AbGradCon 2018

1
00:00:00,260 --> 00:00:12,510
[Music]

2
00:00:16,330 --> 00:00:14,109
hi everyone

3
00:00:18,730 --> 00:00:16,340
yeah so I'm a postdoc in Jack's lab and

4
00:00:21,040 --> 00:00:18,740
one of the overall I'd say like the main

5
00:00:22,960 --> 00:00:21,050
overall goal of Jack's lab the

6
00:00:25,660 --> 00:00:22,970
overarching theme and we all want to

7
00:00:27,580 --> 00:00:25,670
happen is to build a minimal cell and

8
00:00:29,470 --> 00:00:27,590
that's what we call a protocell that's

9
00:00:31,810 --> 00:00:29,480
capable of growth division of

10
00:00:34,389 --> 00:00:31,820
replication and eventually selection and

11
00:00:36,040 --> 00:00:34,399
evolution and so most of the people in

12
00:00:37,299 --> 00:00:36,050
the lab actually organic chemists and

13
00:00:39,220 --> 00:00:37,309

they're very talented and they're trying

14
00:00:41,079 --> 00:00:39,230
to get the middle of that diagram

15
00:00:43,540 --> 00:00:41,089
they're happening so like genetic

16
00:00:46,360 --> 00:00:43,550
material and information replicating

17
00:00:48,279 --> 00:00:46,370
without the help help of enzymes but the

18
00:00:49,889 --> 00:00:48,289
other aspect is actually packaging this

19
00:00:52,479 --> 00:00:49,899
all up and Marcus gave a very nice

20
00:00:54,759 --> 00:00:52,489
introduction earlier of putting it all

21
00:00:57,549 --> 00:00:54,769
inside a compartment to resemble x' like

22
00:00:58,419 --> 00:00:57,559
life as we know it cellular life and so

23
00:01:01,419 --> 00:00:58,429
that's where I come in

24
00:01:03,910 --> 00:01:01,429
I worked on the membranes and I just

25
00:01:06,390 --> 00:01:03,920
like to add that in addition to having a

26
00:01:09,270 --> 00:01:06,400
compartment to sort of condense and

27
00:01:11,560 --> 00:01:09,280
compartmentalize important things like

28
00:01:13,780 --> 00:01:11,570
molecules that might want to react with

29
00:01:15,760 --> 00:01:13,790
each other it's also used to have useful

30
00:01:19,090 --> 00:01:15,770
to have a compartment to keep away

31
00:01:20,920 --> 00:01:19,100
things like parasites are safe you're an

32
00:01:24,670 --> 00:01:20,930
RNA molecule that's evolved the ability

33
00:01:26,440 --> 00:01:24,680
to replicate other RNA molecules you

34
00:01:28,270 --> 00:01:26,450
want to maximize your Fitness by only

35
00:01:30,730 --> 00:01:28,280
replicating yourself and so having that

36
00:01:32,500 --> 00:01:30,740
membrane there stops parasites from

37
00:01:34,900 --> 00:01:32,510
coming in and are compromising your

38
00:01:37,270 --> 00:01:34,910

fitness and also having a selectively

39
00:01:39,880 --> 00:01:37,280
permeable barrier is nice because you

40
00:01:41,800 --> 00:01:39,890
want to be able to you know flirt around

41
00:01:44,530 --> 00:01:41,810
and your pond or whatever and take up

42
00:01:49,510 --> 00:01:44,540
nutrients but also not just take up

43
00:01:53,140 --> 00:01:49,520
anything great so this picture appeared

44
00:01:56,800 --> 00:01:53,150
earlier so basically life as we know it

45
00:01:59,800 --> 00:01:56,810
is cellular life and most organisms have

46
00:02:01,870 --> 00:01:59,810
cells that are comprised mostly of

47
00:02:04,080 --> 00:02:01,880
phospholipids so these are and

48
00:02:06,160 --> 00:02:04,090
fulfilling molecules that consists of a

49
00:02:08,499 --> 00:02:06,170
hydrophilic head group so have some

50
00:02:10,690 --> 00:02:08,509
charges there that's just a picture and

51
00:02:13,479 --> 00:02:10,700
then you see the two tails that are

52
00:02:15,760 --> 00:02:13,489
going out a very fatty chains and these

53
00:02:19,559 --> 00:02:15,770
things actually don't like being in

54
00:02:21,239 --> 00:02:19,569
water and their fatty acids and so what

55
00:02:22,709 --> 00:02:21,249
happens when you throw this in water is

56
00:02:25,289 --> 00:02:22,719
that these molecules spontaneously

57
00:02:26,910 --> 00:02:25,299
assemble into a bilayer and this

58
00:02:29,099 --> 00:02:26,920
maximizes the entropy of the system

59
00:02:30,929 --> 00:02:29,109
because the water molecules that would

60
00:02:33,690 --> 00:02:30,939
otherwise have to awkwardly crowd around

61
00:02:36,119 --> 00:02:33,700
the fatty acid tails become liberated to

62
00:02:37,830 --> 00:02:36,129
explore space when all the fatty acid

63
00:02:40,050 --> 00:02:37,840

tails are actually nestled with each

64
00:02:41,970 --> 00:02:40,060
other and so this is cool it's a

65
00:02:44,250 --> 00:02:41,980
spontaneous process you can buy like

66
00:02:45,750 --> 00:02:44,260
phospholipids from it's kind of

67
00:02:46,920 --> 00:02:45,760
expensive so maybe your live combines

68
00:02:49,020 --> 00:02:46,930
with phospholipids and then you

69
00:02:52,619 --> 00:02:49,030
basically like throw it into water and

70
00:02:55,379 --> 00:02:52,629
you see membranous structures form

71
00:02:57,119 --> 00:02:55,389
luckily for us there are much simpler

72
00:03:00,270 --> 00:02:57,129
molecules that can also self assemble

73
00:03:03,420 --> 00:03:00,280
into membranes if you want to make

74
00:03:05,670 --> 00:03:03,430
phospholipids and a prebiotic a possible

75
00:03:07,860 --> 00:03:05,680
manner you can give chemists it's a lot

76
00:03:11,670 --> 00:03:07,870
of headaches I think but fatty acids are

77
00:03:13,470 --> 00:03:11,680
basically soaps and what happens when we

78
00:03:16,800 --> 00:03:13,480
use the most soap is that we're using

79
00:03:18,479 --> 00:03:16,810
that quite quite a high pH although the

80
00:03:19,920 --> 00:03:18,489
carboxylic acid head groups are

81
00:03:21,929 --> 00:03:19,930
deprotonated so there's a lot of

82
00:03:23,789 --> 00:03:21,939
negative charge and those charges repel

83
00:03:25,830 --> 00:03:23,799
each other and they want to take up alot

84
00:03:28,110 --> 00:03:25,840
of space and they form these really sort

85
00:03:30,629 --> 00:03:28,120
of high curvature structures that we

86
00:03:33,869 --> 00:03:30,639
referred to as micelles and that's what

87
00:03:36,119 --> 00:03:33,879
we used a soap every day when you buy

88
00:03:38,909 --> 00:03:36,129

these from a company they actually come

89
00:03:41,129 --> 00:03:38,919
as a neat oil so if you drop the pH way

90
00:03:43,439 --> 00:03:41,139
down so that all of the carboxylic acid

91
00:03:45,030 --> 00:03:43,449
head groups actually protonated then all

92
00:03:46,649 --> 00:03:45,040
the negative charges are gone nothing's

93
00:03:49,229 --> 00:03:46,659
repelling anything and actually sit

94
00:03:51,659 --> 00:03:49,239
around as neat oil and you can I pet it

95
00:03:53,520 --> 00:03:51,669
it's a little bit viscous but you know

96
00:03:55,439 --> 00:03:53,530
pretty much oil and then the magic

97
00:03:57,839 --> 00:03:55,449
happens at this sort of like a middle pH

98
00:03:59,819 --> 00:03:57,849
the Goldilocks sort of pH and you have

99
00:04:01,530 --> 00:03:59,829
some head groups that are deprotonated

100
00:04:03,449 --> 00:04:01,540
and some that are protonated and these

101
00:04:07,349 --> 00:04:03,459
things can actually hydrogen bond and

102
00:04:10,110 --> 00:04:07,359
almost make this sort of two-tailed sort

103
00:04:12,360 --> 00:04:10,120
of fake phospholipid looking structure

104
00:04:13,649 --> 00:04:12,370
like I showed you before but there are

105
00:04:15,360 --> 00:04:13,659
no real bonds it's I mean the no

106
00:04:18,749 --> 00:04:15,370
covalent bonds they just sort of paired

107
00:04:21,060 --> 00:04:18,759
up weakly and um you can form Philo's

108
00:04:22,770 --> 00:04:21,070
just cool and the reason why were quite

109
00:04:24,749 --> 00:04:22,780
interested in this other than fatty

110
00:04:27,390 --> 00:04:24,759
acids being relatively cheap because we

111
00:04:29,550 --> 00:04:27,400
use it as soap is that they're they're

112
00:04:31,740 --> 00:04:29,560
quite our prebiotic lee plausible roots

113
00:04:33,330 --> 00:04:31,750

just synthesizing fatty acids and if you

114
00:04:35,790 --> 00:04:33,340
look at the organic material

115
00:04:38,040 --> 00:04:35,800
on carbonaceous chondrites fatty acids

116
00:04:41,520 --> 00:04:38,050
actually comprised I think the largest

117
00:04:44,570 --> 00:04:41,530
sort of fractional that most most well a

118
00:04:47,370 --> 00:04:44,580
lot of the molecules of fatty acids

119
00:04:49,020 --> 00:04:47,380
great so then like I said earlier you

120
00:04:50,550 --> 00:04:49,030
can throw them into water and they will

121
00:04:53,250 --> 00:04:50,560
just spontaneously assemble into

122
00:04:55,320 --> 00:04:53,260
membranes and what it might look like on

123
00:04:57,510 --> 00:04:55,330
the is what I'm showing on the left here

124
00:04:59,879 --> 00:04:57,520
and we have some structures that are

125
00:05:01,920 --> 00:04:59,889
quite dark and visible and these are

126
00:05:04,320 --> 00:05:01,930
multilamellar vesicles that means they

127
00:05:05,940 --> 00:05:04,330
have lots and lots of membrane layers

128
00:05:07,770 --> 00:05:05,950
kind of stacked up like an onion ring

129
00:05:11,520 --> 00:05:07,780
and they're pretty easy to see under a

130
00:05:13,590 --> 00:05:11,530
microscope you can also get vesicles

131
00:05:16,350 --> 00:05:13,600
that just have one sort of cell membrane

132
00:05:19,200 --> 00:05:16,360
or not cell 1 membrane and we refer to

133
00:05:21,240 --> 00:05:19,210
these as uni lamella vesicle ok

134
00:05:23,159 --> 00:05:21,250
you also get things to a tubular and

135
00:05:26,129 --> 00:05:23,169
there are things for the spherical and

136
00:05:28,950 --> 00:05:26,139
all sorts of things it's quite a zoo and

137
00:05:31,080 --> 00:05:28,960
we can also die the membranes red in

138
00:05:32,760 --> 00:05:31,090

this case and then stop some RNA inside

139
00:05:35,670 --> 00:05:32,770
and the RNA doesn't leak out the RNA

140
00:05:37,620 --> 00:05:35,680
screen and this is cool people in lab

141
00:05:39,930 --> 00:05:37,630
and other labs have done all sorts of

142
00:05:44,670 --> 00:05:39,940
really good experiments using this

143
00:05:46,290 --> 00:05:44,680
system but basically uh I'm a physicist

144
00:05:48,240 --> 00:05:46,300
by training so when I look at this I

145
00:05:51,210 --> 00:05:48,250
just think it's too messy

146
00:05:52,770 --> 00:05:51,220
this is what cells look like they just

147
00:05:54,960 --> 00:05:52,780
have that one membrane on the outside

148
00:05:58,350 --> 00:05:54,970
usually quite well separated from any

149
00:06:03,240 --> 00:05:58,360
other membranes and one way of referring

150
00:06:06,270 --> 00:06:03,250
to this sort of membrane topology is as

151
00:06:08,040 --> 00:06:06,280
a giant uni lamellar vesicle so then the

152
00:06:09,170 --> 00:06:08,050
giant thing refers to than being so big

153
00:06:11,520 --> 00:06:09,180
that you can see them under a microscope

154
00:06:13,800 --> 00:06:11,530
the Uni lamella because there's just

155
00:06:16,110 --> 00:06:13,810
that one membrane and I'm calling this

156
00:06:17,969 --> 00:06:16,120
yeast cell a vesicle because I look on

157
00:06:19,740 --> 00:06:17,979
vesicles but um that's essentially what

158
00:06:23,190 --> 00:06:19,750
they are they're just a fluid sac that's

159
00:06:24,390 --> 00:06:23,200
sitting around um that makes beer and

160
00:06:28,500 --> 00:06:24,400
stuff anyway

161
00:06:31,020 --> 00:06:28,510
pretty useful so what is kind of crazy

162
00:06:33,779 --> 00:06:31,030
is that this is kind of maybe one of the

163
00:06:36,089 --> 00:06:33,789

sort of really big challenges and

164
00:06:38,490 --> 00:06:36,099
synthetic followed biology we want to be

165
00:06:40,560 --> 00:06:38,500
able to make these giant uni lamella

166
00:06:43,050 --> 00:06:40,570
vesicles to mimic cells to be able to

167
00:06:44,880 --> 00:06:43,060
try out synthetic biology systems even

168
00:06:47,309 --> 00:06:44,890
to use them for drug delivery so that

169
00:06:49,170 --> 00:06:47,319
when they fuse with a cell membrane you

170
00:06:51,809 --> 00:06:49,180
have other extra little bits floating

171
00:06:54,359 --> 00:06:51,819
around and in order to be able to make

172
00:06:56,579 --> 00:06:54,369
these single uni lamellar membranes

173
00:06:58,230 --> 00:06:56,589
people have employed all sorts of

174
00:07:00,239 --> 00:06:58,240
different microfluidic techniques so you

175
00:07:03,179 --> 00:07:00,249
need a lot of sort of cleanroom

176
00:07:04,619 --> 00:07:03,189
experience and a lot of I don't know it

177
00:07:06,869 --> 00:07:04,629
can get quite frustrating but basically

178
00:07:09,089 --> 00:07:06,879
it's a technological challenge to make

179
00:07:12,089 --> 00:07:09,099
membranes that are you Neela Mela and

180
00:07:13,920 --> 00:07:12,099
this is nice because at some point I

181
00:07:16,290 --> 00:07:13,930
guess it's easy for cells now because

182
00:07:19,309 --> 00:07:16,300
cells beget cells but at some point if

183
00:07:23,279 --> 00:07:19,319
you rewind these unity lamellar single

184
00:07:25,019 --> 00:07:23,289
membrane forms became favored and so

185
00:07:26,399 --> 00:07:25,029
these primitive cells just did it like

186
00:07:29,909 --> 00:07:26,409
they didn't have micro fluidics or

187
00:07:32,100 --> 00:07:29,919
anything and so the thing I wanted to

188
00:07:34,439 --> 00:07:32,110

kind of understand was how did this sort

189
00:07:38,189 --> 00:07:34,449
of well separated or uni lamellar

190
00:07:40,439 --> 00:07:38,199
structure form become favored and then

191
00:07:43,049 --> 00:07:40,449
because I'm a physicist I'm not going to

192
00:07:44,999 --> 00:07:43,059
try and ask you know how did this maybe

193
00:07:47,010 --> 00:07:45,009
come about because of selective

194
00:07:48,809 --> 00:07:47,020
pressures instead I'm gonna ask maybe

195
00:07:51,570 --> 00:07:48,819
how did this come about on from

196
00:07:53,219 --> 00:07:51,580
thermodynamics and then that sort of

197
00:07:56,249 --> 00:07:53,229
changes the sort of question you can ask

198
00:07:58,439 --> 00:07:56,259
so instead I would say what I'm trying

199
00:07:59,489 --> 00:07:58,449
to understand is how can we get these

200
00:08:01,860 --> 00:07:59,499
things to self-assemble

201
00:08:04,019 --> 00:08:01,870
without much you know that means no

202
00:08:06,449 --> 00:08:04,029
intervention right tools are no enzymes

203
00:08:09,329 --> 00:08:06,459
so self-assembly not do-it-yourself

204
00:08:11,100 --> 00:08:09,339
assembly which is like you know you're

205
00:08:15,659 --> 00:08:11,110
forcing it into that structure with my

206
00:08:18,029 --> 00:08:15,669
footing so when I say no micro fluidics

207
00:08:19,320 --> 00:08:18,039
that's rather specific and I just mean

208
00:08:22,019 --> 00:08:19,330
like we don't want to go into a clean

209
00:08:23,339 --> 00:08:22,029
room because that's a lot of work but it

210
00:08:26,609 --> 00:08:23,349
doesn't mean we can't use other tools

211
00:08:28,199 --> 00:08:26,619
and so there aren't really many tools at

212
00:08:31,019 --> 00:08:28,209
our disposal in terms of you know

213
00:08:33,689 --> 00:08:31,029

prebiotic earth but we had rocks and

214
00:08:35,459 --> 00:08:33,699
early humans had rocks and this is a

215
00:08:36,779 --> 00:08:35,469
really tenuous line of reasoning but

216
00:08:39,360 --> 00:08:36,789
there was no more logic to it when I

217
00:08:42,509 --> 00:08:39,370
tried it in lab and because I had some

218
00:08:46,590 --> 00:08:42,519
rocks sitting around and I made vesicles

219
00:08:48,540 --> 00:08:46,600
as usual but I put some clay in and then

220
00:08:50,400 --> 00:08:48,550
what you see is that as soon as you add

221
00:08:52,620 --> 00:08:50,410
mineral particles the sorts of vesicles

222
00:08:54,809 --> 00:08:52,630
that you form are the sort of like high

223
00:08:56,939 --> 00:08:54,819
contrast low contrast sort of variety

224
00:08:59,400 --> 00:08:56,949
that you see on the left but you get

225
00:09:01,170 --> 00:08:59,410
these sort of giant rings forming

226
00:09:04,230 --> 00:09:01,180
instead

227
00:09:07,950 --> 00:09:04,240
and so that to me was very surprising I

228
00:09:10,440 --> 00:09:07,960
tried other sister zoomed-in and then I

229
00:09:12,180 --> 00:09:10,450
tried a different way of visualizing

230
00:09:13,920 --> 00:09:12,190
this and this is just you know you make

231
00:09:15,870 --> 00:09:13,930
some vesicles and then you add a little

232
00:09:18,090 --> 00:09:15,880
bit of fluorescent dye to the outside

233
00:09:19,650 --> 00:09:18,100
and the dye isn't able to cross the

234
00:09:21,300 --> 00:09:19,660
membrane so now we're able to see that

235
00:09:23,730 --> 00:09:21,310
in the control experiment when you just

236
00:09:26,940 --> 00:09:23,740
you know dump and fulfilling like don't

237
00:09:30,150 --> 00:09:26,950
fatty acids into buffer they make tiny

238
00:09:31,470 --> 00:09:30,160

vesicles then a lot of them are multi

239
00:09:32,610 --> 00:09:31,480
laid we can't really see that structure

240
00:09:34,860 --> 00:09:32,620
there but basically they're not

241
00:09:38,220 --> 00:09:34,870
encapsulating much of the of the liquid

242
00:09:40,620 --> 00:09:38,230
but as you add things like mineral

243
00:09:42,810 --> 00:09:40,630
particles then all of a sudden you're

244
00:09:44,460 --> 00:09:42,820
getting much much more of the volume be

245
00:09:48,030 --> 00:09:44,470
encapsulated those are the voids that

246
00:09:49,830 --> 00:09:48,040
you see actually um and so you see this

247
00:09:52,560 --> 00:09:49,840
sort of like cheese Swiss cheese like

248
00:09:54,330 --> 00:09:52,570
structure where you know this there's

249
00:09:55,740 --> 00:09:54,340
the same amount of lipid in the image

250
00:09:57,750 --> 00:09:55,750
that's on the left that's on the right

251
00:09:59,310 --> 00:09:57,760
but in the image on the left a lot of

252
00:10:01,140 --> 00:09:59,320
the lipid is wasted in making onion

253
00:10:02,850 --> 00:10:01,150
structures whereas on the right all of

254
00:10:04,950 --> 00:10:02,860
the lipid is being used to make single

255
00:10:07,650 --> 00:10:04,960
layered structures and so you take up a

256
00:10:09,390 --> 00:10:07,660
lot more space and so this is kind of

257
00:10:11,010 --> 00:10:09,400
weird like there's no reason this should

258
00:10:14,970 --> 00:10:11,020
have worked really there was no logic

259
00:10:16,800 --> 00:10:14,980
behind me trying it but then I think

260
00:10:18,690 --> 00:10:16,810
there might actually varies and that is

261
00:10:20,550 --> 00:10:18,700
that all of these are mineral particles

262
00:10:22,380 --> 00:10:20,560
sitting around in the organic chemistry

263
00:10:25,140 --> 00:10:22,390

lab are there for a reason they used for

264
00:10:27,450 --> 00:10:25,150
ion exchange or whatever basically

265
00:10:30,780 --> 00:10:27,460
cations can like bind to the surfaces of

266
00:10:33,960 --> 00:10:30,790
things like one Laurel and oak clay of

267
00:10:35,670 --> 00:10:33,970
silicates and they're usually used to

268
00:10:38,490 --> 00:10:35,680
sort of switch out what cation you're

269
00:10:40,050 --> 00:10:38,500
however whatever but maybe what's

270
00:10:41,820 --> 00:10:40,060
happening is by adding these mineral

271
00:10:44,130 --> 00:10:41,830
particles the cut ions are sticking to

272
00:10:46,320 --> 00:10:44,140
the cat clay instead of two mo vesicles

273
00:10:48,540 --> 00:10:46,330
and that's allowing the negative charges

274
00:10:49,020 --> 00:10:48,550
that are on the surface actually repel

275
00:10:51,540 --> 00:10:49,030
each other

276
00:10:54,620 --> 00:10:51,550
and so that's sort of just sort of

277
00:10:56,640 --> 00:10:54,630
putting the unis lamellar structure in

278
00:11:00,330 --> 00:10:56,650
but this peak it makes them more

279
00:11:03,210 --> 00:11:00,340
favourable ok so then this is the simple

280
00:11:06,270 --> 00:11:03,220
experiment this is just your vesicles

281
00:11:08,490 --> 00:11:06,280
with the usual buffer concentration 200

282
00:11:10,380 --> 00:11:08,500
milli molar almost everyone who works on

283
00:11:13,230 --> 00:11:10,390
fatty acid vesicles chooses to use this

284
00:11:14,850 --> 00:11:13,240
concentration it's kind of inherited but

285
00:11:18,300 --> 00:11:14,860
if you just

286
00:11:21,180 --> 00:11:18,310
even reduce the salt concentration by 25

287
00:11:22,530 --> 00:11:21,190
milli molar and I know this here is a

288
00:11:24,210 --> 00:11:22,540

different number but even if you reduce

289
00:11:25,890 --> 00:11:24,220
the salt concentration by a little bit

290
00:11:28,350 --> 00:11:25,900
then you start getting these really big

291
00:11:32,520 --> 00:11:28,360
vesicles with well separated membranes

292
00:11:33,690 --> 00:11:32,530
and you can encapsulate dye inside they

293
00:11:35,460 --> 00:11:33,700
don't leak a lot just having one

294
00:11:38,190 --> 00:11:35,470
membrane um doesn't compromise their

295
00:11:39,930 --> 00:11:38,200
sort of integrity okay so I've been

296
00:11:41,730 --> 00:11:39,940
claiming that these structures the Uni

297
00:11:44,760 --> 00:11:41,740
lamella for the last however many

298
00:11:47,160 --> 00:11:44,770
minutes so the final step is just to

299
00:11:49,230 --> 00:11:47,170
show you that they actually are and then

300
00:11:50,940 --> 00:11:49,240
because making uni lamellar vesicles is

301
00:11:52,680 --> 00:11:50,950
kind of a big deal in synthetic biology

302
00:11:54,960 --> 00:11:52,690
there's quite a well-developed technique

303
00:11:58,110 --> 00:11:54,970
that people use they throw some dye in

304
00:12:00,360 --> 00:11:58,120
the dyes like lipophilic and it goes

305
00:12:01,800 --> 00:12:00,370
into the membrane and the more membranes

306
00:12:04,950 --> 00:12:01,810
you have the brighter they look under

307
00:12:06,270 --> 00:12:04,960
fluorescent microscope and so on the

308
00:12:09,060 --> 00:12:06,280
sample on the right here is made with

309
00:12:10,650 --> 00:12:09,070
the high salt buffer conditions and you

310
00:12:12,510 --> 00:12:10,660
can see that in this one field of view

311
00:12:13,740 --> 00:12:12,520
there are multiple vesicles but there

312
00:12:16,500 --> 00:12:13,750
are different brightnesses and it's

313
00:12:19,020 --> 00:12:16,510

actually quite pretty and then if you

314
00:12:22,320 --> 00:12:19,030
make a low salt concentration sample

315
00:12:25,470 --> 00:12:22,330
then they all look kind of the same like

316
00:12:27,960 --> 00:12:25,480
intensity so we can write some code to

317
00:12:29,520 --> 00:12:27,970
kind of not just eyeball this but our

318
00:12:31,590 --> 00:12:29,530
quantified the average intensity of

319
00:12:32,880 --> 00:12:31,600
these rings and you can see for the low

320
00:12:35,190 --> 00:12:32,890
salt sample on the left you get this

321
00:12:37,500 --> 00:12:35,200
sort of single peak distribution and it

322
00:12:39,510 --> 00:12:37,510
lines up with like the the sort of demo

323
00:12:41,460 --> 00:12:39,520
strings that we find in the sort of

324
00:12:43,980 --> 00:12:41,470
sample on the right that's a bit Messier

325
00:12:45,810 --> 00:12:43,990
and so this is kind of pretty good

326
00:12:47,660 --> 00:12:45,820
evidence that we've found a way to make

327
00:12:50,280 --> 00:12:47,670
these sort of greeny lamellar structures

328
00:12:51,750 --> 00:12:50,290
literally without doing anything you mix

329
00:12:54,660 --> 00:12:51,760
it together you go to sleep you come

330
00:12:56,490 --> 00:12:54,670
back and you get the sample of that and

331
00:12:57,930 --> 00:12:56,500
this is sort of going towards the goal

332
00:13:00,240 --> 00:12:57,940
of making these membranous structures

333
00:13:04,860 --> 00:13:00,250
that resemble more sort of the life as

334
00:13:06,420 --> 00:13:04,870
we know it like Easter and I think the

335
00:13:08,280 --> 00:13:06,430
reason we're able to do this with fatty

336
00:13:09,600 --> 00:13:08,290
acids and you know most people in

337
00:13:11,760 --> 00:13:09,610
synthetic biology work with

338
00:13:13,560 --> 00:13:11,770

phospholipids it's because fatty acids

339
00:13:15,450 --> 00:13:13,570
are you know have some special property

340
00:13:17,160 --> 00:13:15,460
the first thing is that they do carry

341
00:13:18,690 --> 00:13:17,170
negative charges half of the molecules

342
00:13:20,610 --> 00:13:18,700
are negatively charged when you can make

343
00:13:22,560 --> 00:13:20,620
vesicle and that really helps the

344
00:13:24,900 --> 00:13:22,570
membranes repel each other and like have

345
00:13:26,910 --> 00:13:24,910
that separation and the second reason is

346
00:13:28,710 --> 00:13:26,920
that these molecules are small so they

347
00:13:30,749 --> 00:13:28,720
actually moved between vesicles whereas

348
00:13:32,939 --> 00:13:30,759
if you make a phospholipid vesicle the

349
00:13:35,040 --> 00:13:32,949
phospholipid R is actually kinetically

350
00:13:36,569 --> 00:13:35,050
trapped in its vesicle that it happens

351
00:13:38,129 --> 00:13:36,579
to be in they can't move to a

352
00:13:39,990 --> 00:13:38,139
neighboring one and so if you have a

353
00:13:44,509 --> 00:13:40,000
complex energy landscape you can't sort

354
00:13:47,129 --> 00:13:44,519
of remove anywhere yeah so basically

355
00:13:49,350 --> 00:13:47,139
these are simple amphiphilic molecules

356
00:13:51,569 --> 00:13:49,360
that may have been around on earlier

357
00:13:54,420 --> 00:13:51,579
that can form membranes they have their

358
00:13:55,769 --> 00:13:54,430
advantages and it can help us fight form

359
00:13:59,040 --> 00:13:55,779
these structures that we couldn't with

360
00:14:01,860 --> 00:13:59,050
phospholipids also that you know having

361
00:14:03,600 --> 00:14:01,870
low ionic strength buffers seems to help

362
00:14:04,499 --> 00:14:03,610
us form giant any lamellar vesicles

363
00:14:07,350 --> 00:14:04,509

which is cool

364
00:14:09,179 --> 00:14:07,360
and if you can't afford to use lower

365
00:14:10,559 --> 00:14:09,189
ionic strength buffers it seems like

366
00:14:12,389 --> 00:14:10,569
that you can just chuck some mineral

367
00:14:15,030 --> 00:14:12,399
particles in and that'll do the job okay

368
00:14:17,100 --> 00:14:15,040
so I'd like to thank everyone here for

369
00:14:19,019 --> 00:14:17,110
organizing this it's this space where

370
00:14:22,499 --> 00:14:19,029
you know we can all learn a lot with low

371
00:14:24,749 --> 00:14:22,509
pressure and I don't know this is being

372
00:14:28,410 --> 00:14:24,759
recorded hey nurse we're all very happy

373
00:14:29,429 --> 00:14:28,420
we're all very happy and I can all be

374
00:14:32,490 --> 00:14:29,439
very happy together

375
00:14:35,040 --> 00:14:32,500
no it's really fun um and and I'd like

376
00:14:38,610 --> 00:14:35,050
to thank my lab jack dining out loud

377
00:14:40,710 --> 00:14:38,620
manage is amazing and also to Chris Carr

378
00:14:42,240 --> 00:14:40,720
and Kendall sobota for the olivine that

379
00:14:44,819 --> 00:14:42,250
I used I was actually collected from the

380
00:14:54,240 --> 00:14:44,829
green sand beach in Hawaii pretty much

381
00:15:02,129 --> 00:14:54,250
thank you thank you Anna do you have any

382
00:15:05,369 --> 00:15:02,139
questions okay very fascinating work I

383
00:15:09,389 --> 00:15:05,379
would be curious if is any temperature

384
00:15:12,480 --> 00:15:09,399
dependence the phenomena that you of

385
00:15:15,569 --> 00:15:12,490
Europe inter experiment also the other

386
00:15:20,040 --> 00:15:15,579
thing for pantry application and I think

387
00:15:21,720 --> 00:15:20,050
that oleic acid is bonding

388
00:15:23,850 --> 00:15:21,730

I don't remember what would be the bond

389
00:15:27,090 --> 00:15:23,860
isn't the boric acid we met right but

390
00:15:30,259 --> 00:15:27,100
all we have we try some shorter fatty

391
00:15:35,220 --> 00:15:30,269
acids like the canary acid we do see

392
00:15:36,420 --> 00:15:35,230
some kinds of cell dumplings yeah I'll

393
00:15:38,009 --> 00:15:36,430
answer the second question

394
00:15:41,790 --> 00:15:38,019
that's cuz I already forgot the first

395
00:15:42,310 --> 00:15:41,800
one but yeah I did use our like asset

396
00:15:44,889 --> 00:15:42,320
every

397
00:15:46,509 --> 00:15:44,899
and it's probable its abundance is

398
00:15:48,850 --> 00:15:46,519
probably close to nothing on meteorites

399
00:15:50,079 --> 00:15:48,860
right so this is just like a cheap thing

400
00:15:52,990 --> 00:15:50,089
that's easy to work with room

401
00:15:55,389 --> 00:15:53,000
temperature in live I've tried sure to

402
00:15:57,280 --> 00:15:55,399
ensure their chance and so my sort of

403
00:16:00,819 --> 00:15:57,290
behavior where I see these giant uni

404
00:16:02,559 --> 00:16:00,829
lamella vesicles form the pH range at

405
00:16:05,139 --> 00:16:02,569
which it happens will change with the

406
00:16:08,170 --> 00:16:05,149
with the fatty acid chain length so for

407
00:16:11,970 --> 00:16:08,180
fourteen carbons I see them form at a

408
00:16:15,069 --> 00:16:11,980
lower pH the problem is when you have

409
00:16:18,519 --> 00:16:15,079
these giant vesicles that are about five

410
00:16:20,769 --> 00:16:18,529
micrometers in diameter so I'll

411
00:16:23,530 --> 00:16:20,779
backtrack a bit if you pack oranges into

412
00:16:25,449 --> 00:16:23,540
a box the maximum packing fraction you

413
00:16:27,850 --> 00:16:25,459

can get is like 60 something said I

414
00:16:30,220 --> 00:16:27,860
think and so if you calculate the amount

415
00:16:33,280 --> 00:16:30,230
of material of lipid that you need to

416
00:16:35,949 --> 00:16:33,290
fill a box like fill your sample it is

417
00:16:38,079 --> 00:16:35,959
actually only 5 mili molar and so that's

418
00:16:40,210 --> 00:16:38,089
below the critical aggregation

419
00:16:42,040 --> 00:16:40,220
concentration for these shorter chain

420
00:16:44,740 --> 00:16:42,050
fatty acids which is why I didn't use

421
00:16:47,199 --> 00:16:44,750
them if you do sort of use more lipid

422
00:16:49,059 --> 00:16:47,209
and instead of them just all forming big

423
00:16:51,370 --> 00:16:49,069
vesicles and getting stuck they start

424
00:16:54,460 --> 00:16:51,380
forming like vesicles inside vesicles

425
00:16:56,650 --> 00:16:54,470
and so I mean that's a cool behavior in

426
00:16:58,689 --> 00:16:56,660
and of itself but um yeah I think it'd

427
00:17:01,809 --> 00:16:58,699
be nice to go to a like shorter chain

428
00:17:04,140 --> 00:17:01,819
system like c10 and then just see if we

429
00:17:12,819 --> 00:17:04,150
can make the well separated membranes

430
00:17:12,829 --> 00:17:22,830
one

431
00:17:28,030 --> 00:17:25,120
all right let's talk I have you have

432
00:17:31,540 --> 00:17:28,040
tried unbuffered system like bison is

433
00:17:33,400 --> 00:17:31,550
not very biotic and the pika what it

434
00:17:35,050 --> 00:17:33,410
seems to happen that the giant vesicles

435
00:17:37,330 --> 00:17:35,060
are formed around the pKa of the fatty

436
00:17:40,030 --> 00:17:37,340
acid so if you use the fact that it does

437
00:17:42,040 --> 00:17:40,040
a buffer have you tried this system no I

438
00:17:44,980 --> 00:17:42,050

haven't the thing that I tried that was

439
00:17:46,030 --> 00:17:44,990
the most sort of the simplest system I

440
00:17:48,130 --> 00:17:46,040
tried was no buffer

441
00:17:49,870 --> 00:17:48,140
I'm just hard to chloric acid and sodium

442
00:17:51,790 --> 00:17:49,880
chloride and the same thing happens it

443
00:17:54,520 --> 00:17:51,800
just it's just annoying to like titrate

444
00:18:00,010 --> 00:17:54,530
so I just stop do you think I used

445
00:18:02,680 --> 00:18:00,020
buffer instead yeah thanks for the great

446
00:18:04,630 --> 00:18:02,690
talk I was just wondering if there was

447
00:18:08,440 --> 00:18:04,640
any significant difference in stability

448
00:18:12,270 --> 00:18:08,450
of the vesicles between the different

449
00:18:15,250 --> 00:18:12,280
concentrations or the addition of notes

450
00:18:17,980 --> 00:18:15,260
yeah so I didn't actually check on their

451
00:18:19,630 --> 00:18:17,990
permeabilities too like all again weakly

452
00:18:25,930 --> 00:18:19,640
tides off anything

453
00:18:28,480 --> 00:18:25,940
oh you mean oh they're very stable like

454
00:18:31,180 --> 00:18:28,490
they sit around for like a month and I'm

455
00:18:32,860 --> 00:18:31,190
pretty sure the fatty acid I use like as

456
00:18:35,400 --> 00:18:32,870
oxidized and they still look the same

457
00:18:40,620 --> 00:18:35,410
under the microscope I don't know why

458
00:18:44,200 --> 00:18:40,630
yeah okay one more

459
00:18:46,120 --> 00:18:44,210
I'll go everyone else great talk so

460
00:18:48,880 --> 00:18:46,130
early on you showed where you guys put

461
00:18:51,220 --> 00:18:48,890
RNA inside one of these double membrane

462
00:18:53,280 --> 00:18:51,230
and things could you do the same well

463
00:18:56,320 --> 00:18:53,290

first of all it was there any kind of

464
00:18:57,400 --> 00:18:56,330
activity with that or purpose or is it

465
00:18:58,720 --> 00:18:57,410
just kind of show that you could put him

466
00:19:02,770 --> 00:18:58,730
in there and then could you also do the

467
00:19:04,960 --> 00:19:02,780
same with the GU v's yeah so I've tried

468
00:19:07,120 --> 00:19:04,970
encapsulation experiments with these TVs

469
00:19:11,770 --> 00:19:07,130
on with the the low salt concentration

470
00:19:14,290 --> 00:19:11,780
everything and if I the largest thing

471
00:19:16,510 --> 00:19:14,300
I've stuffed inside a 400 nanometer

472
00:19:17,800 --> 00:19:16,520
large polystyrene particle and so

473
00:19:19,660 --> 00:19:17,810
there's no problem getting nano

474
00:19:21,300 --> 00:19:19,670
particles inside or like colloidal

475
00:19:24,310 --> 00:19:21,310
particles inside as long as they're

476
00:19:25,780 --> 00:19:24,320
diffusing they'll go in so there's like

477
00:19:27,280 --> 00:19:25,790
huge defects when these things are

478
00:19:28,870 --> 00:19:27,290
forming right so I don't really know

479
00:19:31,060 --> 00:19:28,880
what the encapsulation mechanism is but

480
00:19:32,620 --> 00:19:31,070
you can stuff RNA inside what people

481
00:19:34,840 --> 00:19:32,630
normally do in love when they put RNA

482
00:19:36,160 --> 00:19:34,850
inside is do primary extension

483
00:19:38,050 --> 00:19:36,170
experiment so they're trying to see can

484
00:19:40,570 --> 00:19:38,060
we elongate a primer non enzymatically

485
00:19:43,090 --> 00:19:40,580
and then they'll usually crack these

486
00:19:45,600 --> 00:19:43,100
vesicle vesicles open and then run the

487
00:19:49,420 --> 00:19:45,610
results in a gel to see if it worked

488
00:19:51,970 --> 00:19:49,430

yeah so I think as we move towards

489
00:19:54,810 --> 00:19:51,980
developing like optical Reed sort of

490
00:19:57,460 --> 00:19:54,820
feedback for successful primer extension

491
00:19:59,020 --> 00:19:57,470
the G V's will be more useful for that

492
00:20:00,490 --> 00:19:59,030
purpose because then we can like look at

493
00:20:03,880 --> 00:20:00,500
something big under a microscope right

494
00:20:06,340 --> 00:20:03,890
off that'll be pretty cool okay thank