NOVICK

OCTOBER 1951

2	280	270	260	250	240	230
Bact+Super	775	925	1.10	1.35	2.60	
Fmed	052	057	.076	.10	.18	0.70
Δ_{B+S}	.723	.868	1.02	1.25	2.42	
Super	520	615	.740	1.10	2.18	
Fmed	052	057	.076	.10	.18	0.70
Δ_S	.468	.558	.664	1.00	2.00	
$\frac{\Delta_{B+S}}{\Delta_S}$	1.54	1.55	1.54	1.25	1.21	

10/10/51 Took supernatant of 10/9/51 from 297. Added HCl to pH 5. Incubated 37° overnight

350	300	290	280	270	260	250	240
029	.108	.378	.619	639	562	475	

284 – 10/6/51 – Saturday

at 5 pm – added 1 more pin (8 → 9). Should give τ = 1.93 hours.

10/7/51 Sun at 5 pm measured τ = 1.96 hours

Assay + T1 $\begin{matrix}119\\132\end{matrix}\ \begin{matrix}143\\146\end{matrix}\}210^5\ \begin{matrix}212\\240\end{matrix}\}10^5 = 2.48\times10^7$

+T6 $\begin{matrix}256\\251\end{matrix}\ \begin{matrix}234\\251\end{matrix}\}10^6 = 2.48\times10^8$

Changed τ to 2.95 hours (9 – 6 pins)

10/8/51 Mon τ at 3 pm = 2.99 hours

Assay at 12 noon

+T1 $\begin{matrix}213\\206\end{matrix}\ \begin{matrix}193\\199\end{matrix}\}210^5 = 4.06\times10^7$

+T6 $\begin{matrix}248\\243\end{matrix}\ 255\}10^6 = 2.49\times10^8$

At 3^{15} pm lowered pH to 5.7 – switched to 4 pins from 6

10/9/51 Mon 11 pm +T1 $\begin{matrix}251\\258\end{matrix}\}210^5 = 5.09\times10^7$ should give τ = 4.5 hr

+T6 $\begin{matrix}243\\257\end{matrix}\}10^6$

10/9/51 Tues 11^{30} am +T1 $\begin{matrix}235\\236\end{matrix}\}210^5$ 483 $\}10^5$ =

+T6 245 $\begin{matrix}268\\258\end{matrix}\}10^6$

Bact + Super vs H_2O

350	300	290	280	270	260	250	240	230
.280	.440	.596	.775	.925	1.10	1.35	2.60	

400 Super vs H_2O

.015	.116	248	375	520	615	.740	1.10	[218]	315	325
									.030	.184

x2 dilution → .54 1.09

Sample from 11 pm last night Super vs H_2O

350	300	290	280	270	260	250	240	230
095	203	305	430	.515	620	.870	1.70	very large

Sunday Sept 29 /51

Sample from 284 running at about 4 hrs collected at 2 pm centrifuged [illegible].

Obs. at 280 nanos H_2O

0.246

pr M prems ant

9/30/51 Sunday 12 noon
+T1 271 247 236 259 $\}210^5$ $= 5.06 \times 10^7$
+T6 307 253 275 264 $\}10^6$ $= 2.75 \times 10^8$

τ measured 9/31/51 = 3.75 hours

10/2/51 Reautoclaved, reinoculated B/1.t at 5 pm 10/2/51
Ran overnight at τ = 3.1 hr - 6 pins
10/3/51 Switched to 9 pins (τ=2) at 10^{15} am
Assay B/1.t 227 212 $\}210^5$ $= 4.39 \times 10^7$

Inoculated D84/6 at midnight
Assay B/1.t before inoculation of D84/6 187 173 212 208 $\}210^5 = 3.9 \times 10^7$

10/4/51 Thurs Measured τ at 7^{30} pm = 1.85 hours
Removed 1 pin. This should give τ = 2.06
10/5 Fri 11am +T1 270 308 $\}10^5$ $= 2.89 \times 10^7$
+T6 85 108 $\}10^6$ 164 137 $\}510^5$ $= 8.6 \times 10^7$

10/6 Sat Measured τ at 3^{45} pm = 2.17 hours
4pm +T1 119 104 $\}210^5$ 238 249 $\}10^5$ $= 2.44 \times 10^7$
+T6 268 290 $\}10^6$ $= 2.79 \times 10^8$
Beckman @ 350 H_2O = 0.193

Continue on preceding face

284

9/26/51 Set up chemostat with 100 γ/l tryp and 2.3 mg/l arginine.
Inoc with B/it on 9/26/51
Ran at τ = 6.5 hr

Assay 2pm 9/27/51
218 218 } 2.10^5
270 270 } 2.10^5
215 215
239 239
942
2

~ [5 10^7] l. h. = 4.71 x 10^7

Inoculated with 084/6 (arg⁻) ran at τ = 4.8 hr overnight. In morning (9 am) changed rate to 2.5 hr

At 4pm Beckman = 0.178 vs H_2O @ 350

Assay +T1 162 161 145 } 2.10^5 323 292 615 = 3.08 x 10^7
+T1 147 } 2.10^5
9/28/51 +T6 253 248 227 } 10^6
7^39 pm +T6 268 } 10^6 = 2.49 x 10^8

At — pm on 9/28 — chged from 8 pins to 5 pins
Beckman

9/29/51 Solenoid valve broke down (~ 2am). Volume collected ~94 cc
Sat. during the period from 3^55 pm 9/28 to 12 noon 9/29.
Analyzed at 12^30 and resumed operation with
τ set at 5 pins ~~(4 hrs)~~ (3.6 hrs) measured

12^30 pm +T1 75 81 } x 10^6 300 387 } x 2.10^5 = 7 x 10^7
+T6 159 148 } 2 x 10^6 271 289 } x 10^6 ~ 28 x 10^8

10 pm +T1 287 286 274 244 } 2 x 10^5 = 5.46 x 10^7
at τ = 3.6 +T6 258 293 } 10^6 = 2.76 x 10^8

10/5/51 To 10cc B @ 10^8 added 2cc T2r (2.7×10^9), $\frac{1}{10}$ cc of $\frac{1}{100}$ T2rh

A) = 10^8 B, 5×10^8 T2r, 10^6 T2rh

B) Ditto above but used 1cc of $\frac{1}{100}$ T2rh

10^8 B 5×10^8 T2r, 10^7 T2rh

Incubated 1 hr @ 37° – susp lysed.

After lysis expect in A ~ 10^{10} T2r, 2×10^7 T2rh

" " " in B ~ 10^{10} T2r, 2×10^8 T2rh

To B @ (10cc) 5×10^8 add $\frac{1}{10}$ cc of each stock in sep tubes

A

5×10^8 B, { 10^8 T2r; 2×10^5 T2rh

5 min ads

Add T1 (1cc of 10^{10})

5 min ads

Plate on B/2/5

9, 9 } 10^3

316, 360 } 10^8

49, 41 } $2 10^2$

B

5×10^8 B { 10^8 T2r; 2×10^6 T2rh

5 min ads

Add T1 (1cc of 10^{10})

5 min ads

Plate on B/2/5

522 } 200

9, 12 } 10^4

101, 87 } 2×10^3

~~Plate on B~~ ~~C~~

C

~~Ditto A, but no BoT1~~

Plate stock A on B/2/5 directly

having made 1:100 dil first

6, 9 } 10^3 30, 23 } $2 10^2$ 4, 4 } 10^1

Similarly on B+B/2/5

~~Plate on B~~

D

Plate stock B on B/2/5 directly

Having made 1:100 dil first

7, 8 } 10^4 55 } 10^3 337, 300 } $2 10^2$

Similarly on B+B/2/5

10/2/51 Unmasking expt on T2h

Used T2 r (in F) obtained from T2 Kozloff

I 0.83 cc B @ 10^9
0.17 cc T2r stock
incubate 5 min 37°
Add 1 cc T1 (10^9)
incubate 5 min
plate ½ cc on B/2/5⁻ 0 4 } ≅ 2/10^8
= 0.17/4 = .0425 cc = 1.15×10^8 plated

II ditto above but used broth instead of B. 0 0 } ≅ 0/10^8

III ditto I but added broth instead of T1. 13 22 } ≅ 18/10^8

IV Assay T2r on B 268 }10^7 = 2.68×10^9

Next expt.
1) B + 5 T2 + 1/10 T2h
B + 5 T2 + 1/100 T2h } grow stock
then plate as above

Made stock of T2rh by picking h above + growing in 1/2/5⁻
Bef filt assay on B 143 }10^8
on B/2 123 }10^8

Glucose B/t

291

10/4/51 500 c/c typ 2gm/l glucose + (F-lactate)
Inoculated with B/t on 10/4/51
Flow rate set at $\tau = 4$ hr

10/5/51 Collected 11am – 12^{30} pH was 6.8
Supernat bact vs med Assay ($\times 10^6$) = 1.23×10^8

350	300	290	280	270	260	250	240	230
.109	.164	.310	.437	463	403	332	317	363

Supernatant alone vs med

350	300	290	280	270	260	250	240	230
0.015	0.055	0.215	0.360	0.360	.275	0.179	0.150	0.355

$\frac{.360}{4} \times \frac{140}{109} = 0.116$ / hr [illegible] I have beat that!

We shall add glucose to bring up conc. to 6 gm/l ; added at 6 pm.

Friday

292 (Trypt. Ckd)

500 γ/l typ + 9.3 mg/l arginine + F

Inoc 084/12 (old stock) on 10/4/51

4.52×10^8

10/5/51 11 AM

t = 2.87 hr

with Bruchers mmm H_2O 455 / 450 $\}10^6$

350	300	290	280	270	260	250	240	230
.330	.450	0.585	0.750	0.885	0.980	1.00	1.15	2.00

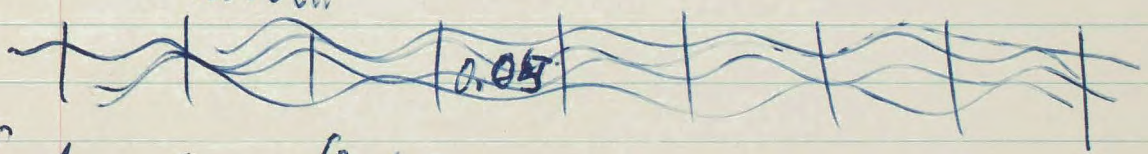
supernatant vs med

350	300	290	280	270	260	250	240	230
0.054	0.059	0.060	0.083	0.142	0.189	0.208	0.300	0.460
vs H_2O .065		0.167	0.232	.290	.312	335	0.401	

Mediums vs H_2O

0.015	0.050	0.097	0.131	0.134	0.124	0.122	0.175

F vs H2O at 280 is 0.05



Saturday Oct 6

3 pm Broth supernatant vs H_2O

at 350 0.443

at 280 1.03

			plates mat pour mat
Assay	97 / 107		9.7×10^8
+T1	84 / 107		
+T1	410 / 2×10^6		8.3×10^8

Friday

293 (Avg Lbd)

2.3 mg/l arginine + 2 mg/l tryp + F

Inoc 084/1t (old) on 10/4/51

10/5/51

τ = 2.79hr

mth Bacteria vs. H$_2$O $\frac{241}{223}$ $\}10^6 = 2.32 \times 10^8$ (11AM)

350	300	290	280	270	260	250	240	230
0.185	0.265	.350	.454	.540	.600	.633	.790	1.58

Supernatant vs medium

350	300	290	280	270	260	250	240	230
0.047	0.059	0.060	0.090	0.135	0.173	0.210	0.315	0.530

(of 1 gm/l)

Added 4 cc hypophosphate to 1.3 l. bacteria making up conc. to about 3 mg/l in Shreve tank at 225 pm

(T = 3.8 hrs) Suspect actually $5\frac{55}{}$ (10/24 a.m.)

Recalculation of specimen

Before .465 / .70 / .395

After 333 / 76 / .257

Typ 40 / .217

Ratio 0.55

At $6\frac{55}{}$, drained contents

Collected $\frac{18\text{ ml}}{3.5} = 5.15\text{ ml/hr} = \frac{18.6}{5.15} = 3.63\text{ hr}$

Supernatant + bacteria vs H_2O

350	300	290	280
0.290			0.890

Supernatant alone vs H_2O

350	300	290	280	270	260	250	240	230
0.028	0.086	0.218	0.333	0.359	0.339	0.303	0.372	

$e^{-\frac{3.5}{3.8}} = e^{-0.92} = 0.40$ $\quad e^{-\frac{3.5}{3.8}}$

~~If there were no~~

0.09 is about typ.

0.333 − 90 = 0.243

0.333 − 76 = 0.243 − 40 = ~~0.183~~ = 203

for calc. w/ 2 mg/l of inj.

In camp. before addition hr.

243 − 56 − 50 = 137

365

$\frac{137}{365} = 0.375$

Friday sudden rise of hypostaphase for precursor

294 500 f/l Hypostaphase in F med. —

τ = 3 hrs B/l to test

Bacteria x supernatant vs. H$_2$O

350	300	290	280	270	260	250	240	230
0.137			0.503					

supernatant vs H$_2$O

	350	300	280	260	270	260	250	240
	023	077	189	292	315	304	277	345
F vs H$_2$O				50	54	72	100	164
supr vs F				.242	261	232	177	

τ = 2.85 hr $242/2.85 hr$ =

Changed from 4 to 3 pms at 5^{30} pm

10/6/51 τ overnight = 3.8 hr

Saturday

at 2^{10} pm

with bacteria vs. H$_2$O

350	300	290	280	270	260	250	240	230
0.138			0.672					

supernatant vs. H$_2$O

0.025	0.091	0.290	0.465	0.490	0.436	0.380	0.448	

0.390/3.8 ≈ 0.100/hrs

$NH_4Cl = \frac{14}{54} N = 0.26\ N$

~~Thursday Oct 8/51~~

207

$0.819 \Rightarrow \frac{0.819}{0.62} \Rightarrow (1.32) \times 20 = 26.4$ mg/l precursor

26.4 mg/l precursor contains $\frac{14}{203} \times 26.4 = (0.069)(26.4) = 1.82$ mg N

1.82 mg N $\approx$ 1.82 $\frac{1}{.26}$ = 7 mg/l NH_4Cl

10/11/51 τ overnight = 11.6 hr

Bact + Super vs H_2O at 350 0.138
at 280 1.04

Collected 11 am to 3 pm 7.8 ml = 1.9 ml/hr = 11.6 hr τ

Super vs H_2O

350	300	290	280	270	260	250	240
035	147	.550	.895	.908	.770	615	.660

Bact resusp in H_2O vs H_2O

350	280
.103	.220
.138	1.115

0.895
.052
0.843 / 11.6 = 0.073/hr

1 mg NH_4Cl ~~or Cl2~~ = 0.26 mg N = $\frac{203}{14}(.26)$ mg Pre = 3.77 mg Prec/l

= $\frac{3.77}{20}(0.62)$ = 0.117 in optical density

Doubled flow rate at 4 20 pm

10/12/51 τ overnight = 2.8 cc/hr = 5.8 hr

Collected 10^{20} – 12 noon

Bact + Sup vs H_2O 350 0.155
280 0.825

Super vs H_2O

350	300	290	280	270	260	250	240
037	125	390	.630	653	580	480	547

0.630
.050
.580 / 5.8 = 0.100/hr

10/13/51 τ = 6.7 hr Bact + S 350 .144
280 .822

350	300	290	280	270	260	250	240
025	106	370	620	635	552	458	520

10/14/51 τ = 7.7 hr Bact + S vs H_2O 350 0.147
280 0.901

Continued 3 pages ahead

Tuesday Oct 9/51

|296| Inoc pt Ehrl 5ml/1l 25 mgm/l NH_4Cl

τ = ~~about~~ 4.2 hrs

At 345 pm Bact + Supernatant vs H_2O

Beckmann at 350 0.150

at 280 0.720

$$\frac{420 - 50}{370/4.2} = 0.089$$

Supernatants vs. H_2O

350	340	290	280	270	260	252	240	230
0.029	0.105	0.325	0.530	0.555	0.497	0.441	0.540	

$$\frac{530 - 60 - 50}{420/4.2} = 0.1/\text{hr.}$$

Flame tube dismissed at 3:54 pm

10/10/51 τ overnight = 5.6 hrs

Collected 12 noon to 3 pm 5.7 ml = $\frac{5.7}{3}$ = 1.9 ml/hr = 11.6 hr

@

Bact + Supernatant vs H_2O at 350 = 0.130

at 280 = 1.02

Supernatant vs H_2O

350	300	290	280	270	260	250	240
033	.144	.563	0.935	.945	.800	623	.640

$$\frac{.935 - .66 - .50}{.819/11.6} = 0.070/\text{hr}$$

Bact resusp in H_2O vs H_2O

350	300	290	280	270	260	250	240
.105	.144	178	.215	244	265	273	288

10/11/51 τ overnight = 10.7 hr at 350 0.157

Bact + Super vs H_2O at 280 1.50

Collected 11am to 3pm 9.1ml = 2.27 ml/hr ~ τ = 10.6 hr

Supernatant vs H_2O

350	300	290	280	270	260	250	240
029	162	.810	1.33	1.32	1.07	0.757	0.702

Resuspended bacteria in H_2O vs H_2O

350	~~350~~	280
.120		0.260

1.33 − .05 = 1.28 / 10.7 = 0.120/hr

10/12/51 τ overnight ~~4.5cc/hr~~ = ~~4.8hr~~ 5.35 hr

Collected 10^{20} — 0.140

Bact + Super vs H_2O 350 280 0.980

Super vs H_2O

350	300	290	280	270	260	250	240
034	120	490	810	820	680	500	497

0.810 − 0.050 = .760 / 5.35 = 0.14/hr

10/13 τ = 8.65 hr Bact + Super 350 280 0.141 1.36

350	300	290	280	270	260	250	240
025	140	725	1.21	1.20	0.960	.670	630

10/14 τ = 10 hr Bact + S vs H_2O 350 280 0.149 1.49

10/15 τ = 11.3 hr Bact + S vs H_2O 350 280 0.143 1.55

350	300	290	280	270	260	250	240
028	167	862	1.428	1.408	1.12	0.760	0.700

10/16 τ = 10.8 hr Bact + S vs H_2O 350 0.142 280 1.500

Tuesday Oct 9/51

297 tube E296 last 35 mgm/l NH4Cl

at 345 pm. $\hat{C} = 3.8$ hr

Bact + Supernat. in H2O
at 350 = 0.141
at 280 = 0.820

820
−50
770
300
470/3.8 = 0.123

supernatant vs. H2O

350	300	290	280	270	260	250	240
0.025	0.103	0.377	0.630	0.640	0.530	0.445	0.485

630
−50
−50
530/3.8
= 0.14

Flowrate changed at 354 pm

10/10/51 t overnight = 11.6 hr
Collected 12 noon to 3 pm 5.9 ml = 5.9/5 = 1.97 ml/hr = 12.2 hr

at 350 0.156
Super + Bact at 280 = 1.50

Super vs H2O

350	300	290	280	270	260	250	240
035	175	.840	1.37	1.36	1.12	0.800	0.740

Bact resusp in H2O vs H2O

350	280
.142	.293

1.37
−.07
−.05
1.25/12 = 0.105/hr

296 continued

10/15/51 τ = 7.9 hr Baef + S vs H_2O 350 0.140 / 280 0.885

350	300	290	280	270	260	250	240
025	100	408	671	685	586	470	521

10/16/51 τ = 8.0 hr Baef + S vs H_2O 350 0.144 / 280 0.890

295 Tuesday Oct 9/51

Strain 212 in 7

at 4^{45} pm ~~stuff growing~~ ; at $\tau = 5$ hrs

Bacteria + supernatant vs H$_2$O

at 350 0.301 this sample may be 20% diluted
at 280 0.860 because we forgot to add

Supernatant vs. H$_2$O

350	300	290	280	270	260	250	240	230
0.025	0.049	0.140	0.200	0.245	0.269	0.290	0.365	
—			052	057	076	100	180	
			.150	.188	.193	.190	.185	

not pursuing

10/10/51 Flow rate overnight $\tau = 6.2$ hr

Apparently O$_2$ limited

Supernatant (undiluted) vs H$_2$O

350	300	290	280	270	260	250	240
.054 0.540	~~.770~~ .710	~~.770~~ .640	1.15	1.60	2.15	2.40	3.00

10/8/51 Substrate expts

	24 hr	48 hr
B/tC (all + 10 mg/l typ)		
10 gm/l lactate	+	+
5 gm/l "	+	+
2 gm/l "	—	—
30 gm/l succinate	—	—
10 " "	—	(+)
3 " "	—	—
10 gm/l glucose	—	+
3 " "	—	+
1 " "	—	+
0,3 " "	—	(+)

B

30 gm/l succinate	—	—
10 " "	—	+
3 " "	—	—
10 gm/l glucose	—	+
3 " "	—	+
1 " "	—	+
0,3 " "	—	—

at 4:15 pm added 50 cc of gm/l anthranilic acid to bacterial culture, to about 30 mgm/l

---

Sunday Oct 21/51

at 1 pm Bact + Supern vs H2O

$\bar{t}$ = 3.4 hr

350 = 0.211
280 = 0.865
400 = 0.112
320 = 0.690

bacteria low by 25%

Supernatant vs H2O

400	350	300	290	280	270	260	250	240
0.015	0.097	0.633	0.650	0.645	0.580	0.701	1.40	

340	330	320	310
0.220	0.395	0.570	0.667

20 mg anthranilic in ↑ vs H2O

400	350	300	290	280	270	260	250	240
0.009	0.071	0.435	0.320	0.215	0.160	0.295	0.880	

340	330	320	310
0.158	0.282	0.410	0.472

Conclusion anthranilic has no effect:

280	270	260
0.645	580	701
− 247	143	330
0.398	340	350
	437	371

280 = 0.398
270 = ~~0.340~~ = 437
260 = ~~0.XXX~~ 371

296a

Fahrenheit
Oct 20/51

$\tau = 3.38$ hr

Bact + Supernat @ 350 = 0.140, @ 280 = 0.720 (vs H_2O

Supernatant vs H_2O

350	300	290	280	270	260	250	240
0.034	0.096	0.315	0.515	0.533	0.470	0.377	

Added anthranilic acid to 1 mg/l at 12[15] pm

296a at 3[15] pm

Bact + Supernatant vs H_2O

at 350 = 0.143

at 280 = 0.760

Supernatant vs H_2O

350	340	290	280	270	260	250	240
0.030	0.097	0.330	0.525	0.535	0.458	0.362	

no change

20 mgm/l anthranilic in H_2O vs H_2O

400	350	340	290	280	270	260	250	240
0.012	0.140	0.285	0.190	0.123	0.101	0.195	0.660	0.920

340	330	320	310
0.225	0.307	0.357	0.352

Sunday Oct 21/57

at 1 pm

T = 3.36 hrs

Bact + Supernatant vs H2O
at 350 = 0.125
280 = 0.665

Bact + supernatant vs. H2O

400	350	300
0.099	0.125	

10/20/51 297a (more N than 296a)
Saturday

$\tau = 3.97$ hr

Bact + Supernatant vs H_2O @ 350 0.123
@ 280 = 0.730

350	300	290	280	270	260	250	240
0.029	0.090	0.335	0.555	0.570	0.488	0.377	

Added indole to 1 mg/l at 12[15] pm

29
-15
14 x 2 = 28

0.555
-52
-28
0.475/4 ≈ 0.12

297a at 3[15] pm

Bact + Supernatant vs Hr
350 = 0.165
280 = 0.803

~~Supernatant vs H2O~~

Supernatant vs H2O

350	300	290	280	270	260	250	240
0.030	0.100	0.360	0.600	0.612	0.525	0.440	0.440

No change

298

at 340 [2 hrs after raising Pyro]
(call for 15 min around 4)

Brett + Supern vs. H$_2$O
350 = 0.204
280 = 0.733

Supernatant vs H$_2$O

350	300	290	280	270	260	250	240	230
0.034	0.105	0.272	0.420	0.440	0.401	0.350		

-100
38
212

420 − 90 = .330

733 − 50 − 380 = 430 → 0.300

full 0.785 or full 0.74

~~n_i / n_{i-1}~~

i in hours $\alpha = \ln n(i) - \ln n(i-1) + \frac{+1}{t}$

or for ½ half hour points

$n = e^{\alpha t}$ $\quad n_1 = e^{\alpha t}$ $\quad n_2 = e^{\alpha(t+1/2)}$ $\quad \frac{n_1}{n_2} = e^{1/2\alpha}$

$\alpha = \frac{1}{t} = 2 \ln \frac{n_1}{n_2}$

$\alpha = 2 \ln \frac{n_1}{n_2} + \frac{1}{t}$

Tuesday Oct 23/51

298 3/1t 500 f/l Tryptophane

Purpose: Sudden increase of tryptophane by about 3 mgm/l in stock tank. How does this l effect bacterial density and „precursor",—

added 1½ cc M/1 gm/l tryptophane added about 600 cc at 140 pm

~~t = 3.75 hours~~ t = 3.5 hrs

at 140 pm Bertrand Bact + Super vs H2O { 350 : 0.149 ; 280 : 0.723

723 − 268 − 318 = 405

Supernatant vs. H2O

350	300	290	280	270	260	250	240	230
0.031	0.097	0.318	0.500	0.519	0.452	0.368		

[280] − 52 = 450 ; [250] − 100 = 268 ; 1240 ; multiplier² = 1.68

500 − 50 − 30 = 420/3.75

266 − 30 = 230 = 0.112

at 240 pm [all trans 235 to 245] { Bact + Super vs H2O at 350 : 0.169 ; at 280 : 0.690

Supernatant vs H2O

350	300	290	280	270	260	250	240	230
035	.118	.324	.510	.529	.462	.385		

90 / 420 − 24 ; 247 − 12

690 − 50 − 308 = 358 ; 332

multiplier² = 1.62 = (510 − 50)/(385 − 100) = 460/285

350/423 = 0.83

Weden Oct 24/57

at 11 AM

350 = 0.483

280 = 2.14

Supernatant vs H2O

350	340	290	280	270	260	250	240
0.037	0.203	0.980	[illegible] 1.58	1.56	1.28	0.90	0.84

It pours out!

2.3 mgm

0.5 "

$$\frac{[280]-52}{[250]-102} = \frac{1.530}{800}$$

= 1.91

2.81 mgm/l tryptophane

about 1 mg/l tryptophane must be left over unless eaten up by tryptophanase. So two alternatives 2.7 / mgm/l trypt does not

Plans for repeat expt

1. Use 0 Alanine
2. Add typ to growth tube to come in reservoir
3. Use 250 tryptophan
4. Will use 75cc growth tube again

298

at 4⁴⁰ pm Bact + supern vs H₂O

350 : 0.259

780 : 0.822

Supernatant vs H₂O

350	300	290	280	270	260	250
0.035	0.099	0.224	0.340	0.371	0.355	0.330

$\frac{290}{230} = \boxed{1.26}$ $\frac{340}{-90} = .250$

822 − 488 − 50 } 538

244

0.284

antilyses 1.26

aeration increased at 5 pm ///

at 5⁴⁰ pm Bact + Supernat vs H_2O

350 = 0.327

280 = 0.980

antilyses 1.28

980 − 50 − 624 } 674

306

Supernatant vs. H₂O

350	340	290	280	270	260	250
0.045	0.115	0.247	0.378	0.414	0.392	0.364

$\frac{378 - 50}{364 - 100} = \boxed{1.28}$

378 − 110 = 268

[$\bar{c}$ = 3.5 hrs measured at 6 30 pm] //

at 6⁴⁰ pm $\frac{434-50}{355-100} = \frac{384}{255} = 1.5$ Bacteria + Supern. vs H₂O

350 : 0.381

280 : 1.190

antilyses 1.5

Supernatant vs H₂O

350	340	290	280	270	260	250	240
0.038	0.105	0.275	0.434	0.460	0.420	0.355	0.403

At $2\frac{40}{\mu m}$ $c_2 = 0.250$ $c_1 = .370$

$a_1 = 0.066$

At $1\frac{40}{\mu m}$ $c_2 = 0.268$ $c_1 = 0.450$

$a_1 = 0.030$

350	325	300	295	290	285	280	275	270	265
.010	.018	.087	.221	.562	.784	.960	.960	.938	.845

260	255	250	245	240	235	230	231
.722	.585	.468	.398	.351	.476	1.05	.840

232	225
.698	72.0

Spectrum of factory supernatant
10/30/51

$$\frac{A_2}{B_1} = \frac{\rho_{250} + a_{250}}{\rho_{250} + a_{250}} = \frac{\rho_1 + a_1}{\rho_2 + a_2} = 1 + n$$

$$\frac{\rho_1}{\rho_2} = 1.8 \qquad \frac{a_1}{a_2} = 0.8$$

$$A_1 = \rho_1 + a_1 = \rho_1 + a_1 \rightarrow \rho_1 = C_1 - a_1$$
$$B_2 = \rho_2 + a_2 = \frac{\rho_1}{1.8} + \frac{a_1}{0.8}$$

$$C_2 = \frac{C_1 - a_1}{1.8} + \frac{a_1}{0.8}$$

$$C_2 - \frac{C_1}{1.8} = a_1\left(\frac{1}{0.8} - \frac{1}{1.8}\right) = 0.695\, a_1$$

$$a_1 = 1.44\left(C_2 - \frac{C_1}{1.8}\right) = 1.44\left(C_2 - 0.55 C_1\right)$$

at 4^{40}_{pm} $C_2 = 0.230$, $C_1 = 0.290$

$a_1 = \underline{0.088}$

at 5^{40}_{pm} $C_2 = 0.264$ $C_1 = 0.338$

$a_1 = \underline{0.112}$

at 6^{40}_{pm} $C_2 = 0.255$ $C_1 = 0.384$

$a_1 = \underline{\text{0.167}}$ 0.063

at 2^{40}_{pm} $C_2 = 0.285$ $C_1 = 0.460$

$a_1 = 0.045$

10/30/51 Flask Pouring Out Expt.

Idea is to simulate chemostat using large flask, and simple feeder – no overflow.

Used 500 cc, 5000 #/l typ in flask, in feeder used 6 mg/l typ; Inoc with ~10^2 B/T/cc at 7pm on 10/29. Flask started at 10 am on 10/30/51. Visibly turbed by 3pm on 10/30

At 10 am on 10/31 took 10cc sample

Bact + Super vs H_2O 350 0.197
280 1.70

Supernat vs H_2O

Drop rate was 1/90"

350	300	290	280	270	260	250	240
034	.179	0.902	1.47	1.45	1.19	0.846	0.742

$\frac{197}{145} = 1.36$ times usual bacterial density

$1.47 - 0.05 - .04 = 1.38$

$\frac{1.38}{1.36} = 1.02$ which corresponds to ~ $\frac{1.02}{0.14} = 7.3$ hours

At 3pm Bact + Super 350 0.240
280 2.14

$\frac{dc}{dt} = kN = kN_0 e^{\frac{t}{\tau}} \Rightarrow c - c_0 = \frac{k\tau}{e} N_0 \left[e^{\frac{t}{\tau}} - 1 \right]$

~~$2.14 - 1.70 = 0.44 = \frac{0.14\tau}{e} N_0 e^{\frac{t}{\tau}} = \frac{\tau 0.14}{e} (1.37) e^{\frac{5}{\tau}}$~~

Mistake!
4.6 mg/l Arg

Also dl Trypt was added

305 Mutual feeding B/lt + D84/6 Vgt=25.6

10/27 Inoc Chemostat containing [0.50 mg/l Trypt, 0.46 mg/l arg.]

with B/lt at 1 pm

10/28 2 pm assay (B/lt) $\left.\begin{matrix}145\\140\end{matrix}\right\}10^6 = 1.43\times10^8$

Sunday after assaying, inoc D84/6 ~ 5×10^7/ml

$\tau = 3.16$ hours.

10/29 12 15 a.m.

Mon +T1 $\left.\begin{matrix}48\\70\end{matrix}\right\}2\cdot10^6 = 1.18\times10^8$

+T6 $\left.\begin{matrix}306\\358\end{matrix}\right\}10^6 = 3.32\times10^8$

12 noon

+T1 $\left.\begin{matrix}121\\111\end{matrix}\right\}10^6 = 1.16\times10^8$

+T6 $\left.\begin{matrix}253\\220\end{matrix}\right\}2\cdot10^6 = 4.73\times10^8$

10/30 11 30 am

Tues +T1 $1158\}10^5 = 1.16\times10^8$

+T6 $\left.\begin{matrix}225\\255\end{matrix}\right\}2\cdot10^6 = 4.8\times10^8$

305a Stopped. Set up new Chemostat to contain the originally desired concentrations of amino acids.

10/31 Inoc with overnight B/lt (~ 10^8 in growth t.)

Wed. at 11 am.

11/1 Thursday 11 30 am Assay +T1 $\left.\begin{matrix}212\\215\\215\end{matrix}\right\}10^6 = 2.17\times10^8$

D84/6 reverted

2 mg/l hypophosphate; 30 mg/l NH_4Cl

308 $\tau = 4.9$ hrs no Ψ change

4 pm

bacteria + supernatant

350	300	290	280	270	260	250	240	230
0.090	0.100	0.135	0.195	0.272	0.317	0.348	0.530	

supernatant vs H_2O

350	0.254

280	0.540

195 − 50 = 145 → 150 → 200

540 − 50 = 490 → 480 → 000

does not pass QC

Conclusion: Nitrogen limited chemostat with 2 mg/l trypt does not pass QC.

Wed Friday; Nov 2/51

307 $\bar{c} = 4.3$ hrs

30 mg/l NH_4Cl, 2 mg/l trypt, 10 mg/l L alanine

4 pm Bacteria + supernatant vs H_2O

350 280

0.245 0.620

Supernatant vs H_2O

350	290	280	270	260	250	240	230
0.040	0.207	0.298	0.325	0.331	0.340	0.460	

300
0.119

298 620
52 52

246 568
−50 460

196 to comp 108

does not pour out

Measure out more:

Saturday Nov 3

Bacteria + Suspens. vs H_2O

3 pm 350 = 0.240 280 = .619

$\bar{c}$ =

Supernatant vs H_2O

350	300	290	280	270	260	250	240	230
0.045	0.115	0.163	0.218	0.254	0.291	0.318	0.444	

−60 −102
−52 −80

.106 182

.136

−21

.115

does not pour out

O.K.

11/13/51 Strain 159-11, 5000 typ, $\tau = 3.2$ hr

Sample #2 vs H_2O

350	300	290	280	270	260	250	240	230
0.039	0.119	0.376	0.610	0.630	0.658	0.478	0.596	1.61

Sample #1 vs H_2O

350	300	290	280	270	260	250	240	230
0.035	0.100	0.263	0.411	0.434	0.410	0.386	0.531	1.45

why higher than in exp. started on 11/8/51

~~[illegible] 2 mg/l tryp [illegible] 10 mg/l [illegible]~~

11/7/51 Low NH_3 factory. Supernat dil $\frac{1}{5}$ in H_2O vs H_2O

350	300	290	280	270	260	250	240	230
026	.110	.632	1.07	1.065	.840	.563	.475	1.19

$\frac{1.07}{0.62} \times 20 \cong 170$ mg/l of X

11/8/51 Strain 159-11 grown in chem on 5 γ tryp (NO 317)
Thursd. τ = ~~[illegible]~~ 3.25 hrs
12 noon Bacteria + supernatant vs H_2O
350 = 0.167   280 = 0.750

Supernatant vs H_2O

350	300	290	280	270	260	250	240	230
0.027	0.089	0.294	0.460	0.465	0.408	0.345	0.395	1.140

it pours out!

89
−35
54

390

0.245 395 −164 231 30 .200

−607 0.533 −.045 0.488

this is like tryptophane not like indol! —

Collected 42cc 11¹⁵ − 4⁰⁵ 42cc $\frac{42}{4.83}$ = 8.7cc/hr = $\frac{25.6}{8.7}$ = 2.95 hr = τ

4–5 pm collection:
Bact. + supernatant vs H_2O
at 350 = 0.166   at 280 = 0.4775

supernatant vs H_2O

350	300	290	280	270	260	250	240	230
0.029	0.095	0.325	0.510	0.513	0.440	0.362	0.407	1.150

it pours out. —

11/9/51 Flask Conversion Expt.

Use 1 mg/l tyrosine in flask (500cc)

If dropper adds 20cc/hr and we want to add tyrosine at 0.15 mg/l/hr

½ l = 0.075 mg/hr in 20cc = .0375 mg/cc

= 37.5 mg/l in feeder

Will use 100 mg/l Cal, 34 mg/l [illegible] X (⅓ ml / 7 feet)

Thursday 5 pm

11/8/5-1 Bio Assay Expts

	Expt	"X"	Tryp	φAl	Tyr	Bact	Beckman	vs H_2O
X from low MW fraction	1.	17 mg/l	⊕	100 mg/l	0.3 mg/l	MF3II4P1	0.163	very addl
	2	"	"	"	1.0	"	0.276	
	3	"	"	"	3	"	0.410	
	4	"	"	"	Xa	"	0.440	
	5	0.1 mg/l	⊖	100 mg/l	20 mg/l	MF3II4P1	(10⁷)	
	6	0.33	⊖	"	"	"	(2·10⁷)	
	7	1.0	⊖	"	"	"	(3·10⁷)	
	8	3.3	⊖	"	"	"	(5·10⁷)	
	9	⊖	0.1 mg/l	100 mg/l	20 mg/l	MF3II4P1	0.104	very addl.
	10	⊖	0.33	"	"	"	0.204	
	11	⊖	1.0	"	"	"	0.475	0.195 x 3
	12	⊖	3.3	"	"	"	0.645	0.260 x 3
	13	0.1 mg/l	⊖	⊖	⊖	121-35	(—)	(—)
	14	0.33	⊖	⊖	⊖	"	(—)	(—)
	15	1.0	⊖	⊖	⊖	"	(5·10⁶)	(—)
	16	3.3	⊖	⊖	⊖	"	(2·10⁷)	turbid
	17	⊖	0.1	⊖	⊖	121-35	0.089	very addl
	18	⊖	0.33	⊖	⊖	"	0.235	
	19	⊖	1.0	⊖	⊖	"	0.210 x 3 = 0.63	
	20	⊖	3.3	⊖	⊖	"	0.265 x 5 = 1.32	

(320) before adding trypt. :

Supernatant vs F

350	300	290	280	270	260	250	240	230
0.014	0.044	0.171	0.293	0.305	0.270	0.206	.200	

+ off this 0.274 may be precursor

Trypsin plasmin added at 11 AM

Sample No1 coll. 10^{24} to 11^{36} AM [12 min]

[11 30 AM]

Back + Super vs. F

350 = 0.154 280 = 0.604

Supern vs F

350	300	290	280	270	260	250	240	230
0.018	0.063	0.215	0.355	0.364	0.306	0.229	0.223	

0.355
−0.075
280
−8
272

Sample No2

Sample No 2 at 12 noon

Back + Supernatant vs F

350 = 0.158 280 = 591

Supernatant vs F

350	300	290	280	270	260	250	240	230
0.016	0.057	0.201	0.325	.336	.294	0.225	0.220	0.460

Expt 320 B/16 (500 γ/l Tryptophane
20 γ/l Nbenthiamidole
10 mg/l Phenylalanine)

11/15/51 τ = 5.9 hr
2^10 – 2^30 pm Bact+super vs H2O 350 0.114
280 0.510

~~11/15/51~~ Flow rate increased at 3 pm
Measured 2^54 – 9^10 pm = 163 cc = 3.05 hr
9^30 pm Bact+super vs H2O 350 = 0.144
280 = 0.570

Nov 16/51
Friday τ = 2.97 hr at 10 AM

840 cc estimated in storage tank
V = 79 cc
add to storage tank 2.11 cc of 1 gm/liter C-Tryptophane sol.
to bring conc to 3 mγ/l
to V we add N-Benthiamidole 1 cc of 1:5 dilution
of 1 gm/l tryptophane to bring conc to 2.5 mγ/l.–
T vs Thio

0.9 NBA	350	280	240
	0.013	0.059	0.165

τ = 2.97 hr

At 1030 AM before adding tryptophane

350	280
0.135	0.526

Bact + Super vs T

135
012
1254
2146

526
270
256

526
2
2566/3

320

Bacteria at 215 pm
undiluted ; Bact + Super vs F
at 350 : 0.265
diluted 1+02 in F vs F
at 350 : 0.145 x 2 = 0.290

Sample No 7 at 230 pm
Bact + Sup vs F
at 350 : 0.25 at 280 : 0.783
or 0.158 x2 = 0.316

Super vs F

350	300	290	280	270	260	250	240	230
0.045	0.076	.158	.247	.295	0.305	0.284	0.335	0.560

At 435 pm bact + sil 1/5 in F vs F = 0.133

Saturday Nov 17
collected 1130 AM (marked out)
at

Bact + Sup diluted 1:3 in F vs F

350	300	290	280	270	260	250	240	230
0.122								

x3

Bact + Super dil in F vs F
at 350 : 0.122 x 3 ; 0.270 x 2

(320)

Sample No 3 at 12 30 pm iced
lost

Sample No 4 at 1 pm iced

Bact + Supern vs F

at 350 = 0.195 at 280 = 0.690

Supernatant vs F

350	340	290	280	270	260	250	240	230
0.023	0.061	0.185	0.298	0.315	0.279	0.220	0.230	0.450

Sample No 5 at 130 pm (not iced)

Bact + Sup vs F

at 350 = 0.215 at 280 = 0.673

Supernatant vs F

350	340	290	280	270	260	250	240
0.023	0.050	0.147	0.235	0.260	0.246	0.211	0.235

Sample No 6 at 2 pm (not iced)

Bact + Supern vs F

at 350 = .245 at 280 = 0.700

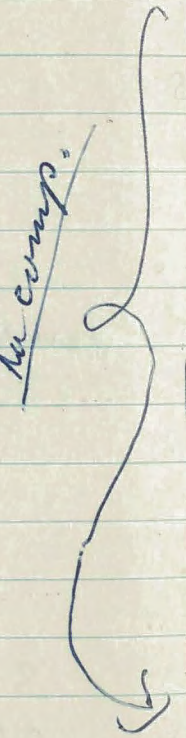
Supernatant vs F

350	340	290	280	270	260	250	240	230
0.029	.055	.143	.227	0.260	0.256	0.230	.270	.497

$T_2 r$ in F vs F

400	390	380	370	360	350	340	330	320	310	300
0.029	0.034	0.037	0.045	0.083	0.119	0.149	0.170	.187	.196	0.220

300	290	280	270	260	250	240	230
0.220	0.340	0.600	0.940	1.20	1.42	2.10	2.60



(320) Juice of Chemostat 320 vs F

350	300	290	280	270	260	250	240	230
0.000	0.005	0.045	0.065	0.063	0.053	0.041	0.037	0.088

↳ this is mainly tryptophane

this is O.K.

Bottle continued:

Supernatant vs F

400	350	300	290	280	270	260	250	240	230
0.011	0.094	0.046	0.072	0.136	0.233	0.280	0.301	0.453	0.640

does not pour out.

↳ mix! τ ≃ 3 hrs at levels of 0.122 x 3

↳ compare this with same lysate!

Nov 18/51 3:30 pm (Sunday) back to Friday overnight with bacteria; centrifuged at

content of growth tube standing

Supernatant vs F

400	390	380	370	360	350	340	330	320	310	300
0.019	0.020	0.031	0.060	0.092	0.109	0.110	0.099	0.084	0.074	0.073

300	290	280	270	260	250	240	230
0.073	0.101	0.170	0.275	0.320	0.325	0.475	0.640

to compare with Tr lysate

Bacteria shaking

to compare with Tr in F.

(lysate; thickened)

supernatant vs. own juice containing anthranilate

350	340	330	320	310	300
0.051	0.051	0.046	0.045	0.044	0.054

300	290	280	270	260	250	240	230
0.054	0.075 .080	0.127	0.180	0.201	0.174	0.184	0.285

↓ this is high compared with Tokes

supernatant vs own juice containing anthranilate and anthranilate is still here

no "Indol" produced.

(312 Sunday Nov 18 added at 6 pm
16 cc 1 gm/l Antr. acted as
soap on juice in Taxes
Chemostat (312) to see if
"initial" is present and, — B (+ lungs)
Monday Nov 19/57 N limited
11 AM Taxes $\bar{\tau}$ = 6.4 hrs mm/super vs. auth. juice

350	340	330	320	310	300	290	280	270
0.055	0.060	0.075	0.110	0.159	0.179	0.197	0.253	0.335

11 40 AM pushed out pure Chemostat
sample also pushed out juice.
$\bar{\tau}$ = 6.7 hrs
juice (with authramycillic) vs juice of Tax

350	340	330	320	310	300
0.040	0.093	0.171	0.250	0.294	0.260

12

350	340	330	320	310	300	290	280	270	260	250	240	230
0.040	0.260	0.175	0.093	0.043	0.120	0.498						

Sample with Tax vs auth juice containing authramycillic

350	340	330	320	310	300	290	280	270	260	250	240	230
						0.365	0.485	0.596	0.650	0.600	0.590	

350	340	330	320	310	300
0.220	0.230	0.230	0.232	0.244	0.274

Tax used to run 0.216 - 12 = 0.204 O.K.

3/21/5

5^30 pm

all 3 pm to 5 30 pm.

Sample with Bact vs own juice

350	300	290	280	270	260	250	240	230
0.256			1.305					

unchanged

supernatant vs. own juice

350	300	290	280	270	260	250	240	230
0.032	0.088	0.523	0.860	0.860	0.690	0.479	0.410	0.915

drained from 3 pms to 4 pms at 5^30 and added tryptophane to increase trypt. in bottle by 100 γ/l [8.4 cc of 0.3 cc (of 1 g/l) into 10 cc] at 6 pm

9^30 pm τ = 3.4 hr

9^30 – 11 pm Bact + Supes vs own juice

Supernat vs own juice

350	300	290	280	270	260	250	240	230
043	1.088	1.387	0.650	0.670	0.550	0.403	0.350	0.740

350 = 0.273

280 = 1.085

at 12^30 a m added tryptophane to bottle to increase concentration by 1 mg/l.

Wednesday

10^45 AM τ = 3.75 hrs

Sample with Bact vs own juice

(not cont. added to not 1.1 mg/l Tryptophane)

Mo Nov 19/57

(321) 2 mg P 1 mg/l Tryp

$\bar{t}$ = 4.8 hrs

Is it up?

collected 2 pm to 4 pm

Bacteria + supernatant vs own juice

350	300	290	280	270	260	250	240	230
0.132	0.183	0.248	0.343	0.410	0.448	0.440	0.488	

Supernatant vs. own juice

0.012	0.027	0.066	0.120	0.140	0.139	0.135	0.188	0.408

True Nov 20

$\bar{t}$ = 4.7 hrs 11 AM

call from 9:45 to 11:30

Bacteria + Super vs. own juice

350	300	290	280	270	260	250	240	230
0.255	0.371	0.845	1.255	1.335	1.235	1.037	0.980	

Supern. vs. own juice

0.023	0.078	0.479	0.839	0.840	0.670	0.458	0.386	0.440

ok pours out! but a little low.

$$\frac{0.39 - 46}{\sim 800/1.8 \times 4.7} = 0.095$$

Saturday 11/24/51

$\tau = 7.2$ hr

Bact + Supe vs own juice — 350 = 0.462, 280 = 2.10

Bact 1/5 in own juice vs own juice — 350 = 0.112, 280 = 0.685

Supe vs own juice

350	300	290	280	270	260	250	240	230
064	1.170	1.120	1.815	1.839	1.480	0.980	0.800	1.680

at 2:30 pm switched from 2 pins to 4 pins

Sunday 11/25/51

$\tau = 3.67$ hr

~~Bact~~ Collected 2:45 – 3:45 pm — 350 = 0.450, 280 = 1.325

Bact + supernatant vs own juice

at 1/3 dil in own juice — 350 = 0.175, 280 = 0.500

Supernat vs own juice

~~350~~	~~300~~	290	280	270	260	250	240	230
0.075	0.069	0.305	0.547	0.610	0.540	0.425	0.468	0.820

Supernat vs own juice

Wedn Nov 28/51

call on Monday $\tau = 13.4$ hrs

~~Bacteria~~ 350 = 0.508 280 = 2.20 vs. own juice

Sample with bacteria dil 1:5 vs own juice

at 350 = 0.120 × 5 at 280 = 0.800 × 5

call on Tuesday $\tau = 8.25$ hrs vs own juice

Sample with bacteria dil 1:4 at 350 = 0.134 × 4, 280 = 0.935 × 4

Supernat vs own juice	350	300	290	280	270	260	250	240	230	
$\tau = 13.4$	0.077	0.252	1.60	2.17	2.15	1.98	1.40	1.14	1.90	2.17/13.4 × 4
$\tau = 8.25$	0.055	0.220	1.48	2.09	2.09	1.87	1.28	1.01	1.86	2.1/8.25 × 4

321 4

Sample with Bact. vs "own juice"
[not containing additional 1.1 mg/l Trypt.

350	300	290	280	270	260	250	240	230
0.340			0.690					

con 0.370

does not pour out

Supernatant vs "own juice"
[not containing added 1.1 mg/l Tryptophane

350	300	290	280	270	260	250	240	230
0.075	0.045	0.069	0.128	0.200	0.229	0.221	0.315	0.460

−20

↑ rise

does not pour out

400	390	380	370	360	350	340	330	320	310	300
0.014	0.017	0.026	0.043	0.065	0.078	0.080	0.070	0.061	0.048	↑

@ 0.43

peak

At 330 pm added 1.15cc of 3mg/cc P to 2300 ml c. flask

Thurs 11/23/51 τ = 3.63 hrs

7 pm - 1 3/4 pm Sample + bact vs own juice

350 = 0.450
280 = 1.19
does not pour out

Switched from 4 to 5 pms at 7 30

Friday 11/24/51 τ = 2.90 hrs Collected 12 30 — 1 45

Bact + Supernat vs P

350 = 0.432
280 = 1.03

Supernat vs own juice

350	300	290	280	270	260	250	240	230
072	040	.112	.214	.290	.287	.249	.320	0.500

At 2 pm switched to 7 pms (from 5)

At 2 30 switched to 2 pms should be 7.2 hrs

4=0
=0.063

continued P.T.O.

323 224

Weeks Nov 21

11 25 AM (call from 9 AM + 11:30 AM)

323 $\hat{c}$ = 2.5 (overnight)

324 $\hat{c}$ = 2.03 (overnight)

323 sample with Bachera vs F

Becton 350 = 0.232 | 280 = 0.755

Supernatant vs F

350	300	290	280	270	260	250	240	23
0.005	0.045	0.192	0.327	0.342	0.277	0.187	0.157	

it pauvant changed from 5 pms to 6 at 1145

324 sample with Bachera vs F

at 350 = 0.212 at 280 = 0.637

Supernatant vs F

350	300	290	280	270	260	250	240	230
0.008	0.026	0.099	0.173	0.195	0.180	0.140	0.126	0.240

changed from 12 pms to 3 pms at 12 15 pm

rate of Bact. from 0.212 to 0.244

324 Sample pushed out at 1 15 pm

Sample with Bachera vs F

0.244		0.779					

Supernatant vs F

0.009	0.032	0.146	0.252	0.267	0.230	0.171	0.177	0.414

gives 0.060/hr

Sample collected from 12 15 pm to 1 15 pm vs F

dil 1:2 vs F

at 350 = 0.125 X 2 at 280 = .395 X 2

to comp with .212

Tue Nov 20

(323) $\bar{c} = 1.95$ kn 11AM 1 mg/l Tryptophane
(324) $\bar{c} = 2.15$ kn

at 4 pm (323)

→ Sample with Bacteria vs H_2O

350	300	290	280	270	260	250	240	230
0.135	0.217	0.300	0.388	0.457	0.507	0.530	0.617	

−65

at 4 pm (324)

→ Sample with Bact vs H_2O

0.188	0.290	0.400	0.521	0.617	0.680	0.690	0.780

→ Supernatant of [323] vs. (F)

350	300	290	280	270	260	250	240	230
0.009	0.021	0.050	0.081	0.100	0.105	0.115	0.145	0.230

−15

does not pass out ×

→ Supernatant of [324] vs (F)

0.015	0.026	0.063	0.097	0.122	0.135	0.147	0.183	0.276

does not pass out ×

In one hour [of which may be 20 min more spent in exhausting the tryptophane present] abs at 280 rose from 0.173 to 0.252 i.e. by 0.080 in place of
$0.72 \times 1.7 \times 0.14 = 0.170$ This argues for adsorption phenomena

Saturday 11/24/51

323 τ = 10.4 hr Bact + Super vs F 350 = 0.237
280 = 1.855
Bacteria + Superm (iced) Known and Nov 20/51

324 τ = 8.5 hr Bact + Super vs F 350 = 0.215
280 = 1.603
Bacteria + Superm (iced) Known and Nov 20/51

At 2[00] pm switched 324 to 12 pins from 3 pins
323 to 5 pins from 1 pin

Sunday 11/25/51 323 τ = 2.02 hr
324 τ = 2.04 hr
Collected 2[45] - 3[30]

323 Bact + Sup vs F 350 = 0.250 !? th.
280 = 0.768

Super vs F

350	300	290	280	270	260	250	240	230
0.015	0.049	0.161	0.278	0.298	0.260	0.221	0.214	0.350

324 Bact + sup vs F 350 = 0.224
280 = 0.693

Super vs F

350	300	290	280	270	260	250	240	230
0.019	0.048	0.149	0.249	0.273	0.251	0.218	0.225	0.385

At 3[15] pm switched 323 from 5 pins to 1 pins
" " 324 " 12 (15 min clock) to 3 pins

323

collected 4 pm to 5 pm

$\tau = 2.5 \times \frac{5}{6}$ Complete with Bact vs F

$= 2.08$ at 350 = 0.210; at 280 = 0.620

Thursday 11/23/51

324 - drew off 15 cc at 7 pm $\tau = 8.13$ hr (323 $\tau = 1.97$ hr)

bact + super vs F 350 = 0.226 280 = 1.58

Supernat vs F

350	300	290	280	270	260	250	240	230
016	.127	.690	1,190	1,178	.920	.586	.445	1.09

$\frac{1.19}{8.13} = 0.146$ $0.146 \times \frac{145}{226} = 0.094$/hr

Friday 11/24

322 $\tau = 1.98$ hr

collected 12^{30} — 14^{45}

vs F

Bact + Super 350 0.216

280 0.640

Supernat vs F

350	300	290	280	270	260	250	240	230
005	025	089	0.160	0.175	0.154	0.114	0.095	0.196

$\frac{0.160}{1.98} = .081$ $.081 \times \frac{145}{216} = 0.0545$/hr

Actually running on 5 pins - one of 6 missing contact

at 2 pm switched to 1 pin → 10 hr

Acquired

~~Readout~~ at 3^{30} pm 350 0.266

Bact + Super vs F 280 0.890

Supernat vs F

350	300	290	280	270	260	250	240	230
.010	040	.185	.325	332	.270	.186	.176	.460

1.5 hrs

0.325

325 − 160 = .165

$\frac{.165}{1.52} = \frac{0.085}{1.5}$

186 − 114 = 72

325 − 160 = 0.165

$0.165 \times \frac{145}{216} = 0.81$/hr

Sunday
cont'd

4:45 pm drained 324

I = 8.2 hrs computed

350 = 0.257

250 = 0.850

Back + Supernat vs F

350	300	290	280	270	260	250	240	230
014	045	185	315	335	288	240	.261	.520

315 − 249 = 66

240 − 228 = 12

Δ = 1 hr

5:15 pm drained 323

I = 10 hours computed

350 = 0.275

250 = 0.980

Δ = 1.5 hrs

Supernat vs F

350	300	290	280	270	260	250	240	230
015	053	.242	.410	.419	.346	.268	.277	0.593

410 − 278 = .132

268 − 221 = 47

2 × 1.5

[325] call on Tuesday
Sample with Buch vs F
at 350 = 0.163 at 280 = 0.448

[326] call on Tuesday
sample with barbara vs F
at 350 = 0.245 at 280 = 0.462 ~~Sample vs~~
[326] Sat. Dec 1 10 AM τ = 8.2 at 350 = .228 at 280 = .40

[325] changed from 4 pins to
2 pins at 1035 AM [should give τ = 8 hrs]

Thursday 11/29 [325] τ = 8.3 hr
[326] Glucose changed at 12 noon from 4 pins to 2 pins

[325] 1115 AM
Sample with Buch vs F
at 350 = 0.164 at 280 = 0.488
supernatant vs F

350	300	290	280	270	260	250	240	230
0.020	0.092	0.145	0.320	0.255	0.267	0.312	0.414	0.690

v<F
0.135
0.420

Dec 1 4 pm added histidine to raise conc by 5 mgm/L see later pages = 40/2.72

does not pass out X

~~Sample vs~~

To see if l-histidine in histidine precursor we take chemostat 324 and add l-histidine Exp [324B]. This chemostat summary at τ = 7.7 hours at 345 pm see later page

11/25 Friday
τ = 8.3 at 350 = 0.150 at 280 = 0.480

Sunday Nov 25/57 Histidine-less Tryptophane-less

[325] F + 5000 f/c tryptophane + 0.04 mgm/c Histidine

[326] ~~325~~ F + 2000 f/c tryptophane + 1.26 mgm/ Histidine

Bmps: B62/1c

No 66 is histidineless from Potts

[325] Bact + super vs F 350 = 0.160 280 = 0.458 $\bar{c}$ = 4.2 hrs

Super vs F

350	300	290	280	270	260	250	240	230
0.009	.045	.074	.115	.135	.140	.163	.215	.345

[326] Bact + super vs F 350 = 0.190 280 = 0.469 $\bar{c}$ = 4 hrs

Super vs F

350	300	290	280	270	260	250	240	230
.027	.055	.081	.122	.155	.176	.203	.290	.478

Wednesday Nov 28/57

[325]

call on Monday

Sample with Bacteria vs F

350 = 0.145 280 = 0.415

[326]

call on Monday

Sample with bacteria vs F

350 = 0.206 280 = 0.400

Working up [321] 4

Assume $\frac{y_2}{y_1} = \frac{250 \text{ abs}}{280 \text{ abs}} = 2.2$ for autolysed bacteria or something away out from bacteria

$\frac{x_2}{x_1}$ for precursor = 0.5

$x_1 + y_1 = A_1$ [280]

$x_2 + y_2 = A_2$ [250]

$\frac{A_2 - x_2}{A_1 - x_1} = 2.2$

$A_2 - x_2 = 2.2A_1 - 2.2x_1$

$A_2 - 0.5x_1 = 2.2A_1 - 2.2x_1$

$1.7x_1 = 2.2A_1 - A_2$

$x_1 = \frac{2.2A_1 - A_2}{1.7}$

However instead of using this formula we use it only for the first point of 0 hour and for later points we take differences at [200]

328 for precursor with sudden increase at tryptophane

supernatant vs F

350	300	290	280	270	260	250	240
0.001	.027	.171	.300	.300	.231	.137	.099

0.105/hr

230
.274

This sample was collected in ice and centrifuged in cold.

at 2^{05} pm call in ice vs F

Obour 350 = 0.125 280 = 0.539

supernatant cold centr. vs F

350	300	290	280	270	260	250	240	230
0.012	0.048	0.192	0.317	0.325	0.273	0.194	0.168	.363

at 2^{05} pm added tryptophane to bring concentr. up to 1 mg/l

$V = 20.3$ cc

Sample cold in ice 2^{05} to 3^{05} pm vs F

Obolus 350: 0.122 280: 0.543

supernatant centrifuged cold

350	300	290	280	270	260	250	240	230
0.008	0.046	0.203	0.334	0.334	0.264	0.170	0.135	0.326

at 3^{05} pm added trypt. to growth tube to bring conc. another mg/l

first time increases absorption of bacteria at 280!

Wednesday Nov 28/57

[328]

For precursor of L tryptophane

$\bar{\tau} = 5.2$ hours

switched from 2 pins to 5 pins at 12^{15} pm.

at 5^{20} pm $\bar{\tau} = 2.16$ hrs

clocked changed to give $\bar{\tau} = 1.8$ hrs

Thursday Nov 29/57

10^{15} AM $\bar{\tau} = 1.75$ hrs

Sample taken at 11^{30} AM

Sample with Bact. vs T

at 350 : 0.060 at 280 : 0.151

at 3^{15} pm Sample with Bacteria vs T

all (2.45 to 3.15) 350 T = 0.057 280 : 0.128

at 5^{40} pm changed from 22 to 19 pins

Friday Nov 30/57

{ Sample at 10^{30} to 12 noon with bact. $\bar{\tau} = 2.02$

350 : 0.070

changed from 19 pins to 10 pins at 4 pm

to be used for exp. with sudden increase of tryptophane conc. —

Sat Dec 1 10^{15} AM $\bar{\tau} = 3.7$ hrs

Sample with bacteria vs T

350 = 0.114 280 = 0.509

328 from 3:05 to 4:05 pm

Sample cell in ice vs F

at 350 = 0.130 at 280 = .580

supernatant contr. cold vs F

350	340	290	280	270	260	250	240	230
0.010	0.046	0.195	0.324	0.325	0.259	0.170	0.137	0.320

Sample pushed out at 405 vs F

~~vs F~~ at 350 = 0.130 at 280 = .557

supern. contr. cold vs F

350	340	290	280	270	260	250	240	230
0.003	0.025	0.160	0.275	0.275	0.214	0.129	0.093	0.254

At 0 hour at 280	340	340 = 340
— 0.87 × 0.03	~~26~~	
	334	
At 0 hr 1 hour at 280	−26	308 + 2% = 314
	308	
at 1 hr 2 hour at 280	324	278 + 4% = 289
	−26	
	−20	
	278	
		235 + 4% = 245
pushed out	275	
	40	
	235	

This looks like the supernatant

324 B "does histidine inhibit precursor?" No

Thursday Nov 29

Old Chemostat 324 now called 324B running at $\bar{\tau}$ = 7.7 hours [1 mg/l tryptophane; B/10]

Sample collected at 245 pm

Sample with Bact vs. F

350 = 0.240 280 = 1.670

Supernatant vs F

350	300	290	280	270	260	250	240	230
0.026	0.158	0.875	1.413	1.395	1.100	0.720	0.585	1.340

pursuant 0.11/hr

added 3 cc of 5 gm/l l-histidine, to 1.5 litre spring cane. to 10 mg/l in storage tank at 330 pm.

Friday Nov 30/51

$\bar{\tau}$ = 6.8 hrs

Sample with Cadmium vs. O.j.

350	300	290	280	270	260	250	240
0.262			1.03				

it powers out

Supernatant vs O.j.

350	300	290	280	270	260	250	240	230
0.019	0.153	0.935	1.560	1.520	1.180	0.740	0.550	1.400

histidine does not inhibit X

[321] continued [Pa Trypt. control]

Wednesday Nov 28

4 pm τ = 6.4 hrs

Sample collected 4 pm to 5 30 vs own juice (with Boek)

at 350 = 0.459 at 280 = 2.04

dil 1:4 at 350 = 0.132 x 4 at 280 = 0.687 x 4

own juice

supernatant vs. other juice

350	300	290	280	270	260	250	240	230
0.070	0.173	1.03	1.69	1.68	1.375	0.950	0.820	1.620

6 30 pm changed from 8 pins to 10 pins

Thursday Nov 29/51

10 15 AM τ = 4.95

Sample pushed out 10 20 AM

Sample with bacteria vs own juice

at 350 = 0.448 at 280 = 1.86

dil 1:4 350 = 0.140 x 4; 280 = 0.574 x 4

supern. vs own juice

350	300	290	280	270	260	250	240
0.075	0.133	0.760	1.30	1.331	1.08	0.766	0.704

230
1.40

End

2 Chemostats set up by adding 7 without Phosphorus to juice of [321] to increase Volume by factor 4 x Ph adjusted to 7 before autoclaving. These will be called 321 B and 321 C

P.T.O. →

Sat Dec 1st

10:15 AM

321 B Sample with Brot vs. O.j

τ = 9.2 hr 280 350 : 0.119 ; 280 : 0.785

| 0.016 | .061 | 0.359 | 0.603 | 0.606 | 0.483 | 0.332 (250) | 0.265 | 0.600 |

321 C Sample with Brot vs O.j

τ = 7.4 hr 350 = 0.133 ; 280 : 0.890

| 0.008 | .049 | 0.390 | 0.677 | 0.670 | 0.514 | 0.322 | .240 | 1.640 |

[321 B and 321 C] incubated at 1120 AM from 2 pins to 4 pins to give shorter τ's

~~Sunday Dec 2~~

At 3:15 pm collected in ice

~~321 B~~

[321 B] (ice) Sample with Brot vs O.j.

τ = 4.8 hr 350 : .134 ; 280 : 0.633

[321 C] (ice) Sample with Brot vs. O.j.

τ = 3.8 hr at 350 : 0.131 at 280 = .501

Supernatant vs. O.j.

350	300	290	280	270	260	250	240	230
0.023	0.060	.191	.319	.330	.281	.231	.242	.455

~~Monday Dec 3/81~~

280°

Friday Nov 30 '57 [321 B]. [321 C]

[321 B] against own juice at 3 40 pm
Sample 350 = 0.069 280 = 0.155 $\bar{t}$ = 8.8 hrs
with last P.H. 5.6
from blue morning
call 10 30 AM to 12 noon

[321 C] against own juice $\bar{t}$ = 7.6 hrs
call from 10 30 AM to 12 noon
350 = 0.084 280 = 0.227 PH = 5.8

~~Flow tubes~~
321 B and C should have
525 f/d Trypsin plaque

[321 B] sample call 3 30 pm to 5 pm
sample with last. vs O.j.
350 = 0.095 280 = 0.284

supernatant vs O.j.

350	300	290	280	270	260	250	240	230
0.027	0.054	0.114	0.185	0.215	0.233	0.284	0.522	0.160

[321 C] sample call 3 30 pm to 5 pm
sample with last vs. O.j.
350 = 0.105 280 = 0.415

supernatant vs. O.j.

350	300	290	280	270	260	250	240	230
0.019	0.040	0.140	0.240	0.265	0.257	0.264	0.420	0.960

321 B changed from 4 to 5 pm at 3 pm

Wednesday Dec 5

11:30 AM

$\tau = 3.77$ hrs

Supern vs. O.j.

350	300	290	280	270	260	250	240	230
0.013	0.033	0.096	0.167	0.191	.190	.204	.294	.583

Trypsin vs. O.j.

350 = 0.148

280 = 0.441

12/5/51 Thursday Wednesday. Repeat of morning

Collected 2 – 3:30 pm (ice)

321 B

Bact + Super vs O.j. at

350 0.154

280 0.441

Super vs O.j. (ice)

350	300	290	280	270	260	250	240	230
0.010	0.020	0.083	0.150	0.169	0.165	0.175	0.265	0.550

+15

$x_1 = \frac{2.2 A_1 - A_2}{1.7} = 0.091 + 15 = 0.105$ / 3.77

$y_2 = .134$

321 C $\tau = 3.88$ hr

Bact + super vs O.j

350 = 0.121

280 = 0.500

Supernat vs Oj (ice)

350	300	290	280	270	260	250	240	230
~~0.012~~ 0.010	0.073	0.142	0.256	.265	0.275	0.182	0.192	.391

+15

$x_1 = 0.226 + 15 = 0.241$ / 3.88

$y_2 = 0.079$

At 4:30 pm B put on 15 pins slow clock

C put on 12 pins slow clock

(to give about $\tau = 5$ hrs for each)

321 B 321 C

Monday Dec. 3/57

321 B $\bar{t}$ = 4.85

321 C $\bar{t}$ = 3.85

Samples in ice at 11^{45} AM vs o.f.

350 = 0.126 280 = 0.603

[321 B] Supern. vs. o.f.

$\bar{t}$ = 4.85	350	300	290	280	270	260	250	240	230
	0.005	0.029	0.197	0.358	0.361	0.294	0.205	0.182	0.430

[321 C] Sample (in ice) vs o.f.

$\bar{t}$ = 3.85 350 = 0.119 280 = 0.490

Supernatant vs. o.f.

350	300	290	280	270	260	250	240	230
.005	0.012	0.140	0.249	0.255	0.217	0.168	0.166	0.368

added 100 γ/l tryptophane at room (0.23 oc of stock solution)

His may have been an error and may be we added to 321 B

Tuesday Dec 4

Collected 10 am – 11^{30}

321 B $\bar{t}$ = 4.8 hr

vs organic 350 = 0.139

Bact + Supen 280 = 0.532

Supernat vs O.f. (in ice)

350	300	290	280	270	260	250	240	230
0.003	0.018	0.140	0.253	0.263	0.222	0.178	0.170	0.367

321 C $\bar{t}$ = 3.8 hr

vs organic 350 = 0.118

Bact + Supen 250 = 0.458

Supernat vs O.f (in ice)

350	300	290	280	270	260	250	240	230
005	020	0.133	0.240	0.248	0.214	0.178	0.188	0.386

Supernatant

321B ; 321C
Friday Dec 7/51 collected 220 pm to 420 pm
at 4 pm

B ; τ = 6.44 ← ~~Supernatant~~ Sample (ice) ~~vs. o.f.~~ → 350 : .155 280 : .776
C ; τ = 7.17 ← ~~Supernatant~~ Sample ~~vs. o.f.~~ 350 : .141 280 : .940
(ice)

B Supernatant ~~vs.~~ o.f. τ = 6.44

350	300	290	280	270	260	250	240	230
0.007	.040	.270	.470	.471	.378	.265	.227	.508

C Supernatant vs. o.f. τ = 7.17

350	300	290	280	270	260	250	240	230
0.007	.053	.396	0.688	0.675	.523	.340	.268	.662

B changed from 11 to 9 pins } 530 pm
C " " 8 to 7 pins }

12/8/51 Saturday Collected 8am - 10 am

321B τ = 7.94 hr ν Supernat vs Of

350	300	290	280	270	260	250	240	230
0.011	0.051	0.375	.638	.635	.498	.334	.274	.642

PH 7.45

321C τ = 8.25 hr ν Supernat vs Of

350	300	280	280	270	260	250	240	230
0.008	0.057	.463	.800	.780	.600	.384	.298	.753

PH 7.43

B switched from 9 pins to 13 pins } at 11 30 pm
C switched from 7 pins to 20 pins }

Sunday Dec 9/51 1030 AM call 10 30 AM to 11 30 AM

B τ = 5.61 Sample vs. o.f. 350 : H 280 : H

C τ = 2.94 Sample vs. o.f. 350 : .115 280 : .417

continued at later page

321 B, 321 C Thursday Dec 6/57

11 A M

B $\delta = 4.82$ Sample vs o.f. 350 = 0.143; 280 = 0.570

C $\delta = 4.85$ Sample vs. o.f. (ice) 350 = 0.133; 280 = 0.670

321 B supern. vs o.f

350	300	290	280	270	260	250	240	230
0.004	0.022	0.157	0.279	0.287	0.238	0.181	0.176	0.372

321 C supern vs. o.f.

350	300	290	280	270	260	250	240	230
0.004	0.028	0.231	0.410	.409	.326	.228	0.200	0.452

B changed from 15 pins (5.52 hr) to 13 pins at 11[50] AM

C changed from 12 pins (5.52 hr) to 10 pins at 11[50] AM

~~Collected from~~ 9[10] pm to 10[30] pm

[9[50] pm]

B ~~C~~ $\tau = 5.56$ hrs } run thru for 10 hours

C $\tau = 5.66$ hrs } 11[50] AM to 9[50] pm

[B] 9[50] pm sample; supern (ice) vs o.f.

350	300	290	280	270	260	250	240	230
0.001	0.034	0.232	0.409	0.407	0.313	0.212	0.182	0.420

[C] 9[50] pm sample; supern (ice) vs. o.f.

360	300	290	280	270	260	250	240	230
0.002	0.032	0.285	0.491	0.486	.376	.247	.202	0.509

B changed from 13 pins to 11 pins

C changed from 10 pins to 8 pins at 11[30] pm

329

to growth tube, make

1/2 cc added at 11[50] AM to ~~broth~~

concentration of mycoplasma by 2 mycs/cc

V = 22.6 cc

collected from 11[50] am to 12[50] pm in ice

sample sample vs F

1/2 350 : .258 200 : 1.220

supernatant vs F [centr. cold]

350	300	290	280	270	260	250	240	230
0.017	0.090	0.440	0.723	0.720	0.567	0.370	0.305	0.758

1 hr sample pooled out and read at 12[50] pm vs F

350 : 0.254 200 : 1.226

First hour increases about 1.0 at

supernatant centr. cold vs. F.

350	340	290	280	270	260	250	240	230
0.012	0.075	0.409	0.687	0.679	0.530	0.333	0.265	0.687

$e^{-\frac{t}{3.7}} = 0.763$

0.687
− 46
0.641 + 2% = 0.654

$\frac{0.654}{0.735} = 0.875$

654
− 75
579

0.86 × 0.88
579/735 = 0.789

curve refined:

$X_0 = 0.744$

$X_1 \approx 0.691$
− 46
0.645 + 2% = .658

$\frac{658}{744} = 0.885$

Friday, Nov 30/51

[329] tube 328 unit / mg/l

precursor

for exp. to increase hypotaphane hypotaphane
antibody. -

4 pm running at $\bar{c}$ = 6.85
Sample with antisera vs F
at 350 = 0.248 at 6800 = 1.920
flowrate set at 4 pm to give about
$\bar{c}$ = 3.5 hours we hope

Ext. Dec 1
1030 AM
$\bar{c}$ = 3.7 hr

Sample vs F
350 = 0.239 280 = 1.198 = 1.200

$\bar{c}$ = 3.7

[322] tube 328 (with 500 f/l hypo)

Ext. Dec 1
11 AM
$\bar{c}$ = 3.8 hr

Sample vs F
350 280

→ Supernatant vs F
[329]

350	300	290	280	270	260	250	240	230
0.010	.075	.430	.735	.730	.569	.356	0.279	0.725

0.42 / hr

this sample was centrifuged and
centrifuged cold

~~V = 22.6 cc~~ 22.6 $\times 10^{-3}$ liter to get ~~mg/ft~~
add $\frac{2}{1000}$ mg/cc or $\frac{22.6 \times 2}{1000}$ mgm

precursor with smaller no of trypt.

333 Monday Dec 3

1 mgm/l Trypp./

V = 109

running since Sunday 9 45 pm

τ = 3.78 Sample (ice) vs F [and F]

Supern. vs. F 350 = 0.232 ; 280 = 1.145

350	300	290	280	270	260	250	240	230
005	0.052	0.370	0.658	0.650	0.502	0.310	0.230	0.628
				+50		+50		

0.11/hr

10 am – 1 45 τ = 3.89 hr

---

0 hour call in ice [at 2 15] Sample vs. F

350 = 0.236 280 = 1.122

Supern vs F

0.015	0.070	0.404	0.680	0.672	.524	0.335	0.256	0.620

F vs H2O

350	340	290	280	260	250	240	230
0.014	0.042	0.056	0.067	0.074	0.093	0.120	0.181	0.605

at 255 pm added trypt to growth tube, to raise conc. by 2 mgm/l and took

0.10 collected in ice 255 to 305 pm

supern vs F

350	340	290	280	270	260	250	240	230
0.012	0.073	0.426	0.698	0.688	0.530	0.339	0.265	0.682

where is trypsophrane? [checked rel. activity 0.K.]

½ hour call, 325 to 335 pm in ice

Supernatant vs F

350	340	290	280	270	260	250	240	230
0.014	0.070	0.379	0.630	0.625	.484	0.312	0.246	0.610

[325]

Question is ~~normal~~ minimal "H" production of "member" involved? If so there should be no rapid fall between 1 hr and 2 hr like either.

~~Dec 3 10 AM~~

[325] running since Sunday noon at $\tau = 8.35$ h (with added lactalbumin)

Sample with test vs F

at 350 = .135 at 280 = 0.420

does not pair out (as before)

will be inoculated with B/1c/5 (B strain II) (added at 330 mm 5000/cc)

12/5/51 Wed 11am assayed for B/5

$\left.\begin{matrix}321\\310\end{matrix}\right\}10^2 = 631$

at 11:40 am added B/1c/5 (298/5 #11)

2 pm assay + TS $\left.\begin{matrix}286\\294\end{matrix}\right\}10^4 = 2.9 \times 10^4$

12/6/51 Thurs 11am

assay + TS $\left.\begin{matrix}175\end{matrix}\right\}10 = 1750$

$\tau = 7.8$ hours

it falls e in 7.5 hours

Dec 7. Friday at 5 pm $\tau = 7.8$ hrs

Sample vs. F

350 = 0.271 at 280 = .678 ?

→ why is density doubled?!

Sunday Dec 9 at 11 AM

Sample vs. F. 350 = .170 280 = .480

→ normal

Rev dec 3 5pm

Exp on 329 to see if trypto-
phane disappears. $\bar{c} = 3.7$ hr

1.) I.c pushed out. dead, No 1
2.) trypt added to growth tube to raise
cells. ½ mg/ml. - after 10
minutes with lights on and
inflow stopped sample No 2 was
pushed out. dead.

Sample No 2 vs F
350 = .244; 280 = 1.193

Supernatant (ice) No 2 vs Supern No 1 (ice)

350	300	290	280	270	260	250	240	230
0.002	0.010	.041	0.057	0.055	0.038	0.015	0.005	0.036

- prec 0.029
+ 0.032

Water 1145 AM inedn dec 5/57

350 — 0.235 — 280 = 0.800

[33]

[1 hour] cell 353 to 405 in ice vs F

supernatant vs F

.012	0.063	.336	.560	.557	.433	.280	.226	.580
			+10			+10		

F vs. kpo

350	300	290	280	270	260	250	240	230
~~0.018~~			~~0.088~~			~~0.138~~	~~0.165~~	
0.017			0.076			0.130	0.195	

1 1/2 hour incl Sample vs F

350 = .310 2000 = 1.180

620

.560

supernatant vs. F

350	300	290	280	270	260	250	240	230
0.018	0.065	0.309	.513	.517	.413	.279	.239	.566

2 hour incl Sample vs F

350 = .347 2000 = 1.220

supernatant vs. F

350	300	290	280	270	260	250	240	230
.022	.064	.293	.481	.490	.395	.274	.244	.560

$x_1 = 462 - 15 = 447$

$\frac{447}{560} = 0.80$

Weden. Dec 5/57

Interesting possibility may have: did we forget to add supernatant to tank? NO

310 : 235 2000 = 0.500

It does not prove out! Is it a contaminant? [Or is it a mutant of B1A selected at high supernatant?]

at 5:30 pm added 10g tryptophane to tank of 32/B to raise conc. by 50 p/c

---

Tuesday Dec 11/51

B (new series with increased light)

$\tau = 5.5$ hr 11:45 AM (red) Sample vs o.f.

350 = .142 280 = 0.598

B Suspens (ice) vs. o.f.

350	300	290	280	270	260	250	240	230
0.018	0.048	0.242	0.360	0.374	0.324	0.264	.255	.472

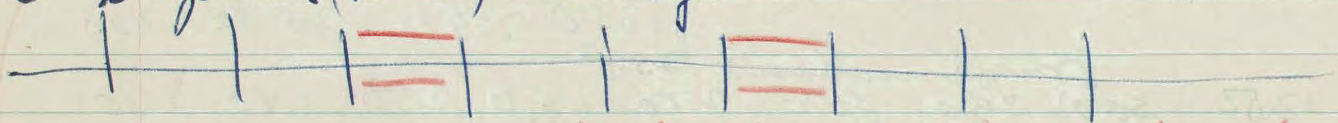
too much repeat below:

C (new series) (calc. 9:30 AM to 11:30 AM)

$\tau = 2.94$ hrs 11:45 AM. Sample vs. o.f.

(ice) 350 = .146 280 = .325

~~C Suspens (beef) vs. o.f.~~



at 350 = .146 is higher (at high tryptophane) than earlier measurement at same τ (at low tryptophane)

at 4:45 pm

B (new series) $\tau = 5.5$ Sample (calc 2:15 pm to 4:45 pm) vs o.f.

(ice) 350 = 0.144 280 = 0.598

Suspensions vs o.f.

350	300	290	280	270	260	250	240	230
0.008	0.020	0.192	0.335	.344	.284	.210	.106	.408

C (new series) (ice) $\tau = 2.94$ Sample (3:15 to 4:45 pm) vs. o.f.

350 = 0.148 280 = 0.332

321B 321C continued

Sunday Dec 9/51

10 30 AM call 10 30 AM to 11 30 AM (ice)

B Supern. (ice) vs. o.j. $\bar{\tau} = 5.61$

350	300	290	280	270	260	250	240	230
.007	0.034	.226	.407	.410	.330	.239	.215	0.483

C Supern (dec) vs. o.j. $\bar{\tau} = 7.94$

350	300	290	280	270	260	250	240	230
0.007	0.019	0.071	0.129	0.143	0.139	0.144	0.203	0.408

C changed from 20 pins to 8 pins 12 35 pm
added 1 mgm/l (raise) of hypo to tank

Monday Dec 10

321B τ = 5.6 hr ~~0.443~~ collected 9 20 am – 11 30 am

B Bact + super vs OJ 350 = 0.143 280 = 0.667

Supernat vs OJ

350	300	290	280	270	260	250	240	230
004	025	.225	.407	406	318	216	0.184	0.432

321C τ = 7.8 hr Coll 9 30 am – 11 30 am

(Chem readings) Bact + super vs OJ 350 = 0.197 280 = 0.393

350	300	290	280	270	260	250	240	230
020	012	025	.056	089	102	.112	.141	0.174

X_1 = .006 ∴ Plain B/it does not pan out

Bacterial kills rose from 0.141 at $\bar{\tau}$ = 7.17 hr before adding hypo. to 0.197 after adding hypo ($\bar{\tau}$ = 7.8 hr)

C changed from ~~8~~ pins ~~to~~ 20 pins at 4 45 pm

5[20] pm

B sample call 2[10] pm to 5[10] pm. 250 o.j.

350 = 0.156 280 = .798

Lupeon vs. o.j.

350	340	240	280	220	260	250	240	230
.015	.045	.310	.534	.536	.428	.290	.243	.558
0.015	.047	.306	.529	.532	.424	.287	.238	.550

switched from 11 pins to 19 pins at 6[05] pm
standard time 3.74

Friday Dec 14/57

B = 3.78 hr = τ ; C = 6.86 hr = $\bar{\tau}$

2[20] pm [B] sample vs. o.j.

θ = 3.78 hr; 350 = 0.130 280 = .458

Bacteria seem sitting at bottom of sample tube

τ = 3.78 hr

350	340	240	280	Lupeon 270	260	vs. o.j. 250	240	230
0.09	0.092	0.120	0.217	0.233	0.205	0.175	0.180	0.343

2[20] pm [C] sample vs. o.j.

$\bar{\tau}$ = 6.86 350 = .196 280 = .430

indol added to boiling conc. to about 20 mg/l at 3 pm

condensed at boiler proof

321 B ; 321 C | ~~Thursday~~ Tuesday Dec 11/57

C switched from twenty pins to 15 pins at 5^05 pm Tuesday Dec 11/57

B switched ~~[illegible]~~ (from 13 pins) to 9 pins at 5^30 pm

Wdn: Samples v.o.f. 350 : .270 280 : .057

12/12/57 321B $\bar{t}$ = 7.80 hr, Supern vs. o.f.

5 pm	310	300	290	280	270	260	250	240	230
	0.015	0.047	.352	.600	.594	0.463	.300	.237	.568

B switched from 9 pins to 11 pins at 550 pm

321 C $\bar{t}$ = 3.92 hr

245 pm Sample vs o.f.

350 : .168 280 : .370

dumped from 15 pins to 12 pins at 250 pm

should give $\bar{t} = 3.92 \times \frac{15}{12} = 4.9$ hrs

~~Friday Dec~~ ~~Thursday, Dec. 13/57~~

→ this sample was probed out and repeat!

350 : .146 ; 280 : .800

~~Fri~~ Thursday Dec 13/57

B ; $\bar{t}$ = 6.46 overnight

C ; $\bar{t}$ = 4.95

C 10^50 AM Sample vs. o.f.

310 : .160 280 : .374

dumped from 12 to 9 pins at 11 AM

B 5^20 pm Sample calc. 9^40 AM to 2^00 pm vs. o.f.

350 : 0.148 280 : .707

Supern vs. o.f.								
0.015	0.045	.310	.534	.536	.429	.290	.243	.558

~~Fri~~ Tuesday Dec 11

336 [like 333] for molded rose of tryptophane 1 mg/l tryptt, V=109cc running at $\bar{\tau}$ = 3.8 hrs

Sample vs F at 5 45 pm

at 350 = 0.263 at 280 = 1.317 O.K.

Wednesday Dec 12

$\bar{\tau}$ = 3.77 hr (10 45 am – 2 pm)

Collected (ice) 2 pm — Boat+Super vs F 350 = .275 O.K.

Supernat vs F 280 = 1.535

350	300	290	280	270	260	250	240	230
0.015	0.090	0.678	0.980	.965	.740	.458	.340	0.880

much higher than 333

PH 6.7

Sample 0 ~~0 hr~~ (0 hr. 10 min but something probably went wrong at 10 min)

Supern vs. F

350	300	290	280	270	260	250	240	230
017	.113	0.673	1.127	1.110	0.850	0.530	0.403	1.03

due to the added tryptophane, this is a bigger increase than that should be 1.040

Sample 1 ~~1 hr~~ (10 to 20 min but must spin at 10 min)

Supern ~~vs.~~ F.

350	300	290	280	270	260	250	240	230
019	.119	.653	1.090	1.070	.834	.535	.410	1.02

Sample 2 25 – 35 min

350	300	290	280	270	260	250	240	230
0.019	0.119	0.630	1.048	1.028	.782	.496	.377	0.960

Sample #3 55 – 65 min

350	300	290	280	270	260	250	240	230
0.019	0.115	0.640	1.080	1.050	0.795	0.488	0.381	0.960

Sample 4 85-95 mm

350	300	290	280	270	260	250	240	230
020	.112	.630	1.045	1.022	.780	.490	.375	0.950

Sample 5 115-125 mm

350	300	290	280	270	260	250	240	230
0.018	0.104	0.601	1.014	1.00	.761	0.480	0.365	0.930

Summary 321 B and C, P and typ. W
low, changing E gives for any E
apparent typ phosphate incubation. Outpouring
of precursor increases with increasing E,
abating but kept to track step
outpouring of precursor. —

Evaluation of 296. –

at 5 hrs = t it gave $A_1 = 0.5$ precursor level

at 10 hrs = t it gave $A_2 = 0.75$ precursor level

at 5 hr rate = 0.1 ; at 10 hrs rate = 0.75×0.1

assuming the proportional to NH_4Cl concentration

$\frac{C_2}{C_1} = 0.75$ $\quad \frac{C_2}{C_2} = \frac{1}{0.75}$ $\quad$ But we also have

$C_2 + \overbrace{\alpha(A_2 - A_1)}^{B} = C_1$ $\quad C_2 + B = C_1$

where $\alpha = \frac{53}{200}\,\frac{20}{0.6} = \frac{53}{6} = 9$

$0.75\,C_1 + B = C_1$

$B = \frac{C_1}{4}$

$C_1 = 4B = 9$ mg/l NH_4Cl

$C_2 = 0.67$

$B = \frac{9}{4}(0.25) = \frac{9}{4}$

338 like 336 and 333 for

12/13/51 Set up with 10 cc growth tube, 1 mg/l trypt. Inoc with B/it. Ran at 4 pins

12/14/51 Flow rate overnight at 4 pins = 6.83 hr
At 9 am increased to 6 pins. At 4:30 pm τ = 4.77 hr
At 4:30 pm switched to 8 pins.

12/15/51 ~~Saturday~~
τ = 3.42 hrs overnight

F vs Thio

350	300	290	280	270	260	250	240	230
0.012	0.038	0.040	0.058	0.063	0.081	0.105	0.168	0.620

1045 AM Sample with heat vs F
350 = .245 280 = 1.195

Supern. vs F (20 min collection)

350	300	290	280	270	260	250	240	230
0.010	0.066	0.397	0.680	0.670	0.520	.335	.265	.675

[same tube removed centrifuge and was left with Roberts] ~~Centrifuge~~ Centrifuge decreased 11 20 pm

2:45 pm τ = 3.50 hr collected 225 pm to 245 pm
Sample with heat vs F.
350 = .247 280 = 1.203

Supern vs F

0.014	0.070	0.413	0.700	0.690	.536	.340	.260	.654

~~Sample (0) collected from 2:20 pm to 2:30 pm~~
cell from 10 min to 0
[0] supern. vs. F.

0.007	0.055	0.400	0.680	0.663	0.510	0.315	.235	.620

0.22 ml dilute of 1 gm/L to 10 gm/L

Input added at 0 time to growth tube to raise by 2 mgm/L

supernatant [10] (call from 0 to 10 min) vs F

350	300	290	280	270	260	250	240	230
0.008	0.066	0.428	0.717	.699	.530	332	.252	.673

supern [20] (call 10 to 20 min) vs F

0.006	0.071	0.420	0.695	.675	.517	.325	.252	.674

supern [30] (call 20 to 30 min) vs F

0.009	0.072	.414	.681	.663	.510	.322	.251	.663

supern 40 (call 30 to 40 min) vs F

0.009	0.072	.400	.672	.655	.505	.319	.246	.660

supern 50 (call 40 min to 50 min) vs F

.007	.069	.405	.672	.657	.502	.318	.245	.653

supern 60 vs F

0.008	.070	.404	.667	.650	.498	.314	.244	.660

supern 70 vs F

008	0.069	.398	.660	.634	.492	.312	.240	.640

321 C with indol

sample with bacteria vs diff. E. c.

$\tau \approx 6.86$ hrs 350 = 0.207 280 = 0.960

. tryptone vs. o.f.

350	300	290	280	270	260	250	240	230
0.050	0.045	0.270	0.665	0.760	0.700	0.502	0.428	0.640

~~0.041~~ ~~extracted~~

extracted with Ether vs o.f.:

0.041	0.022	0.038	0.068	0.113	0.134	.130	.167	.215

does not make precursor from indol in presence of tryptophane

Continued at later pages

~~Sat. Dec 15/51~~

339 tours strain 121-35 ↓ antibacterial loss made M/It $\tau = 8$ hrs

500 γ/l tryptone

sample all 10 AM to 6 PM at 350 = .187 280: 449

350 & 300	290	tryptone vs F	260	250	240	230
0.042	0.081	.095	0.137	.183	.232	.342	.645	1.32

340 same as 339 + 20 mg/l indol

$\tau = 6.1$ hrs

sample all 10 AM to 6 PM 350: .554 280: 2.01

either contaminant or it grows on indol

340 Monday Dec 17/51 ~~not so low~~

$\tau = 8.15$ hrs (why not 6.1 hrs?) samples at 345 pm

350 = 0.362 280 = 1.280

350	300	tryptone vs F	280	270	260	250	240	230
0.146	.148	.422	.900	1.07	1.05	1.00	1.47	2.45

Supern 90 vsF

350	340	240	290	270	260	250	240	230
0.010	0.069	.402	.670	.657	.500	.317	.244	.620

not specially cleaned cultures

Supern 90 vsF

0.009	0.068	0.399	.660	.643	.493	.310	.237	.620

Supern 100 vsF

0.010	0.067	0.394	.660	.644	.494	.312	.237	.618

Supern 110 vsF

0.010	0.068	.397	.660	.642	.493	.310	.235	.619

Sampl 120 vsF 350 = .267; 280 = 1.205

this reached the a full by 40 + 60

Supern ~~Sampl~~ 160 vsF (cell 150 min to 160 min)

0.010	0.066	.392	.665	.650	.498	.314	.237	.613

Sample 170 350 = 0.270; 280 = 1.229

Precursor never really falls; ~~if~~ low if trypt is added only to growth tube

Bacteria do not rise much

continued at later pages

Suspension 20 to 30 [30] vs. F

350	300	290	280	270	260	250	240	230
.012	.070	.419	.698	.685	.531	.352	.289	.705

Suspension 30 to 40 minutes [40] vs F

.008	.065	.480	.660	.651	.506	.335	.273	.670

Suspension 40 to 50 minutes [50] vs F

.008	.063	.379	.641	.630	.487	.312	.262	.648

Suspension 50 to 60 min [60] vs F

0.010	0.066	.384	.640	.629	.484	.323	.265	.645

Suspension 60 minutes 70 [70]

0.009	0.060	.359	.610	.598	.462	.308	.253	.614

Sample with Bract. 70 min to 90 min [90]

350 = 0.294 2090 = 1.220

Suspension [90] vs F

0.008	0.055	.340	.570	.560	.433	.292	.238	.580

Suspension [100] vs F

0.008	0.051	.325	.550	.540	.419	280	.230	.555

Sample [110] with Bract vs F

350 = .302 280 = 1184

Sunday Dec 16/57

338 continued. 4

Shell added hypo to grow tube and tank to raise conc by 1 mg/l. (Lights will be off)
Tavernight 4.2 hrs

350	300	F vs H2O			250	240	230
0.010	0.034	0.048	0.055	0.059	0.072	0.127	0.558

11 AM (1050 to 1100 CST)

Supern vs F [-10]

0.010	0.064	0.426	0.728	.712	.552	.363	.295	.723

Simple with bact vs. F
350: .255 200: 1.285

$\frac{.728}{3.42} - \frac{1}{7.76} = .121$ /hr (-10 to 0)

11^05 to 11^15 AM (CST) Supern [0] vs F [0]

0.008	0.060	0.421	.724	.706	.548	.356	.284	.715

hypo added to mouth tube and tank at "11" exp. time. — to raise conc 4 l mg/l

Supern (0 to 10) [10] vs. F

0.009	0.069	.436	.735	.719	.555	.365	.299	.746

Supern (10 to 20) [20] vs F

0.010	0.069	.419	.710	.695	.539	.359	.288	.710

338

Monday 17/57

$\tau = 3.35$ hrs overnight

10.50 AM Sample vs F

350: 0.377 280: 1.108

Supernatant vs F

350	340	290	280	270	260	250	240	230
0.014	0.045	.155	.255	.264	.220	.170	.152	.280

rate = $\frac{.230}{3.4} \cdot \frac{.145}{377+23} = 0.025$/hr

It might be contaminated because were not "sterile" yesterday in our operations. Suggest added to tank was perhaps 4/5 of mg/l rather than 1 mg/l because of amount left in 5 cc pipette. — Density should be 0.450 which should read 0.415. ~~Maybe because of oxygen limitation has shock & is produced and Chromobact is hypothesized to be grown.~~

~~Hollis~~

We should repeat with better oxygenation and may be 2 mg/l added hypothesis

aeration rate increased at 11:20

Sample call 3 pm to 315 pm vs F

350 = .455 280 = 1.635

Supernatant vs F

020	085	422	.710	.710	.575	402	.348	.750

pour up 4 pm next run

End

~~350 [250]~~ Expern. (110 to 120) [120] vs F

| 0.007 | 0.050 | .303 | .516 | .508 | .393 | .264 | .216 | .519 |

Expern (120 to 130) [130]

| 0.010 | .055 | .307 | .512 | .504 | .396 | .269 | .223 | .529 |

Sample with duct [140] vs F

350 = .315 280 = 1.185

~~Expern~~ Expern. [160] vs F

| 0.007 | .047 | .276 | .464 | .458 | 356 | .240 | .200 | .472 |

Sample [170] vs F

350 = 0.320 ; 280 = 1.165

Expern. [190] vs F

| 0.007 | .045 | .260 | .440 | .436 | .339 | .232 | .195 | .450 |

Sample [225] vs F

350 0.335 280 = 1.160

350	340	290	280	F vs N20	250	240	230
0.010			0.060		.091		0.600

aeration slow Arrangement this experiment

337 Wed. 19/5?

3/684/lt strain (No 5 for)

10 mg/l tryptophane

500 γ/l tryptophane

τ = 3.7 hrs

10 50 am sample 350 : 0160 vs F

280 = 0.423

350	300	290	fingers about 270	260	250	vs	240	230
00A	032	050	080	104	119	140	203	352

changed from 5 pins to 3 pins at 11 AM

Thursday 20/51

p.m. τ = 6.0 hrs overnight

4 pm Sample with Rochester vs F

350 : 154 ~~280~~ = 446

fingers about vs F / Rochester F

.0010	.052	.082	.130	.162	.177	.221	.300	.520

+25 +25 does not pour out →

some strain!

did not revert on tryptophane broths. —

blank made 684/lt – 337

End

← this pours out! because of large τ may be tryptophanase ?? May be when inoculated P was introduced and growth tube became P Ltd.

$\frac{480}{16} = 0.030 \times \frac{115}{2}$

organisms running at about 8 hours since

2.30 pm (Dec 20) Friday Dec 21 τ overnight 9.6 hrs

Sat. noon τ = 7.8 hrs

Sample vs. d.f. at 350 : 0.195 at 280 : 500 at 515 pm at 350 : 0.188 280 : 0.464

changed to τ ~ 16 hrs noon

from previous page

321 B and C

Thursday Dec 17

Re B mottled from 19 pins to 9 pins and 2 nigrg hypostaplane added to tank 2 (3/4 liter juice) at 6 45 pm should give T = 7.0 hrs

Tuesday Dec 18/51 B T overnight

Sample call for 12 noon to 2 pm vs. 0.j.

350 : .128 280 : .360

there is some small growth or redimmed does not pass out now, but tracheal tubes low! Put fresh growth tube on it to get Bacterial toxin.

Wed Dec 19/51 321 B T overnight = 17.8 hrs

350 = 0.352 ← 280 = 1.23

vs own juice

does it match?

500 : 0.153

500 : 0.160

450 : 0.175

400 : 0.205 vs.

350 : 0.253 0.j.

280 : .870

Thursday

T = 15.7 hours

Sec call from 1 30 pm to 4 pm (flow rate changed at 230 to 8 hours from 16 hours) mushed out by 10%

Suspension V3 0.j.

0.031	.065	.274	.479	.502	.428	.310	262	.490

341/ 0 to 10 min collection Sample [10]

Supern [10] vs F

.010	.087	.491	.810	.790	.610	.393	.310	.765

Supern [20] vs F

.012	.087	.465	.765	.750	.680	.375	.303	.740

not iced

Supern [30] vs F [not iced]

.013	.091	.450	.740	.726	.563	.367	.296	—

Supern [40] vs F

.014	.090	.438	.717	.703	.547	358	.289	—

Supern [50] vs F

.016	0.093	.426	.693	.680	.534	.348	.279	—

Supern [60] vs F

.014	.081	.403	.660	.650	507	.332	.265	—

Sample [70] vs F 350: 0.302 | 280: 1.274

Supern [80] vs F

0.014	.084	.380	.622	.613	.480	.316	.255	—

Supern [90]

014	082	.370	.603	595	462	308	247	—

Sample [100] with Bact vs F

350: .334 | 280: 1.305

Sample [110] with Bact vs F

350: 0.301 | 280: 1.274

[341] for repeat of 338 Sudden rise of trypt in growth tube and tank. — V = 109 cc
1 gm/l trypt. strong aeration
Media Dec 18 [Dec]
τ = 6.9 hrs
call 4 pm to 4:45 pm
Sample with best vs F
350 : 0.248 ; 280 : 1.532
[280] − 2[350] = 1.036
[rate] = $\frac{1.036}{6.9}$ $\frac{145}{248}$
(bins changed 6 pm) = 0.094/hr

Thursday Dec 20/57
T overnight 3.53 hrs
at 3 pm Sample at 350 : .248 V.S.F
Supern. vs F (back F) at 280 : 1.198

350	340	290	280	270	260	250	240	230	
0.014	.072	.416	.700 +25	690	652	360 +25	300	760	O.K.

aeration slightly changed. (still strong)
Made F pads

Friday Dec 21/57
T = 3.53 hrs
overnight
Sample (4:50 PM - 10:00 AM) vs F
350 : .250 280 : 1.211
Supern. vs. F

0.011	0.068	.418	.710	.700	.544	.347	.265	.651

4 cc 1 gm/per liter trypt in 10 cc H2O
Trypt 1.1 cc into growth tube, Rest into Tank at
at 10:29 AM [exp. time] (about 4 mg/l added)

This indicates that growth rate yesterday was not Oxygen limited but rather that tryptophanase concentration was low because of tryptophanase in growth tube.-

2.15 / 8 x 3,5	= 0.077

P/ It Inoculated tubes show humidity only in samples [10] and [20]

Beckman at 350 a/[10] : 0.213 - 120 = .093 ~ .092

Beckman at 350 a/[20] : 0.187 - 120 = .067 ~ .0565

Beckman at 350 at [30] = 0.140 at 280 = 0.820

Tryptophanase must be reduced by tryptophane curve c very low by the 20 & 30 min collection

Nephelometer on [10] : 71 x 2 = 142 - 4 = 138

[20] = 29 x 3 = 87 - 4 = 83

Heat Stand : 60

Assay of later tube and rate of rise of 6350 should together define c in growth tube and it should be really less than 10 g/l

Sunday Dec 23/57

Beckman at 350 [40] = 0.124 0.117

Sample 80 extracted with ether 280 = .0[illegible] 250 = .154

versus the this is not trusted

To test for indol repeat at experiment needed and for indol tested without unbalancing sample :-

Assay [40] = 3.4 10^6 ; 80 = 2 x 10^6 ; 110 = 2.7 10^6

This would give 5 g/l

341

Supern, [110]

| 0.013 | .078 | .343 | .564 | 538 | 439 | .292 | .237 | — | X_1 = 558

Supern [120]

| 0.015 | .078 | 0.339 | .550 | .546 | .433 | .290 | .239 | — | X_1 = 540

Sample [130] vs F 350 = 0.372 280 = 1.365

Supern [140] vs F

| 0.020 | .086 | .323 | .524 | .525 | .420 | .293 | .249 | — | X_1 = 507

Supern [150] vs F

| 0.018 | 0.080 | .310 | .505 | 505 | .407 | .280 | .236 | — | X_1 = 488

Sample [160] vs F

350 = 0.412 280 = 1.394

Supern [170] vs F

| 0.017 | .074 | .290 | .472 | .472 | .380 | .262 | .220 | — | X_1 = 457

Supern [180] vs F

| 0.020 | .074 | .285 | .460 | .461 | .370 | .260 | .220 | — | X_1 = 442

Sample [190] vs F

350 = .460 280 = 1.470

From chemostats 342 (Sample kept like B/C) Dil 1 to 20 and 1/2 + 10 cc added to supernatants [ambeland] Inoculated with B/C about 4 pm

---

Sat. 10 AM Sample vs F

350 = 0.296 x 4 ; 280 = 1.184 x 4

Supern vs. F

| 0.101 | 0.245 | 1.350 | 2.150 | 2.04 | 1.78 | 1.259 | 1.170 |

342–342A

Expern [0 – 20 min] [20] vs F

| 0.011 | .030 | .140 | .238 | .247 | .210 | .164 | .173 | – | $x_1 = 212$

Expern [20 to 40] [40] vs F

| 0.012 | .038 | .168 | .282 | .294 | .246 | .194 | .208 | $x_1 = 250$

Expern [40 to 60] [60] vs F

| 0.012 | .048 | .174 | .294 | .305 | .259 | .206 | .228 | $x_1 = 259$

Expern [60–80] [80] vs F

| 0.013 | .040 | .183 | .307 | .315 | .270 | .214 | .235 | $x_1 = 272$

See graph continued on later pages

Exp changed 342A to change of flowrate. —

Sunday Dec 23 ~~morning~~ 7 hrs

10 AM Expern vs F

240

| .0.07 | 0.025 | 0.125 | .220 | .228 | .193 | .144 | .143 | $x_1 =$ 200 $x_1 = 220$

switched from 8 pins to 4 pins to slow flow by factor 2 at 10 AM exp. time

Expern (0 to 40) [40] vs F

| 0.007 | 0.023 | 0.131 | .226 | .234 | .195 | .145 | .147 | $x_1 = 207$

Expern 40 to 80 [80] vs F

| 0.008 | 0.030 | 0.146 | .25 | .260 | .221 | .170 | .174 | $x_1 = 224$

(0.029 per hour at T=7 should give a rate of 14 per hour)
– more accurately 15.9 and even more accurately

342 at 25°C for precursor, ~~sudden~~ temp. rise
~~Thurs~~ Weeks dec 1951 $\tau = 10.6$ assay $7.3\ 10^7$
at 5 pm

Thursday
pm 8 overnight 10.67 hrs
at 330 pm Sample with Bact. vs F.
350 : 0.127 ; 280 : 0.604

Supern. vs F. (last F)

350	340	290	280	270	260	250	240	230
0.009	0.037	.208	.358	.362	.297	.225	.237	.750
			+25			+25		

about $\tau = 0.6$ hrs (about 4 pm) Stirred from 5 min to 8 min to give

~~Friday~~ Dec. 21.

8 overnight 7.04 hrs
4 pm Sample with bact vs F
350 .135 : 280 : 0.494

supernatant vs F

0.005	0.020	0.113	0.109	.205	.175	.117	.097	.144

Sat. Dec 22 1951 prints out about 0.030/hr
τ ; 26° at 8 am 345 pm to 926 pm ~~AM~~ [18 hr]
V=103 Sample at 1025 AM vs F
$\tau = 6.92$ 350 : 130 280 : .503

Supern vs F

0.007	0.027	0.127	0.225	.235	1.99	.153	.157	—

~~Temp rose to 37° in 3 min~~

Another subsequent Sample vs F
supern vs. F [0]

.010	.030	.137	0.230	.240	.203	.156	.160	—

V = 206

temp. raised to 37° in 3 min to 37°

Set overnight (4) Suspens 400

022	063	0.299	507	520	440	341	382

Yt pursuant at rate of $0.02\cancel{4} + \frac{200}{14} =$

$= 38.3$ compared with value at $\bar{c} = 7$ hrs

of $\frac{200}{\cancel{7}} = \cancel{38.3}\ 28.6$ or rather approximately

but $\frac{p(\bar{c}=14)}{p(\bar{c}=7} \approx \frac{38}{28} = 1.35$

Dec 24, 1951 From 5^{55} pm to 10^{25} am call 12 hrs → $\bar{c} = 13.7$ hr

Collected $10^{25} - 11^{35}$ am $t = 24$ hr $e^{-t/14} = 0.199$ $(1-e^{-t/14}) = 0.801$

Suspens vs F

015	056	321	.550	.550	438	302	269	$x_1 = 0.534$

Dec 26 $\bar{c}$ from 12/24 to now = 14.4 hr

Collected from 10^{20} am — 11^{20} AM

Suspens vs. F. $x_1 = .592$

0.010	0.055	.0349	.600	.599	1.470	.315	.272	—

$1 - e^{t/\bar{c}} = 0.094$

F vs H_2O at 280 = 0.044 at 250 = 0.084

F vs Nothing

at 280: .091 at 250: .161

Sample call 12^{20} pm to 2^{20} pm vs F $\frac{350}{280} = .142$ / $.853$

0.020	.090	.361	0.624	.622	.498	.344	.304	

$\frac{-21}{0.603}$ $\frac{-26}{.318}$ $x_1 = .592$

$\frac{.592}{14} = 0.0423$ corrected

3.42A

Lupsem [~~100~~ 120] vs T

| 0.010 | .023 | .161 | 0.274 | .280 | .236 | .181 | 189 | X = 248

Lupsem [160] vs T

| .010 | .034 | .170 | 0.290 | .298 | .249 | .188 | .194 | X = 264

Lupsem [200] vs T

| .010 | .033 | .176 | 0.303 | .310 | .256 | .192 | 195 | X = 279

Formula:

$$C = [\text{Rate } \bar{C}(14\text{hr}) - C_0(\bar{C}=7\text{hr})]\left(1 - e^{-t/\bar{C}}\right) + C_0$$

$$C - C_0 = [\quad]\left(1 - e^{-t/\bar{C}}\right)$$

	$1-e^{-t/\bar{C}}$	
20	~~.0242~~	~~0.0024~~ .0024
60	~~.0715~~	~~0.0071~~ .0071
100	~~.1192~~	~~0.0118~~ .0118
140	.1663	~~0.0212~~ 0.0166
180	~~.2120~~	~~0.0212~~ .0212
220	→	.0258
260	→	.0305
300	→	.0350
340	→	.0397
380	→	.0442

~~Remark rise of 27.0 hr~~
~~Logmean gradient rate of~~
~~140~~ ~~180~~ 27.0 + 14.3 = 41.3 at $\bar{C} = 14$
in place of 28.6 at
$\bar{C} = 7$ Inversely factor 1.45

(0) Lupsem [240]

~~see graphs~~ | 0.010 | 0.030 | .181 | 0.318 | .325 | .268 | .198 | .203 | X_1 = 295

(1) Lupsem [280]

~~duplic~~ | 0.010 | 0.033 | .195 | 0.335 | .342 | .280 | .206 | 209 | X_1 = 312

(2) Lupsem 320

| 0.010 | 0.036 | .205 | 0.349 | .354 | .293 | .215 | .219 | X_1 = 325

(3) Lupsem 360

| 0.008 | 0.032 | .208 | .354 | .357 | .292 | .241 | .218 | X_1 = ~~373~~ 335

to correct for impurities
we add 0.0025 and
get for rate at 37°C 0.660
at $\bar{t}$ = 6.6 hours

The End

~~[illegible]~~ same day
Collected 2^40 pm - 3^10
Supernt vs F

350	300	290	280	270	260	250	240
013	066	392	659	647	500	328	270

X_1 = .660

$\frac{0.660}{6.66} = .099/hr$

At 3^10 pm switched to air + 5% CO_2
~~Collected~~ 4^40 pm ~~to 5 pm~~ pushed out 3cc
Supernt vs F

350	300	290	280	270	260	250	240
0.013	.065	.390	.650			326	

after 1 hour 25 min) (no change

321B also 342

321B continued.

Sunday Dec 23

running since yesterday 5-30 pm

at $\bar{t} = 15.5$ hrs does it pursue? Yes we should blame hyperkinase!

Sample call 10 AM to 1:30 pm ; 350 : 0.223 ; 280 : 615

Supernatant vs. 0.1

350								
0.083	0.071	0.203	0.363	.430	.413	0.360	0.415	$x_1 = 0.250$

It pursues at $\bar{t}$? No. $\frac{250}{15.5} \times \frac{145}{223}$

$0.016 \times \frac{145}{223} = 0.010$ /hr

The End

342 continued

Wednesday Dec 26 3:45 pm heated up to 37° (see 342A) and incubated from $\bar{t} = 14$ hours to 7 hours. — Purpose is to see if final point on graph of 342. —

Thursday Dec 27 (at 37°) Overnight = 6.66 hr

10:30 am 11:20 am Bact + super via F net 350 = 0.137 280 = 0.900

Super via F

350								
0.018	300	290	780	720	280	350	240	$x_1 = .649$
	.074	.400	.668	.660	.520	350	.296	
			-17			-21		
			.651			329		

$.649 / 6.6 = 0.0975$

For

Fox Breeder Sat. Dec 29/51

Inoculated flask on shaker in 7
at 305 pm. with density 0.059 vs F.—

at 355 pm Beckm. 350: 0.097 vs F.
4 30 pm " " 0.130 " + reduced amount (to 1/2 cc)
4 45 pm " " 0.151 "
5 15 pm " " 0.198
5 35 pm " " 0.228
5 50 pm " " 0.251

Gots B/lt

Chemostat 343. Lee isolated B/lt from Gots wild type B
F + 500 ϒ/l trp V = 24 cc
Inoculated Wednesday Dec 26. Ran at r 6 hr
Friday Dec 28 τ = 5.85 hr
Collected 11^{15} am – 12^{15} in ice
Supernat vs F

350	300	290	280	270	260	250	240	
016	060	095	141	167	170	187	262	$X_1 = .072$

Collected 12^{15} — 2^{30}
Bact + Super vs F 350 = 0.172 280 = 0.480
Supernat

350	300	290	280	270	260	250	240
015	059	.094	.142	.168	.172	.191	.267

switched from r to r/min at 6 pm

---

343 Sat. Dec 29/57 T elevat 11 hrs
3 pm pushed ant (in ice)
350 = .184 ; 280 = .543

Beckmann calibration

undiluted	0.459
1+4 (F)	0.117
2+3	0.226
3+2	0.301

---

100 mg/l Histidine in F vs F

350	300	290	280	270	260	250	240	230	228
0.007	0.020	0.025	0.025	0.037	0.045	0.066	0.080	.375	.600

226.
.870.

Beckmann calibration [number to the right]

Sample dil in F vs F at 350

1B+9F = 0.048 (0.038)

4B+6F = 0.193 (0.175)

⇒ B+3F = 0.327

2B+8F = 0.100 (0.082)

5B+5F = 0.240 (220)

8B+2F = 370

3B+7F = 0.145 (0.127)

6B+4F = 0.285

9B+1F = 0.410

10B+0F = 0.440

F vs H₂O = 0.018

Isopropanol vs. F =

New Calibration of Nephelometer

Steel Standard 60 Neph (75-4)x2

Beckmann at 350 136 x 1

or 136 Beckmann gives 142 in Neph

Graph vs. H₂O

700	600	500	450	400	350	300
0.900	0.061	0.115	.204	.405	.800	1.86

625 575 550 525
.140 .052 .084 0.0[illegible]

to comp with Beckmann vs H₂O

at 350 = 0.190 at 575 = 0.100

Calibration of Nephelometer

Stand. 43.5

7cc in No I tube 1:20 dil of a suspension [56] in Neph.

1:5 dilution in Beckman vs H$_2$O - H

at 350 = 0.155

$\frac{-2}{0.153}$

0.153 corresponds to Neph 56 x 4

$\frac{0.153}{4} = 0.038$ corresponds to 56

100 mg/l Phenylalanine ninh

350	340	290	280	270	260	250	240	230
0.040	.005	.006	.007	.027	.112	096	040	020

20 mg/l Anthranilic ninh vs H$_2$O

400	350	340	330	320	310	300	290	280	270	260
0.002	0.140	0.225	0.307	0.357	0.354	.285	0.190	.123	.101	.195

250	240	230
0.660	0.420	

20 mg/l anthranilic ninh vs H$_2$O

400	350	340	330	320	310	300	290	280	270	260
0.009	0.071	0.158	0.282	.410	.472	.435	.320	.215	.160	.295

250	240	230
0.880		

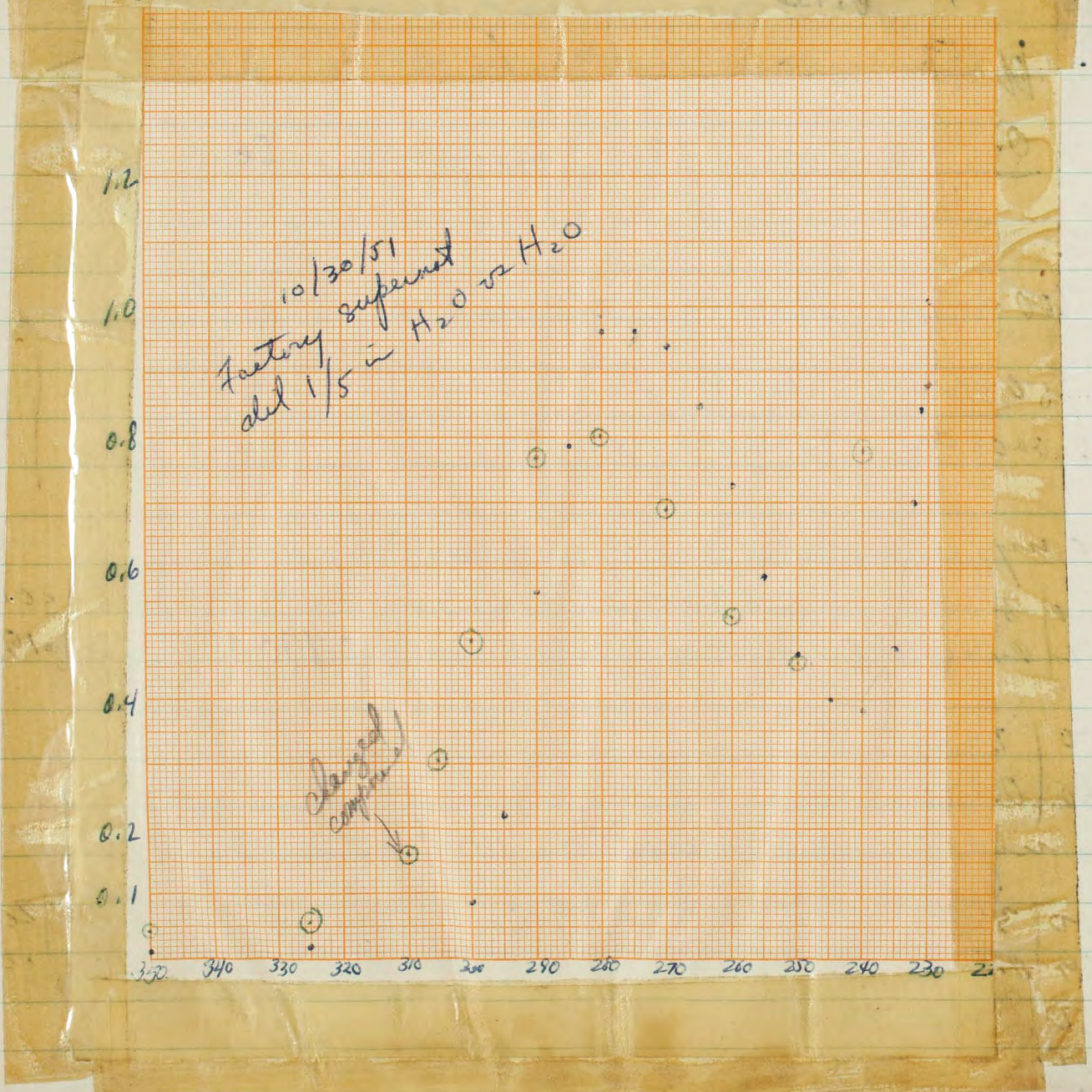
F — NH_4Cl

2 1/2 % broth in [illegible] vs H_2O

350	300	290	280	270	260	250	240	230
0.037	0.095	0.140	0.217	0.294	0.388	0.539	0.755	1.45

Bubble 31 mμ/l H_2O vs H_2O

350	340	330	320	310	300	290	280	270	260	250	240	230
0.008	0.052	0.600	1.550	1.690	1.480	0.970	0.650	0.940				



F vs H₂O

350	300	290	280	270	260	250	240	230
0.013	0.035	0.045	0.052	0.060	0.075	0.102	0.164	0.607

Glycine

Phenylalanine 19 mg/l vs H₂O
in H₂O

350	300	290	280	270	260	250	240	230
0.01	0.012	0.013	0.015	0.020	0.035	0.021	0.004	0.120

Compound X } of 17 mgm/l vs Phosphate
in Phosphate buffer PH 8.3 } dil into H₂O 50x Nov 2/57 (Original conc = 0.85 mg/ml) buffer

350	300	290	280	270	260	250	240	230	220
0.017	0.105	0.169	0.175	0.143	0.083	0.054	0.141		

Correction 0.076 / .250

Ed containing X evaporated and then acetyl added until precip forms

A dissolved 1.12 mg in 2 ml of 1/15 N Phosphate buffer PH 7

0.57 mg/cc or 560 mg/l

B dissolved 1.05 mg in 5 cc 0.02 normal Base
20 mg/l of Compound Mix 1:10 in Buffer PH 7 vs H₂O

350	300	290	280	270	260	250	240	230	220
0.021	0.245	0.384	0.401	0.345	0.262	0.226	0.390	0.21	

325	310	305
0.030	0.080	0.153

$$\frac{dN}{dt} = W_1 + \frac{W_2}{V_2} + \alpha N$$

$$\frac{dN_2}{dt} = W_1 N_1 - \frac{N_2}{\tau_2} + \frac{N_2}{\tau^*} = 0$$

$$\frac{dN}{W_1 - \frac{1}{\tau_2} + \frac{N}{\tau_1}} = dt$$

$$\frac{N_2}{\tau^*} = \frac{N_2}{\tau_2} - W_1 N_1$$

$$N_2 = \frac{\tau^*}{\tau_2} - \frac{W_1}{\tau_2} N_1$$

$$N_2\left(\frac{1}{\tau^*} - \frac{1}{\tau_2}\right)$$

$$N_2\left(\frac{1}{\tau_2} - \frac{1}{\tau^*}\right) = W_1 N_1 \frac{\tau^*}{\tau_2} = \frac{N_2}{\frac{1}{\tau_2} - W_1 N_1}$$

$$\frac{N_2}{\tau^*} - \frac{N_2}{\tau_2} = -W_1 N_1$$

$$\frac{N_2}{\tau^*} = -W_1 N_1 + \frac{N_2}{\tau_2}$$

$$\frac{1}{\tau^*} = \frac{1}{N_2}\left[\frac{N_2}{\tau_2} - W_1 N_1\right]$$

$$\frac{1}{\tau^*} = \frac{1}{\tau_2} - W_1 \frac{N_1}{N_2}$$

$$W_1 a = W_2 c + \left(\frac{N_2}{V_2} - \frac{N_1}{V_2} \right) Q$$

~~$W_1 a = W_2 c + Q \frac{dN}{dt}$~~