streaks. 5/5 b

same for phase test — Mocmass to b agar. → several buds. Pick for test (D7B:1-)

10/4 B. Plant individual colonies from original pool to b agar. (two kinds of diphasic ??)

→ 3 swarms: inagglutinable b, 12, enx

1 ~~so~~ does not swarm on b agar 2 swarms slowly.

: D7A2 gives weak reaction after swarming !! enx ?

of A7 streak and save D7B1 Rechuck.

4/6 b strong = D7A1 = b
2/6 b weak! D7A2 b

Rev D7A1 A2 and B0 give weak b SW677
B1 inaggl (2d phase?) 678

c 1,7 D3 10/3 From swarm from transplant to b agar. → non b, i. 1,7 ?
streak out and save as SW675. Z33

10/5. Second run, plant swarms to b agar and save selection: slow outgrowths
(probably second phases again) < 1 - still b
2 - non-b Z33

D3: ~~a : c ✓ repurify as SW903~~
a, b : not c. (probably "j") 3/5/53: D3b = b

T.O.

r; 1,2,3 D5 10/3 - FA mg. 10/?.. 3 buds → streak and test single colonies. see SW683

D15. 10/24. Numerous single swarms. Pick + test : see 979-4

D18 (LT-2^II) 1/8/53. FA18 → x666. 5i 52b Note concordance
with 22 → x666 see 986. 4/5 were Lp^s.

D6 10/7 3 swarms. 2 non-b 1 b. Save non-b as SW 669
FA39 (secondary) 12/4/52. 30b: 2a (22?: 4 mixed) swarms may
have been mixed. SW & some rough

3/4/53 "weak" and "strong" b have not been carefully studied. May represent
fractions of b? Throw out

#3

533 → 534 $\xrightarrow{\text{sp+}}$ 588

para B 0 (1,2)

note: 588 still susc. like 533

543b+ are resist

S W. cultures.

RACK 1

SW			
351	538	548	566
414	539	549	567
435	540	550	568
530	541	550R	569
531	542	552	573
534	543	553	574
534R	544	556	576
534	545	558	577
536	546	563	578
537	547	565	579

RACK 2

SW		
580	600	628
584	609	629
586	618	630
587	619	
588	620	
595	621	
596	622	
597	623	
598	625	
599	626	

RACK 3

SW			
653	669	680	696
654	670	681	697
655	671	682	698
656	672	683	699
662	673	684	700
664	674	685	
665	676	687	
666	677	688	
667	678	694	
668	679	695	

RACK 4

701	770R	829	LT-2 I
703A	771	834	Staphi VI FI
703B	774	837	LT-2 II
704	775	839	2nd PRE TO
704B	776	840 A	LT-15
706	787	840 B	SY 79
715	791	842	LT-12
721	803A	862	
760	803B		
764	825		

RACK 5

901	913	923	933
902	914	925	933
904	915	926	934
905	916	926 new	935
906	917	927	936
908	918	928	937
909	919	929	938
910	920	930	939
911	921	932	940
912	922	932 b,2	941

RACK 6

942	954
943	954 +
944	956
945	979
946	
947	
948	
949	
950	
952	

FA56 (SW960) appears inactive re F1a, (SW666; SW967)
FA55 and FA57 → SW666 give b only
~~FA~~ → SW967 " swarms:

(57 → 967 swarms unusually slow)

3 trials each FA54 → mainin (d, a, 123 serum) to secure z_6 phase gave only 1,5 ; 1,2 resp. FA54A is designated as $\underline{z_6}$ but should be reinvaid.

missing:

A	B	C	D	D
1394	112	1827	1728	618
1177	125	1969		
1895	1368			
1922	1606			
1918				
H 310				

pulled for re nerdal

8848,
1979
1895
1896
1590
1635
518
1325
1452
1618
884
961D8
760
1015
1022
1023
1024
1033
1327
2069H+
1674
1666
1649
1688
1517
1529
1541
1585
2069p+
1832
1852
1846
1903
1920
1939X
1970
2020
2019
1957
Y 28
679
1801
383
1742
1813
1730
1734
1729
543
513
404

X-phage

9/29/52

Survey host range by cross-streak on EMB Lac:

1	ty[illegible]	R	11	SW541	R
2	S[illegible]		12	enteritidis	R
3	[illegible]		13	Boyd 1404	R
4	Boyd 1411		14	coli 5	R
5	coli 3		15	stanley	S
6	LT2		16	derby	R
7	altendorf		17	london	R
8	eastbourne R		18	SW553	R
9	sandiego		19	montevideo	R
10	abony		20	eastbourne	R

(2)

1	coli 2	R	11	LT15 #292	R
2	coli 1	R	12	12 114	R
3	H901	RS	13	1 ✓ i, 1, 2	S
4	SW435	R	14	3	R
5	"coli 4"	R	15	4	R
6	SW539	R	16	5	R
7	typhi Vi F1	R	17	6	R
8			18	8	R
9			19	9	R
10			20	10	R

check motility of S, RS. 9/30

A

1	971 "A2"	R	F 1	R	634	R	645	R
2			2		635		6	
3			3		636		7	
4	=A1		4		637		8	
5	A3		SW569	S	638		9	
6	"A2'"		653		639		650	
7	=A2		562		641		51	
8			561		2		52	
9			572		3			
10			588	SR	4			
			547	SR				

C

reported sensitive to adapted X

B

1	592	R	6 16	R
2	594		6 17	
3	546	S	6 18	
4	609		623	
5	610		25	
6	611		26	
7	12		28	
8	13		631	
9	14		32	
10	15		33	

Records on SW543 b+?

Notes

Stocker reported that SW588 was more susceptible to X grown on SW592.

H901, H901 i both S

S479; S479 i both R

History of 588: ~~[illegible]~~ 544 (tymer 0 form) $\xrightarrow{\text{spont +}}$ 588 ∴ original strain seems. ~~[illegible]~~

χ-phage

10/7/52

Compare χ 942-1 and regrown on SW592

	χ_{or}	$\chi_{\text{or}\,92}$
609	R	R
610		
~~673~~		
617		
618		
623		
625		
633		
LT1	S	S
LT2	R	R
SW435 ~~LT22~~	1	1

no improvement.

SW543 line seems resistant.

Typhi derivatives:

Later, recheck φ. χ / SW592

		942-1	χ/592
	703	+	±
	588	++	±
	633	±	−
(typhi)	537	+	+
	976-5A.	−	−

Fermentation markers in Salm. 973

A SW541 5EMB Gal 3 Xyl 8 sec UV (°K).

1 each?? — Gal n.g. Xyl — looks ok. SW665

excellent mutant: no transduction, papillae are clear in 48 hours.

Apparent rather high yield compared to SW435. (should be compared closely)

Plate c̄ PLT22/2. Pick papillae + streak to test stability

12 papillae. Streak to give large but poorly fermenting colonies!

Compare 541, 665 and papillae! (possible a Xyl + weak allele, in this background).

In various tests, inconsistent scoring. Best result: incubate at 37, then room temp. No sign of segregation — not many tests

B SW603 5 Gal 1 Xyl

↓

1 sectored col.

↓

good Gal — SW666

excellent mutant as above. Pick papillae + PLT22 → & streak to test stability.

plaque ridden (e.o.p. ca 10^{-4})

considerable dimorphism on gal, especially after UV, not remedied by repicking large colony.

8 papillae. 1 appears Gal v = 973B1 Restreak several colonies.

All others appear stable.

10/9 Pick addnl. 16. (all large papillae only). All + appear stable on 1st streaking. Restreak: no — sign, but # 2c shows some mottling. Review this as 973B2: repeated + and + mottled (phage?). Repeat once again:

→ appears to throw stable, mosaic types. Repick B1A to look for new stable, "stable" ✓ 1 colony is stable.

Streak of B1A mosaic Gal v → −, v, +. Check +, restreak v. → pure + → −+v

∴ This unstable transduction segregates pure + and −.

~~Gal~~ (over) Isolate pure + and −

973C PLT22 + SW665

973D ⊕ SW541 + SW665 Xyl.

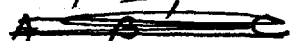
973E PLT22 SW435 Gal. – 20 : all pure + !

D) SW541, transductions of SW665 behave very peculiarly on xylose. Sharp, Xyl+ colonies give – restreakings; SW541 itself gives inconsistent responses on EMS xylose, but not to cause a false negative reading. Only one restreaking gave a typical + reaction. 24 tests 22 – 1++ 1 wart + !

These colonies gave typical + reactions on original plating !

Possibility : interaction with SW655? Try mixture.

Save 1 plate which carries 1++, 1+, 2 –.



973 B

Efficiency of transduction and effect of serum.

FA is PLT22 966-4.

Mix .5ml ca 10^{10}+ cells/ml with .5ml FA (or dil. as ind). 966-4

Let stand 5-10 minutes. plate .1ml.

1 = SW665 on EMBXyl Note self plaquing!

2 = SW666 EMBGal

3 SW435 EMBGal

---

	papillae / ml FA →					
	.05	.025	.01	.025 + .12 serum .02ml	0	
SW665/Xyl	477	218	40	31	1	self plaq.
SW666/Gal	15	6	5	[illegible]	0	
				.5 + .02ml 0 serum.		
				less heavy plaquing 16		
SW435/Gal.	53	50	31			

heavily plaqued all plates. obvious reason for nonlinearity

why non-linear?

$\frac{8000}{8000 \times 10^7}$

JAN 26 1955 This calculate at only 10^{-6} papillae per phage of BADS claim of

Spara B/0 serial transduction 974

10/4/52

Streak out PLT22/2 = FA22 on SW666.

Pick single plaque to broth = FA9

streak out FA9 on SW609 = FA10 (masked plaques)
618 = FA11
623 = FA12

10/2 974-1 SW666 + PLT7/7 → 1 track - swarm: i

Purify again and keep as SW671. No swarms on i, 1-2 agar. (i alone ??)

11/5/52. SW666 + FA as indicated

			gross agglut. b	gross agglut. i	plant on agar anti b	plant on agar i
9	No T or S	(o)	no swarms			
10	Many T+S	(b sp.)	+		−	++
11	"	(b transd.)	+		−	++
12	"	(i transd.)		+	no swarm on i agar	−
22	"	(i)		+	" " "	++ ++

why PLT22 → x 666 different on i agar than FA12 → ? (too few for full sample?)

(over)

Pick swarms to broth. Streak out on EMB Gal to ✓ joint transduction. all Gal -.

Plant on i and b agar to screen for diverse types

After 48 hours, second phases appear i 10 and 11.

22: 5/7 i
2/7 b

Do not keep

B.

1	FA12 + SW609	grow together on agar; plant on b motility agar.	many beds → 5 tested all non b, i	
2	+ SW618	b	6 beds/plate → 4 non b, non i	
3	+ SW673	b	many beds → i. streak out ✓ SW682	
4	FA9 + SW623	i-12	ca 10/7 no beds	10/10 2?
5	FA9 + SW435	i-12	no beds ...	solid
6	FA10 + SW623	i-12	no beds	0
7	FA10 + SW435	i-12	no beds ...	0
8	FA11 + SW623.	i-12	no beds	0
9	FA11 + SW435	i-12	no beds ...	10/10: solid growth

not b or i

Repeat series using larger amts. of FA.

i 12 screen n.g. for this

Repeat 12.

10/5 4+ swarms from 2 addnl. plates
to i,12. no swarm.

10/7 4 addnl. plates: isol beds. 6 to i,12
2 showed slight movement (moisture smear?):
both are b

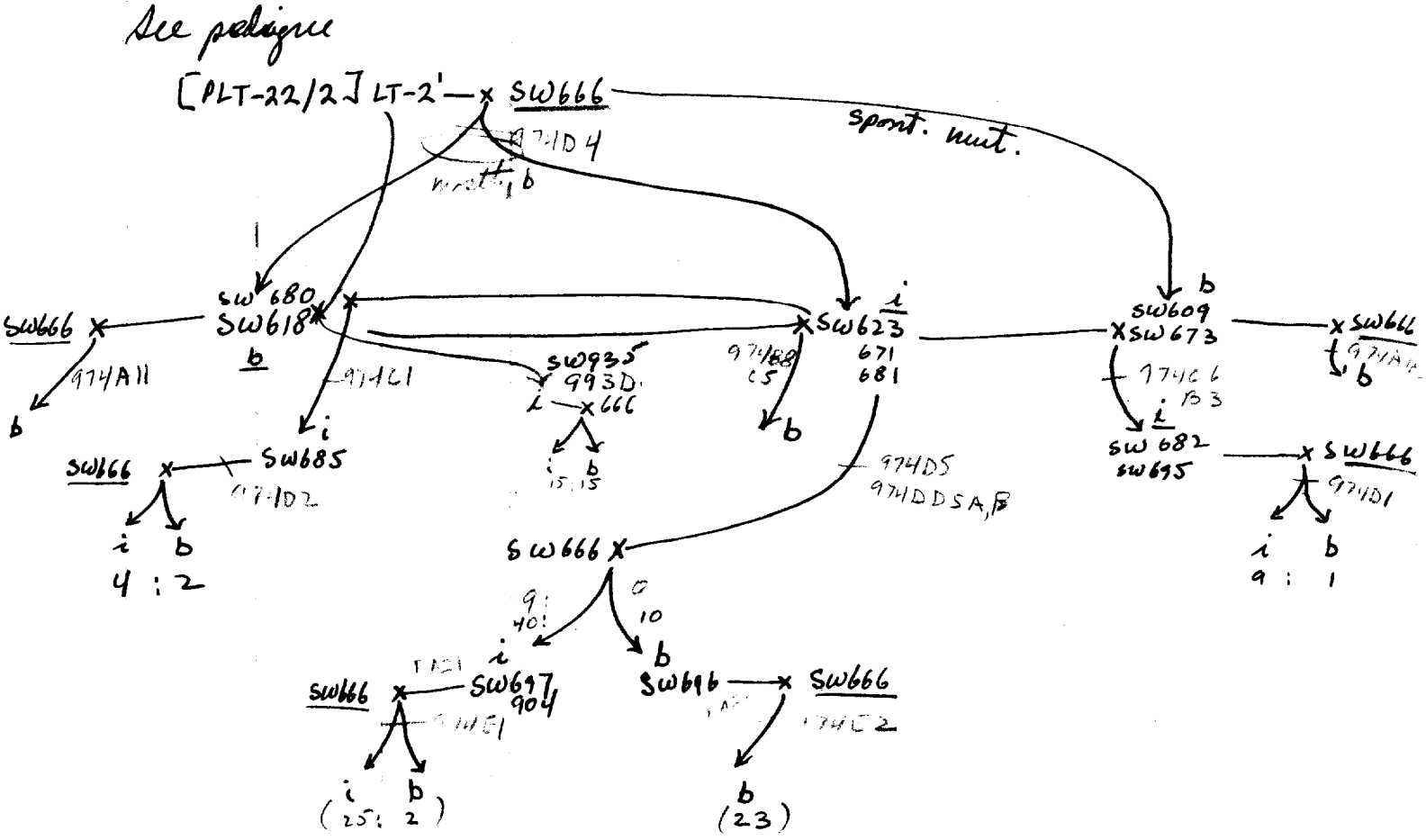
others remained stationary.

10/8 18 addnl swarms from 2 large plates (including pooled swarms)
all stationary on i agar.

Total 30 swarms: all i. ?

Progeny tests on SW666 x — i

See pedigree



Note: 974DD5B

① seems to show purity of individual swarms, but controls may be indecisive.

② In three cases tested, failed to isolate O-forms from initial flare. Trades themselves not tested. Established growth seems to be inhibitory to remoculations. Maynard bitter methods: see 680

974 C

	FA	SW		plate in	
1	12	609	$A^i \to H + A^b$ sp?	b	no buds. cf B3
2	12	618	$A^i \to H + A^b$	b	→ i ✓ pur. SW 685
3	22	618	$A^i \to H + A^b$	b	→ i SW 686.
4	11	435	$A^b \to H + A^i$	i	} no buds! → 1, 2
5	11	623	$A^b \to H - A^i$	i	} swarmed: → i ✓ isolated colony i
Repeat 6	12	609		b	→ i SW 695
7	22	609		b	no buds
8					
9					

i.e. "swarm" is still i

C5. 10 buds 9/10 b 1/10 still i or mixed.
naturals.

974/C3. Repeat 11/21/52. Use relatively light bacterial inoculum: this seems to work much better. Numerous swarms (not well separated when finally seen). Includes 2-3 flares. Pick these as possibly representing segregation of b.

C3A flares (fundam from contam. by blocked b not certain.) Grow in Pinassay, test grossly and select on i agar. : all i

C3B Distinct swarms for an Lp^s i: 15 - all i, Lp^s Prepare FA. (#10, #15 may be Lp^t) See 993.

4/5 blocked on i 1/5 → b. (possibility of contamination). In further experiments use added R cells as a contamination marker. (e.g. 666R).

after pul. test save #1, #4 as SW9

FA from two-step transductions 974D

10/13/52

1. FA16 (SW682) + SW666. only 2 swarms → both i
2. PA (SW685) " plant on i, b agar
3. " (C8C) no "Tot b!" "
4. " PLT22 " many swarms: pool and streak out
5. FA 12 (623 = trans d i) " individual swarms pulled out

1 Repeat: mixed swarms - no motility as i - agar.
test pool growth: (2)
a no agglut! → b, i ± as controls.
b: wanted b, (i ±?)

2. " as above (2): do.

i and b!

although no swarm on i.
Perhaps medium n.g.
Would acct. for failure of C4-5.

Immotile as before on old (i, 12)

5 i i b b i b i i i i i i b i i i i b

4
12 i
5 b

This experiment was recalled to be confused, and these results bear it out! From previous experience, 4 should give a mixture of b and i, 5 i only.

REPEAT 10/17.

4 b+i?) pool was streaked out. Test colonies: 14/14 b
7/7 b pool rx b, not i
7/7 b test rx b agar

This is actually mixed b, i

migrated on b agar

D1 separate T+S 7i 1b

D2 1 plate T+S pool - to both: reacts i ++ b + . Colony tests from strain: 4i : 2b

D3 No T or S need new PA

D5 ~~both~~ individual swarms: b and i ~~8~~ Total

i agar bagar

b swarmed. 8 19

i swarmed.

previous difficulty not reproduced.

D2 affirms F-2 progeny test of limited transduction

974DD

10/21: Reisolate SW623 single colony (i) and prepare FA (FA9 + ...)
FA(SW623) + SW666 → many T+S. (very well defined on large agar plates)

5A Pick reasonably individ. swarms + retest b or i:
10 : i 1 : b (weak, maybe b + i) → purify and retest

5B. FA12B → SW666 on motility agar, well separated swarms and tracks.
Count: 14 lateral swarms (mostly i tracklets) 46 tracks. In many cases, the tracks flare out into swarms. Pick well isolated swarms near origin for later selective test.
Pick flares + streak out to reisolate presumed O component.
see over.

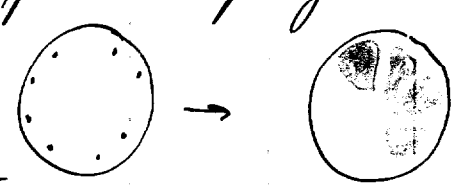
at least 2-3 mm. from streak "FA" from SW686 (2 batches) – no T or S. (check sensitivity to PLT-22).
SW666 controls No T or S.

→ Spot on i (or b) agar to test purity. 5/5 i immobilized.
6 add tests spoiled by bad agar, but repick. Also single b slapped around and retest → all are immobilized. ∴ each swarm was pure.
To check, spot i on b plate and v.v. i/b swarms. b/i grows poorly (plate dried out?)
Repeat test by serial monitum.

6. FA18 (PLT22 / LT2 phase II 1, 2) + SW666.
2/2 b.
16/16 b.

Total 18/18 all b
no 112 ?!

Spot 16 on b agar: no swarms. typical slow spr. growth, some mutual inhibition:



Conclude: these are monophasic
b identical with SW618.

(over)

see 979

5B: 3 flares streaked out.

15 colonies tested from each: all i no O forms!
(45 tests!)

Perhaps the flares are not actually O microcolonies!

---

12 isolated birds tested: 11 → i
mouth from near source 1 → b (#12)

(as over) Spot on NSA., transfer to homologous serum agar.
all immobilized. ∴ each swarm is pure

This agrees with 15/15 individual tests on each of 3 swarms.

Note 11/8. SW688 (Boyd's rough) gives very coarse flares on motility agar, prior to later "smooth" swarm. Reisolate and compare morphology, swarms & original unselected.
This may be the basis of flares. see 978

plaging tests on SW666x — i, b

974E

FA21 → SW666 1st test: again too sloppy 2d test activity?
Repeat preps: try FA9, LT7.
new FA9 prep. apparently lysed but activity still poor.

---

new PLT7 prep. inactive. — v. few Tracks. 2? swarms. but might be carry over?

Test other hybrid i's for susc. to PLT22. cf. 697 which is app. resistant (but lysed?)
e.g. 974DD5b: 1–11 and SW697. All show no lysis on plate
with FA9 except #6. = SW904. However in both 697 does seem to be inhibited by FA9!

FA21A (904) → SW666
27 swarms. 25: i 2: b → 1R, 1S

Note: in 974DD5b of 11i, 10 are LP22R 1S
here of 25 21R 4S / 31 R : 5 S

2 FA20 → activity? / a number of T+ swarms developed later. Maybe contaminants: grew poorly on NSA
as above. 1 swarm → b.
new prep. (FA9) → T+ swarms; fairly separable.
22 → all 6 (not i) ∴ 23 total
Test on FA9: 1S 21R. cf 986

Serum effect on transmotilization.

~~974~~
975

October 4, 1952.

Many fewer swarms occur in antigen substitution experiments (e.g. typhi x typhimurium) than in "transmotilizations" (eg. SW543 x typhimur), and it seems unlikely that this is due to a lower frequency of occurrence. The SW543 transinductions allow a comparison of the techniques.

10/3. Plant D22 (SW-666 + FA PLT22/2) on motility agar ± b, i/2.

No serum: many tracks and swarms (ca. 60 tracks, diff. swarm)

i: 1 bud } A4: transplant — to ~~E~~ i

b no buds, no tracks. }

Could this effect be due to O-antibodies? Absorbed sera should be used.

Also cf. 971D.

D3: 19 ~~buds~~ tracks. heavy swarm D3/b: no buds, swarms or tracks.

Repeat P4, D3/-, /b, /i (to test specificity of i effect)

A5: D3/- swarm; ca 15 tracks

D3/i 1 sw ca 12 tracks.

D3/b No tracks or swarms.

D3/d T+S as above.

In this case, b is specific. cf 974

$D3_0$/b 1 swarm. see D3. Inagglutinable.

NOTE. Inagglutinable phases might come either from FA or alternate phases in SW603. Occasional buds appear on each plate of SW666 H+/b, e.g. SW673 → inagglutinable by b, i, 1.2, enx... Furthermore abony/b (occurs very readily) = enx shows very weak reaction with our enx serum!