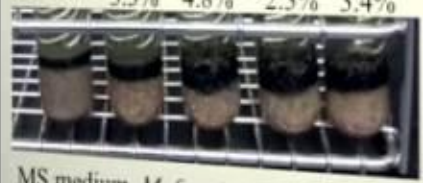
Results

Set 2 Transfer, 5 g sand, 10 mL

MM medium, *M. wolfeii*

1.7% 0.01%

3.3% 4.8% 2.3% 3.4%



MS medium, *M. formicicum*

1
00:00:12,619 --> 00:00:09,980
so why methanogens first off methanogens

2
00:00:14,990 --> 00:00:12,629
can be ideal candidates for life on Mars

3
00:00:17,840 --> 00:00:15,000
as well as other bodies in the solar

4
00:00:21,170 --> 00:00:17,850
system the four strains we used in our

5
00:00:23,650 --> 00:00:21,180
lab are listed down here and like Lucy

6
00:00:27,890 --> 00:00:23,660
said earlier all of our methanogens

7
00:00:30,290 --> 00:00:27,900
utilize only carbon dioxide as a carbon

8
00:00:33,050 --> 00:00:30,300
source and hydrogen as an energy source

9
00:00:35,060 --> 00:00:33,060
and so they're anaerobic which would be

10
00:00:37,010 --> 00:00:35,070
nice and Mars also their

11
00:00:38,660 --> 00:00:37,020
non-photosynthetic so that means they

12
00:00:42,710 --> 00:00:38,670
could live under the subsurface and

13
00:00:47,090 --> 00:00:42,720

finally methanogens were arose early on

14
00:00:50,060 --> 00:00:47,100
earth and so if Mars had a similar early

15
00:00:52,670 --> 00:00:50,070
history perhaps they happen to arise on

16
00:00:54,890 --> 00:00:52,680
Mars as well so these are the four

17
00:00:57,080 --> 00:00:54,900
strains we use in our lab but in this

18
00:00:59,120 --> 00:00:57,090
experiment we only used meth a no

19
00:01:02,870 --> 00:00:59,130
thermal vector wolfie I and meth a no

20
00:01:05,499 --> 00:01:02,880
bacterium for me cecum so we did a

21
00:01:08,690 --> 00:01:05,509
freeze thaw experiment and we wanted to

22
00:01:10,670 --> 00:01:08,700
approach mimicking Mars temperature

23
00:01:13,220 --> 00:01:10,680
conditions and so these are just two

24
00:01:16,730 --> 00:01:13,230
charts from curiosity at Gale Crater and

25
00:01:19,160 --> 00:01:16,740
the first the top chart shows daily

26
00:01:22,580 --> 00:01:19,170
temperature minimum and maximum in the

27
00:01:25,370 --> 00:01:22,590
air from late winter too late spring or

28
00:01:28,430 --> 00:01:25,380
about one hat one fourth of the Martian

29
00:01:29,990 --> 00:01:28,440
air and so you can see that at late

30
00:01:32,690 --> 00:01:30,000
winter temperatures don't really get

31
00:01:35,180 --> 00:01:32,700
above zero degrees and they go down to

32
00:01:37,880 --> 00:01:35,190
about negative 80 Celsius whereas in the

33
00:01:41,930 --> 00:01:37,890
late spring temperatures can rise a tiny

34
00:01:44,120 --> 00:01:41,940
bit above zero and there's not as much

35
00:01:48,890 --> 00:01:44,130
variation there and so we use this data

36
00:01:50,990 --> 00:01:48,900
for our initial experiments yesterday's

37
00:01:53,300 --> 00:01:51,000
talk by Brandon Stackhouse write a good

38
00:01:57,830 --> 00:01:53,310

introduction to thaw features on Mars

39
00:02:00,710 --> 00:01:57,840
and so this is just background for our

40
00:02:04,010 --> 00:02:00,720
experiment that we do see evidence of

41
00:02:09,350 --> 00:02:04,020
what could be thathi such as skeleton

42
00:02:11,089 --> 00:02:09,360
rain pingos and polygons so the

43
00:02:13,670 --> 00:02:11,099
questions we wanted to ask in our

44
00:02:16,220 --> 00:02:13,680
experiment are can limit antigens that

45
00:02:18,690 --> 00:02:16,230
we work with survive these freeze-thaw

46
00:02:21,330 --> 00:02:18,700
cycles or does the stress

47
00:02:23,369 --> 00:02:21,340
happen to kill them do they go dormant

48
00:02:26,729 --> 00:02:23,379
stuff and be like something like that

49
00:02:29,400 --> 00:02:26,739
and so we wanted to oopsy initially look

50
00:02:31,589 --> 00:02:29,410
at just a variation in min and max

51
00:02:35,070 --> 00:02:31,599
temperature of 20 degrees and ultimately

52
00:02:36,780 --> 00:02:35,080
get up to 80 degrees next we want to

53
00:02:39,360 --> 00:02:36,790
know if our methanogens can actively

54
00:02:40,949 --> 00:02:39,370
grow during these freeze-thaw cycles if

55
00:02:44,699 --> 00:02:40,959
the temperature does get above freezing

56
00:02:47,039 --> 00:02:44,709
can they take that opportunity to grow

57
00:02:49,680 --> 00:02:47,049
and finally we wanted to look at the

58
00:02:52,800 --> 00:02:49,690
porosity of the soil or the regolith we

59
00:02:56,430 --> 00:02:52,810
use in the experiments to see if greater

60
00:03:00,569 --> 00:02:56,440
pore size can provide some sort of safe

61
00:03:03,180 --> 00:03:00,579
haven so we conducted four experiments

62
00:03:05,250 --> 00:03:03,190
and these are four different sets in our

63
00:03:07,830 --> 00:03:05,260

first set we looked at porosity where we

64
00:03:10,800 --> 00:03:07,840
used just one of our organisms for me

65
00:03:13,319 --> 00:03:10,810
see come in a standard anaerobic medium

66
00:03:15,420 --> 00:03:13,329
that we grow our methanogens in and we

67
00:03:17,640 --> 00:03:15,430
had two subsets where we had a mixture

68
00:03:20,190 --> 00:03:17,650
of sand and gravel and then just stand

69
00:03:23,250 --> 00:03:20,200
and we had ten military ten milliliters

70
00:03:25,830 --> 00:03:23,260
of medium in there four sets two three

71
00:03:28,770 --> 00:03:25,840
and four we were basically looking at

72
00:03:31,650 --> 00:03:28,780
how the amount of liquid medium and the

73
00:03:33,930 --> 00:03:31,660
amount of dry regolith affected the

74
00:03:36,300 --> 00:03:33,940
survival or growth of our organisms and

75
00:03:38,039 --> 00:03:36,310
so we used both for me see command mole

76
00:03:40,500 --> 00:03:38,049
feei in each of these experiments and

77
00:03:44,220 --> 00:03:40,510
then different amounts of sand and

78
00:03:46,920 --> 00:03:44,230
liquid medium and so set to has five

79
00:03:48,990 --> 00:03:46,930
grams and ten millliliters set three has

80
00:03:51,509 --> 00:03:49,000
10 grams of sand five milliliters of

81
00:03:55,110 --> 00:03:51,519
medium and set for was our saturation

82
00:03:58,259 --> 00:03:55,120
experiment where we only provided enough

83
00:04:02,280 --> 00:03:58,269
medium to saturate just above the soil

84
00:04:05,900 --> 00:04:02,290
layer so here are some of our results

85
00:04:08,339 --> 00:04:05,910
first I just want to explain what we

86
00:04:11,879 --> 00:04:08,349
would see and then I'll explain what we

87
00:04:14,250 --> 00:04:11,889
did see so here on the bottom is the

88
00:04:15,839 --> 00:04:14,260

length of the experiment in days in

89
00:04:18,000 --> 00:04:15,849
order to measure growth of methanogens

90
00:04:21,960 --> 00:04:18,010
since they're anaerobic the easiest way

91
00:04:23,850 --> 00:04:21,970
is to measure the % methane produced in

92
00:04:28,190 --> 00:04:23,860
the headspace of the test tube using a

93
00:04:31,560 --> 00:04:28,200
gas chromatograph and so when our

94
00:04:32,740 --> 00:04:31,570
organisms are producing methane that

95
00:04:37,080 --> 00:04:32,750
basically means that there

96
00:04:40,270 --> 00:04:37,090
growing and so if we did not take any

97
00:04:41,890 --> 00:04:40,280
remove any methane to test remove any

98
00:04:44,920 --> 00:04:41,900
air from the test tube to test it for

99
00:04:47,200 --> 00:04:44,930
methane then this methane would be

100
00:04:48,730 --> 00:04:47,210
constant throughout the whole trial

101
00:04:51,100 --> 00:04:48,740
because there's nowhere for that methane

102
00:04:53,440 --> 00:04:51,110
to go but when we do take measurements

103
00:04:55,540 --> 00:04:53,450
such as which all these data points are

104
00:04:57,220 --> 00:04:55,550
then the methane decreases because we're

105
00:05:01,960 --> 00:04:57,230
removing that methane from the test tube

106
00:05:04,570 --> 00:05:01,970
and so that this decrease here is due to

107
00:05:08,610 --> 00:05:04,580
sampling of the test tubes and I just

108
00:05:11,970 --> 00:05:08,620
want to explain that so um in this graph

109
00:05:14,380 --> 00:05:11,980
you can see here that these are our

110
00:05:16,980 --> 00:05:14,390
different cycles of freeze and thaw I

111
00:05:19,180 --> 00:05:16,990
guess first we initially grew our

112
00:05:22,450 --> 00:05:19,190
organism at their growth temperature

113
00:05:24,910 --> 00:05:22,460

which is 37 degrees Celsius and then we

114
00:05:26,920 --> 00:05:24,920
subjected them to different cycles of 4

115
00:05:29,940 --> 00:05:26,930
degrees Celsius and the light blue is

116
00:05:33,880 --> 00:05:29,950
negative 15 degrees Celsius and so

117
00:05:37,540 --> 00:05:33,890
what's interesting about this set is

118
00:05:39,430 --> 00:05:37,550
that we do see an increase in methane

119
00:05:42,990 --> 00:05:39,440
which means that methane was produced

120
00:05:47,830 --> 00:05:43,000
because there's nowhere for that methane

121
00:05:50,170 --> 00:05:47,840
to go and then come back basically to be

122
00:05:52,480 --> 00:05:50,180
removed when we're taking a sample and

123
00:05:55,480 --> 00:05:52,490
the only way for it to be reproduced is

124
00:05:57,760 --> 00:05:55,490
for methanogens to produce it and so

125
00:05:59,440 --> 00:05:57,770
that's an interesting aspect and then

126
00:06:02,080 --> 00:05:59,450
there's just another one here we're

127
00:06:05,290 --> 00:06:02,090
removing air from the sample didn't

128
00:06:07,840 --> 00:06:05,300
decrease the methane and this again was

129
00:06:10,570 --> 00:06:07,850
set one where we had sand and sand and

130
00:06:14,770 --> 00:06:10,580
gravel and so it does seem that maybe

131
00:06:16,840 --> 00:06:14,780
porosity could play a role in providing

132
00:06:17,920 --> 00:06:16,850
a better growth environment and so this

133
00:06:22,270 --> 00:06:17,930
is just something we're going to look

134
00:06:23,980 --> 00:06:22,280
into further in set two we had five

135
00:06:25,540 --> 00:06:23,990
grams of sand and ten milliliters of

136
00:06:29,050 --> 00:06:25,550
medium and we used our two organisms

137
00:06:31,450 --> 00:06:29,060
wolffian for me see come and I just want

138
00:06:34,930 --> 00:06:31,460

to note that all of these graphs are on

139
00:06:37,950 --> 00:06:34,940
the same axis for comparison they're all

140
00:06:41,830 --> 00:06:37,960
they all go up to forty five percent

141
00:06:43,600 --> 00:06:41,840
methane but again you can see here that

142
00:06:45,719 --> 00:06:43,610
we put them at their initial growth

143
00:06:47,700 --> 00:06:45,729
temperatures which is 37 degrees for

144
00:06:49,890 --> 00:06:47,710
for me to come and 55 degrees Celsius

145
00:06:54,299 --> 00:06:49,900
for Wolfie I and then we subjected them

146
00:06:57,659 --> 00:06:54,309
to alternating growth cycles and so what

147
00:07:00,149 --> 00:06:57,669
we wanted to look at was what was

148
00:07:03,739 --> 00:07:00,159
happening here since we aren't seeing

149
00:07:06,269 --> 00:07:03,749
any real increase in % methane did our

150
00:07:08,790 --> 00:07:06,279
methanogens die are they going into

151
00:07:10,950 --> 00:07:08,800
dormancy are they still active and so I

152
00:07:14,429 --> 00:07:10,960
performed a transfer just to new medium

153
00:07:18,299 --> 00:07:14,439
and at room temperature and at least

154
00:07:21,779 --> 00:07:18,309
Wolfie I is still viable it rebounded

155
00:07:24,899 --> 00:07:21,789
and the organisms are able to again

156
00:07:27,649 --> 00:07:24,909
produce more methane and even with for

157
00:07:30,269 --> 00:07:27,659
me cecum some methane was produced and

158
00:07:32,639 --> 00:07:30,279
hopefully I plan on continuing these

159
00:07:36,510 --> 00:07:32,649
experiments and hopefully those numbers

160
00:07:38,129 --> 00:07:36,520
will go up in set three there was 10

161
00:07:40,679 --> 00:07:38,139
grams of sand and five milliliters of

162
00:07:42,629 --> 00:07:40,689
medium this basically just shows that

163
00:07:46,790 --> 00:07:42,639

the more liquid medium ultimately

164
00:07:49,649 --> 00:07:46,800
available provides greater growth and

165
00:07:53,459 --> 00:07:49,659
inset for which is just the saturation

166
00:07:55,860 --> 00:07:53,469
experiment we had initially put the

167
00:07:58,050 --> 00:07:55,870
organisms directly into four degrees C

168
00:07:59,610 --> 00:07:58,060
we didn't allow for that initial growth

169
00:08:03,059 --> 00:07:59,620
period at the growth temperature and

170
00:08:04,980 --> 00:08:03,069
they didn't grow so we put them then

171
00:08:08,699 --> 00:08:04,990
into their growth temperatures and they

172
00:08:12,570 --> 00:08:08,709
were still viable after being exposed to

173
00:08:15,809 --> 00:08:12,580
4 degrees Celsius and so they did grow

174
00:08:18,570 --> 00:08:15,819
and then were subjected to freeze thaw

175
00:08:21,149 --> 00:08:18,580
cycles again for at least wolfie I we

176
00:08:24,739 --> 00:08:21,159
did see an increase in methane which

177
00:08:26,969 --> 00:08:24,749
means that methane was being produced

178
00:08:31,110 --> 00:08:26,979
another interesting thing that we saw

179
00:08:34,709 --> 00:08:31,120
with our experiment is this black band

180
00:08:36,719 --> 00:08:34,719
within the first sometimes it's just

181
00:08:39,959 --> 00:08:36,729
below the surface or sometimes it's

182
00:08:42,149 --> 00:08:39,969
actually at the surface but we're not

183
00:08:44,250 --> 00:08:42,159
quite sure what it is right now it could

184
00:08:46,680 --> 00:08:44,260
either be an interaction with the sodium

185
00:08:49,590 --> 00:08:46,690
sulfide that we add to our medium with

186
00:08:51,540 --> 00:08:49,600
the soil or it could be some alteration

187
00:08:55,740 --> 00:08:51,550
of the environment by the microorganisms

188
00:08:59,060 --> 00:08:55,750

something that I looked at was the %

189
00:09:02,030 --> 00:08:59,070
methane produced in each test tube

190
00:09:04,610 --> 00:09:02,040
and it does look like the tubes with

191
00:09:06,470 --> 00:09:04,620
more methane produced have wider bands

192
00:09:09,560 --> 00:09:06,480
and so this is also something we're

193
00:09:13,580 --> 00:09:09,570
going to look into further so in

194
00:09:17,210 --> 00:09:13,590
conclusion our organism Wolfie I is very

195
00:09:20,000 --> 00:09:17,220
robust organism it is a thermo file but

196
00:09:22,550 --> 00:09:20,010
is also is able to rebound quickly from

197
00:09:25,330 --> 00:09:22,560
4 degrees Celsius and negative 15

198
00:09:27,140 --> 00:09:25,340
degrees Celsius and it has shown

199
00:09:30,260 --> 00:09:27,150
production of methane at those

200
00:09:32,000 --> 00:09:30,270
temperatures as well another conclusion

201
00:09:35,540 --> 00:09:32,010
is that a greater amount of initial

202
00:09:38,090 --> 00:09:35,550
liquid medium promotes greater growth

203
00:09:40,640 --> 00:09:38,100
such as when we use 10 milliliters

204
00:09:42,860 --> 00:09:40,650
medium and only half as much sand versus

205
00:09:45,500 --> 00:09:42,870
when we used a saturation experiment and

206
00:09:47,870 --> 00:09:45,510
then finally our discoloration of sand

207
00:09:49,340 --> 00:09:47,880
could indicate environmental alteration

208
00:09:52,700 --> 00:09:49,350
but this is something we're going to

209
00:09:53,690 --> 00:09:52,710
look into further for the future studies

210
00:09:55,820 --> 00:09:53,700
we're going to continue these

211
00:09:57,200 --> 00:09:55,830
experiments and also repeat them just to

212
00:10:00,350 --> 00:09:57,210
make sure that those increases in

213
00:10:02,150 --> 00:10:00,360

methane are accurate and that you know

214
00:10:04,910 --> 00:10:02,160
something wasn't going wrong we also

215
00:10:06,530 --> 00:10:04,920
want to explore porosity further with

216
00:10:08,810 --> 00:10:06,540
different Mars simulants oils such as

217
00:10:11,360 --> 00:10:08,820
JSC Mars one and the Mars Mojave

218
00:10:13,490 --> 00:10:11,370
simulant and we're also going to further

219
00:10:15,710 --> 00:10:13,500
our temperature study by using a

220
00:10:17,480 --> 00:10:15,720
negative 80 degree freezer to more mimic

221
00:10:22,160 --> 00:10:17,490
those temperatures on Mars that we see

222
00:10:24,170 --> 00:10:22,170
and so how does this relate to Mars life

223
00:10:28,070 --> 00:10:24,180
is robust I was also going to use the

224
00:10:29,630 --> 00:10:28,080
life finds away but I didn't but life's

225
00:10:32,180 --> 00:10:29,640
robust so these are actually

226
00:10:35,480 --> 00:10:32,190
thermophiles that I'm working with but

227
00:10:38,450 --> 00:10:35,490
at least Wolfie I shows promise of being

228
00:10:42,020 --> 00:10:38,460
able to rebound from freeze-thaw cycles

229
00:10:44,290 --> 00:10:42,030
and if Mars was perhaps similar to earth

230
00:10:46,490 --> 00:10:44,300
in the past if there were dormant

231
00:10:52,900 --> 00:10:46,500
microorganisms they could be existing in

232
00:10:52,910 --> 00:11:08,689
so do we have any questions for Rebecca

233
00:11:11,579 --> 00:11:10,499
thank you for your talk I just want to

234
00:11:14,429 --> 00:11:11,589
make sure i understand correctly

235
00:11:17,789 --> 00:11:14,439
wondering your grass so um on the

236
00:11:21,539 --> 00:11:17,799
graphic showing set to for example think

237
00:11:24,449 --> 00:11:21,549
you should i seen it yeah sir sins

238
00:11:27,419 --> 00:11:24,459

through the so we're in their first red

239
00:11:31,559 --> 00:11:27,429
box where you see a methane increase

240
00:11:33,179 --> 00:11:31,569
that's at minus 15 degrees c so you're

241
00:11:34,949 --> 00:11:33,189
sampling from the headspace but at this

242
00:11:38,549 --> 00:11:34,959
temperature your medium is is frozen i

243
00:11:39,929 --> 00:11:38,559
believe correct yep um so when you start

244
00:11:42,059 --> 00:11:39,939
at the end of the four degree experiment

245
00:11:43,859 --> 00:11:42,069
you take a a time measurement and you

246
00:11:47,309 --> 00:11:43,869
have a concentration of methane and then

247
00:11:48,689 --> 00:11:47,319
after a few few days you take it after a

248
00:11:51,389 --> 00:11:48,699
few days minus fifteen and you see an

249
00:11:53,069 --> 00:11:51,399
increase in methane so does it mean like

250
00:11:55,379 --> 00:11:53,079
so the methane would have died fused

251
00:11:57,539 --> 00:11:55,389
through the ice and go into the

252
00:11:59,999 --> 00:11:57,549
headspace the vial and that's what you

253
00:12:04,409 --> 00:12:00,009
measure is that correct or do you melt

254
00:12:06,210 --> 00:12:04,419
your sample before they when we do take

255
00:12:08,039 --> 00:12:06,220
measurements they we are taking the

256
00:12:10,229 --> 00:12:08,049
measurements at room temperature so it's

257
00:12:12,179 --> 00:12:10,239
possible that there's some melting but

258
00:12:14,789 --> 00:12:12,189
they're basically still frozen when

259
00:12:17,249 --> 00:12:14,799
we're taking our measurements um we're

260
00:12:20,780 --> 00:12:17,259
also interested in why this is occurring

261
00:12:23,340 --> 00:12:20,790
we looked at methane solubility in

262
00:12:25,289 --> 00:12:23,350
liquid water and frozen water and it's

263
00:12:28,079 --> 00:12:25,299

has nothing to do with solubility

264
00:12:30,179 --> 00:12:28,089
because solubility is very low but we're

265
00:12:31,229 --> 00:12:30,189
interested in this as well and it's

266
00:12:35,159 --> 00:12:31,239
something that weren't further

267
00:12:36,389 --> 00:12:35,169
investigation so this is also something

268
00:12:38,699 --> 00:12:36,399
that's see in the field in permafrost

269
00:12:41,729 --> 00:12:38,709
during the onset of freezing you get

270
00:12:44,159 --> 00:12:41,739
this freeze expulsion of methane from

271
00:12:46,289 --> 00:12:44,169
the water phase and it gets pumped out

272
00:12:47,729 --> 00:12:46,299
so if the onset of winter comes on you

273
00:12:49,559 --> 00:12:47,739
get these enormous bursts of methane is

274
00:12:52,499 --> 00:12:49,569
where the majority or in some places of

275
00:12:54,090 --> 00:12:52,509
methane flux comes from so I'm I was

276
00:12:55,139 --> 00:12:54,100
worried about the same thing think

277
00:12:57,269 --> 00:12:55,149
what's happening is that you're freezing

278
00:12:58,379 --> 00:12:57,279
your solution your ex building all your

279
00:12:59,970 --> 00:12:58,389
methane out there so it's not active

280
00:13:01,529 --> 00:12:59,980
production okay you're expelling the

281
00:13:03,090 --> 00:13:01,539
production you've made during the four C

282
00:13:09,419 --> 00:13:03,100
times and then you're dropping back down

283
00:13:13,229 --> 00:13:09,429
again yeah right okay yeah that is um

284
00:13:15,749 --> 00:13:13,239
see that's both very good points that's

285
00:13:18,749 --> 00:13:15,759
something we would expect as well as

286
00:13:19,710 --> 00:13:18,759
methane was being produced earlier

287
00:13:25,460 --> 00:13:19,720
either

288
00:13:27,990 --> 00:13:25,470

at 4 degrees c if it was in the soil or

289
00:13:31,199 --> 00:13:28,000
in the ice and then it was released

290
00:13:39,749 --> 00:13:31,209
later maybe we can have one more

291
00:13:42,780 --> 00:13:39,759
question for Rebecca if there's one can

292
00:13:44,610 --> 00:13:42,790
you comment on the slope of your I mean

293
00:13:49,139 --> 00:13:44,620
it does that mean anything the fact that

294
00:13:52,740 --> 00:13:49,149
they have different um basically wolfie

295
00:13:56,929 --> 00:13:52,750
I is just a more robust grower in itself

296
00:13:59,610 --> 00:13:56,939
it's more robust organism in various

297
00:14:02,069 --> 00:13:59,620
environments it just shows better growth

298
00:14:04,530 --> 00:14:02,079
and survivability and so I think that's

299
00:14:11,670 --> 00:14:04,540
the difference between Wolfie I and for

300
00:14:13,769 --> 00:14:11,680
me to come oh yes that also i don't know

301
00:14:15,780 --> 00:14:13,779
i mean this is these are both very um

302
00:14:18,749 --> 00:14:15,790
this these experiments are all very

303
00:14:21,530 --> 00:14:18,759
preliminary and this is just we've only

304
00:14:23,819 --> 00:14:21,540
looked at how much % methane was

305
00:14:24,900 --> 00:14:23,829
produced over the time period and now we

306
00:14:30,119 --> 00:14:24,910
have to go back and look at everything