



# ABS*ci*CON 2017

MESA, ARIZONA

1  
00:00:12,250 --> 00:00:06,150

you

2  
00:00:16,870 --> 00:00:14,220

[Music]

3  
00:00:19,120 --> 00:00:16,880

thank you for coming everyone just a

4  
00:00:23,160 --> 00:00:19,130

brief update on the title as you may

5  
00:00:27,540 --> 00:00:23,170

have read so I changed the title to

6  
00:00:29,859 --> 00:00:27,550

evidence for Institute iron cycling from

7  
00:00:31,480 --> 00:00:29,869

focusing just on the let's autotrophs

8  
00:00:34,869 --> 00:00:31,490

because I think the iron cycle in

9  
00:00:39,460 --> 00:00:34,879

general makes for a more interesting

10  
00:00:42,100 --> 00:00:39,470

story in my system so to get everyone

11  
00:00:44,680 --> 00:00:42,110

oriented chocolate pots is in

12  
00:00:47,100 --> 00:00:44,690

Yellowstone and it's in the northwest

13  
00:00:51,790 --> 00:00:47,110

corner right along the given river

14

00:00:54,310 --> 00:00:51,800

denoted by the blue X and here we have a

15

00:00:59,470 --> 00:00:54,320

couple pictures of hot springs we have

16

00:01:02,619 --> 00:00:59,480

the oops there we go so we have the vent

17

00:01:05,469 --> 00:01:02,629

pool up here the flow path and then

18

00:01:07,560 --> 00:01:05,479

chocolate pots is very interesting hot

19

00:01:10,810 --> 00:01:07,570

spring because it's circum neutral pH

20

00:01:13,719 --> 00:01:10,820

mildly thermophilic and contains a high

21

00:01:17,740 --> 00:01:13,729

amount of iron and silica in the spring

22

00:01:19,420 --> 00:01:17,750

water from a biological perspective were

23

00:01:23,649 --> 00:01:19,430

interested environments like this

24

00:01:27,999 --> 00:01:23,659

because of the new and recent

25

00:01:30,730 --> 00:01:28,009

discoveries of certain neutral pH like

26  
00:01:33,940 --> 00:01:30,740  
environments on Mars and the evidence of

27  
00:01:37,330 --> 00:01:33,950  
relic hot spring-like features and if

28  
00:01:40,480 --> 00:01:37,340  
we're able to nail down evidence for

29  
00:01:43,840 --> 00:01:40,490  
internally cycling iron base metabolisms

30  
00:01:46,149 --> 00:01:43,850  
in environments like chocolate pots then

31  
00:01:50,260 --> 00:01:46,159  
this could in theory be applied to some

32  
00:01:53,140 --> 00:01:50,270  
of these relic features on Mars so we

33  
00:01:55,060 --> 00:01:53,150  
have two main hypotheses going forward

34  
00:01:57,609 --> 00:01:55,070  
with this study the first is that there

35  
00:02:00,550 --> 00:01:57,619  
are native active iron reducing

36  
00:02:03,490 --> 00:02:00,560  
microorganisms present in the chocolate

37  
00:02:06,010 --> 00:02:03,500  
pots event and also that there are lipid

38  
00:02:08,919 --> 00:02:06,020

atrophic iron oxidizers which are also

39

00:02:12,009 --> 00:02:08,929

actively cycling iron and in addition as

40

00:02:15,970 --> 00:02:12,019

let the autotrophs they are fixing

41

00:02:20,050 --> 00:02:15,980

carbon debt yeah fixing co2 and able to

42

00:02:22,180 --> 00:02:20,060

supplement other sources of carbon for

43

00:02:24,730 --> 00:02:22,190

the heterotrophic community

44

00:02:27,680 --> 00:02:24,740

so backing up a little bit some of our

45

00:02:31,100 --> 00:02:27,690

initial work with materials from

46

00:02:34,449 --> 00:02:31,110

chocolate pots we ran a series of iron

47

00:02:38,510 --> 00:02:34,459

reducing enrichment culture studies and

48

00:02:41,120 --> 00:02:38,520

this is a graph showing the repeated

49

00:02:44,110 --> 00:02:41,130

transfer of these cultures for over four

50

00:02:46,850 --> 00:02:44,120

years and they've been able to sustain a

51  
00:02:52,190 --> 00:02:46,860  
fairly high level of iron reducing

52  
00:02:54,670 --> 00:02:52,200  
activity and through both 16s and meta

53  
00:02:57,560 --> 00:02:54,680  
genomic sequence data we've identified

54  
00:03:00,979 --> 00:02:57,570  
sequences that are related to known iron

55  
00:03:05,380 --> 00:03:00,989  
reducing taxa including a Geo vector and

56  
00:03:09,050 --> 00:03:05,390  
mealy factor moving one step towards

57  
00:03:11,330 --> 00:03:09,060  
choosing apart the infant we ran another

58  
00:03:15,520 --> 00:03:11,340  
series of short duration incubations

59  
00:03:18,590 --> 00:03:15,530  
using just the native sediments and

60  
00:03:20,960 --> 00:03:18,600  
amended with and without additional

61  
00:03:22,729 --> 00:03:20,970  
electron donor and the things I'd like

62  
00:03:25,790 --> 00:03:22,739  
to point out here is in this feature

63  
00:03:27,500 --> 00:03:25,800

which they're the that figure which

64

00:03:30,650 --> 00:03:27,510

corresponds to material collected from

65

00:03:32,750 --> 00:03:30,660

the hot spring vent you have an almost

66

00:03:36,920 --> 00:03:32,760

equivalent level of iron reduction

67

00:03:39,170 --> 00:03:36,930

activity regardless of electron donor

68

00:03:41,870 --> 00:03:39,180

source whereas if you move a few meters

69

00:03:44,660 --> 00:03:41,880

away at site three there's still a

70

00:03:47,479 --> 00:03:44,670

measurable amount of activity of the

71

00:03:51,740 --> 00:03:47,489

organisms using their native carbon

72

00:03:54,170 --> 00:03:51,750

source but it's greatly reduced so this

73

00:03:57,819 --> 00:03:54,180

led us to the question of what is this

74

00:03:59,660 --> 00:03:57,829

internal carbon source is it coming from

75

00:04:03,289 --> 00:03:59,670

chemoautotrophs is it from the

76  
00:04:05,810 --> 00:04:03,299  
cyanobacterial community is it breakdown

77  
00:04:08,479 --> 00:04:05,820  
of plant or animal to trade detritus

78  
00:04:12,020 --> 00:04:08,489  
from the external environment whereas

79  
00:04:15,800 --> 00:04:12,030  
the carbon coming from so and we have

80  
00:04:19,069 --> 00:04:15,810  
16s data from this as well and several

81  
00:04:21,140 --> 00:04:19,079  
Oh to use related to an organism

82  
00:04:24,650 --> 00:04:21,150  
Ignasi bacterium while it is not

83  
00:04:26,600 --> 00:04:24,660  
implicated as an iron reducer it is very

84  
00:04:31,320 --> 00:04:26,610  
similar to this newly or a factor which

85  
00:04:33,790 --> 00:04:31,330  
I identified in the enrichment cultures

86  
00:04:36,340 --> 00:04:33,800  
taking a look now at the in Scituate

87  
00:04:39,630 --> 00:04:36,350  
vironment we collected a series of core

88  
00:04:44,080 --> 00:04:39,640

samples moving down the flow path and

89

00:04:47,200 --> 00:04:44,090

the 16s data from these samples revealed

90

00:04:49,570 --> 00:04:47,210

sequences related to known with

91

00:04:52,690 --> 00:04:49,580

autotrophic iron oxidizers sister

92

00:04:54,970 --> 00:04:52,700

oxidants and again this relative of a

93

00:05:00,040 --> 00:04:54,980

potential iron reducer Ignasi bacterial

94

00:05:03,670 --> 00:05:00,050

ease so magic and ohmic work flow I'll

95

00:05:06,130 --> 00:05:03,680

cover in brief you start out with a very

96

00:05:08,590 --> 00:05:06,140

complex community like chop the pots get

97

00:05:12,430 --> 00:05:08,600

all the DNA sequence it put it back

98

00:05:15,460 --> 00:05:12,440

together separate it out into bins which

99

00:05:17,950 --> 00:05:15,470

are you can think of as being different

100

00:05:19,720 --> 00:05:17,960

taxa within the community and then what

101

00:05:21,909 --> 00:05:19,730

we're really interested in is what is

102

00:05:26,290 --> 00:05:21,919

the metabolic potential of some of these

103

00:05:28,240 --> 00:05:26,300

these bins or taxa and we're taking a

104

00:05:33,010 --> 00:05:28,250

targeted meta genomic approach looking

105

00:05:35,680 --> 00:05:33,020

at which genes are appear to be involved

106

00:05:39,730 --> 00:05:35,690

in iron cycling and which ones are

107

00:05:43,240 --> 00:05:39,740

involved in carbon fixation so here we

108

00:05:47,500 --> 00:05:43,250

have the community of two metagenomes

109

00:05:49,840 --> 00:05:47,510

that I assembled from chocolate pots one

110

00:05:51,880 --> 00:05:49,850

is from water filtered from the vent

111

00:05:54,700 --> 00:05:51,890

pool and another one is a combined

112

00:05:56,560 --> 00:05:54,710

assembly from three of those sediment

113

00:06:02,230 --> 00:05:56,570

cores that I mentioned and once again

114

00:06:04,330 --> 00:06:02,240

you see bins related to this known with

115

00:06:09,640 --> 00:06:04,340

the autotroph and this possible iron

116

00:06:14,590 --> 00:06:09,650

reducer so thank you to Dave Emerson for

117

00:06:16,270 --> 00:06:14,600

the lovely introduction - I see - I will

118

00:06:18,730 --> 00:06:16,280

not be covering that in my talk because

119

00:06:20,969 --> 00:06:18,740

I found no homologues that in my system

120

00:06:23,770 --> 00:06:20,979

but I will be talking about this other

121

00:06:27,719 --> 00:06:23,780

electron or extracellular electron

122

00:06:30,940 --> 00:06:27,729

transfer system a pouring wrapped

123

00:06:33,159 --> 00:06:30,950

version where you have a pour in in

124

00:06:35,620 --> 00:06:33,169

opening in the outer membrane of the

125

00:06:37,659 --> 00:06:35,630

cell and a cytochrome which fits into

126

00:06:39,490 --> 00:06:37,669

that and is able to transfer electrons

127

00:06:41,830 --> 00:06:39,500

into or out of the cell depending on

128

00:06:43,350 --> 00:06:41,840

whether it's an iron oxidizer or I

129

00:06:47,040 --> 00:06:43,360

reducer

130

00:06:48,719 --> 00:06:47,050

and there's two homologs in particular I

131

00:06:52,740 --> 00:06:48,729

want to point out there's this MTO a

132

00:06:55,350 --> 00:06:52,750

system which is identified in said er

133

00:06:59,219 --> 00:06:55,360

oxidants and there's this geo factor

134

00:07:02,279 --> 00:06:59,229

like pouran cytochrome system and it's

135

00:07:06,779 --> 00:07:02,289

also been identified in that Ignasi

136

00:07:08,339 --> 00:07:06,789

bacterium organism these are just

137

00:07:11,100 --> 00:07:08,349

several different search tools that you

138

00:07:13,230 --> 00:07:11,110

can use hidden Markov models blastp

139

00:07:14,850 --> 00:07:13,240

homology many different ways to identify

140

00:07:15,839 --> 00:07:14,860

these genes but one of the more

141

00:07:19,860 --> 00:07:15,849

important things that we're interested

142

00:07:25,170 --> 00:07:19,870

in is the genomic context of these genes

143

00:07:28,080 --> 00:07:25,180

that genomically you need to find a pore

144

00:07:34,850 --> 00:07:28,090

in encoded next to one or several of

145

00:07:38,189 --> 00:07:34,860

these cytochromes okay we're looking for

146

00:07:40,050 --> 00:07:38,199

multiple co2 fixation pathways but for

147

00:07:43,679 --> 00:07:40,060

the sake of brevity I'm only going to

148

00:07:47,969 --> 00:07:43,689

cover Rubisco today so to quote my

149

00:07:51,749 --> 00:07:47,979

advisor if you identify a bin with an ET

150

00:07:54,119 --> 00:07:51,759

system and Rubisco that's a pretty good

151  
00:07:57,379 --> 00:07:54,129  
smoking gun of an organism that may be

152  
00:08:01,140 --> 00:07:57,389  
involved in a litter litho autotrophic

153  
00:08:05,010 --> 00:08:01,150  
lifestyle and we very similar search

154  
00:08:09,329 --> 00:08:05,020  
parameters for these genes so here I

155  
00:08:13,740 --> 00:08:09,339  
have a summary of all of the metagenomic

156  
00:08:16,969 --> 00:08:13,750  
bins from these two assemblies in red we

157  
00:08:23,249 --> 00:08:16,979  
have all of the bins that contained some

158  
00:08:28,249 --> 00:08:23,259  
model EEG system in yellow we have all

159  
00:08:31,230 --> 00:08:28,259  
of the bins that contained a hit for

160  
00:08:34,139 --> 00:08:31,240  
Rubisco and what we're mostly interested

161  
00:08:36,569 --> 00:08:34,149  
in is these bins that contained both

162  
00:08:39,839 --> 00:08:36,579  
systems and may be involved in chemo

163  
00:08:41,459 --> 00:08:39,849

little otter trophy and then down here

164

00:08:44,010 --> 00:08:41,469

at the bottom are bins that were not

165

00:08:47,310 --> 00:08:44,020

involved in any of it

166

00:08:49,680 --> 00:08:47,320

so bringing this back to the two

167

00:08:52,050 --> 00:08:49,690

organisms that I've been mentioning

168

00:08:53,610 --> 00:08:52,060

throughout this talk the sifter oxidants

169

00:08:57,600 --> 00:08:53,620

and the ignominy she really is

170

00:09:00,269 --> 00:08:57,610

I just wanted to summarize the results

171

00:09:02,880 --> 00:09:00,279

in terms of those two organisms so up in

172

00:09:05,340 --> 00:09:02,890

gray we have the published genomes of

173

00:09:07,680 --> 00:09:05,350

these organisms and against utter

174

00:09:12,510 --> 00:09:07,690

oxidants containing this MTO system and

175

00:09:14,550 --> 00:09:12,520

Rubisco and the two related

176

00:09:17,190 --> 00:09:14,560

Ignasi bacterium and me leroy vector

177

00:09:20,519 --> 00:09:17,200

containing only that geo vector like TCC

178

00:09:21,360 --> 00:09:20,529

system whereas looking at the two

179

00:09:24,000 --> 00:09:21,370

metagenomes

180

00:09:27,360 --> 00:09:24,010

here we have a sitter oxidants relative

181

00:09:29,880 --> 00:09:27,370

that all of a sudden has a PCC system

182

00:09:34,769 --> 00:09:29,890

and we have these Ignasi bacterias that

183

00:09:38,100 --> 00:09:34,779

have MgO and Rubisco which is really

184

00:09:40,949 --> 00:09:38,110

interesting that sitter oxidants which

185

00:09:44,699 --> 00:09:40,959

is a known with autotrophic iron

186

00:09:46,590 --> 00:09:44,709

oxidizer contains PCC which so far has

187

00:09:48,990 --> 00:09:46,600

only been implicated in iron reduction

188

00:09:53,040 --> 00:09:49,000

so perhaps this is a relative that is

189

00:09:56,250 --> 00:09:53,050

able to carry out both iron oxidation

190

00:09:58,500 --> 00:09:56,260

and iron reduction and for the Ignasi

191

00:10:02,460 --> 00:09:58,510

bacteria Li's as I mentioned before has

192

00:10:06,389 --> 00:10:02,470

an e ET system and Rubisco so perhaps it

193

00:10:12,099 --> 00:10:06,399

is capable of carrying out a little odd

194

00:10:18,079 --> 00:10:15,710

so to wrap things up once again we have

195

00:10:21,049 --> 00:10:18,089

a highly active dissimilatory iron

196

00:10:23,599 --> 00:10:21,059

reducing community at the chocolate pots

197

00:10:25,819 --> 00:10:23,609

event and it is able to sustain itself

198

00:10:28,280 --> 00:10:25,829

using only the native carbon source

199

00:10:30,619 --> 00:10:28,290

we have metagenomic bins that are

200

00:10:33,799 --> 00:10:30,629

related to known and potential iron

201  
00:10:36,289 --> 00:10:33,809  
cycling microorganisms we have the

202  
00:10:38,720 --> 00:10:36,299  
positive identification of homologs of

203  
00:10:41,629 --> 00:10:38,730  
genes involved in both these eeet

204  
00:10:43,970 --> 00:10:41,639  
pathways and carbon fixation pathways

205  
00:10:48,109 --> 00:10:43,980  
and as a side note we're in the process

206  
00:10:51,109 --> 00:10:48,119  
of identifying lipid biomarkers from

207  
00:10:54,049 --> 00:10:51,119  
these materials to try and nail down a

208  
00:10:57,829 --> 00:10:54,059  
possible source of the carbon that's

209  
00:11:01,749 --> 00:10:57,839  
present in statue and one last thing I

210  
00:11:05,150 --> 00:11:01,759  
would like to mention so I only covered

211  
00:11:06,349 --> 00:11:05,160  
very briefly the model systems that

212  
00:11:09,109 --> 00:11:06,359  
we've been looking at but we've also

213  
00:11:11,720 --> 00:11:09,119

expanded our search to look for non

214

00:11:14,769 --> 00:11:11,730

model ET systems and we're also looking

215

00:11:20,239 --> 00:11:14,779

at all of the known carbon fixation

216

00:11:22,189 --> 00:11:20,249

pathways in these systems and that I'd

217

00:11:25,099 --> 00:11:22,199

like to thank the organizers for the

218

00:11:26,659 --> 00:11:25,109

conference my funding my committee

219

00:11:29,479 --> 00:11:26,669

members who have helped with this

220

00:11:32,419 --> 00:11:29,489

project Eric and Eric and my colleagues

221

00:11:36,669 --> 00:11:32,429

back at UW Madison particularly Xiaomei

222

00:11:39,109 --> 00:11:36,679

hay who without her meta genomic

223

00:11:43,669 --> 00:11:39,119

expertise we would not have been able to

224

00:11:50,450 --> 00:11:43,679

do any of this so with that I think I

225

00:12:01,220 --> 00:11:52,460

so we definitely have time for a couple

226

00:12:03,590 --> 00:12:01,230

of questions Tim yeah yeah I really

227

00:12:06,020 --> 00:12:03,600

enjoy the Yellowstone toxemia yeah

228

00:12:09,680 --> 00:12:06,030

interesting it's a cool area

229

00:12:13,580 --> 00:12:09,690

so what non-model systems do you have in

230

00:12:17,390 --> 00:12:13,590

mind exactly as far as the Ichigo yeah

231

00:12:21,710 --> 00:12:17,400

yeah so again a lot of this credit goes

232

00:12:25,130 --> 00:12:21,720

to Xiaomei and her computer programming

233

00:12:27,950 --> 00:12:25,140

skills but we have scripts that were

234

00:12:31,700 --> 00:12:27,960

able to search our meta genomic

235

00:12:36,320 --> 00:12:31,710

sequences for the Keene binding site

236

00:12:38,480 --> 00:12:36,330

motifs and any genes that have that

237

00:12:41,960 --> 00:12:38,490

appear to be multicam cytochromes we

238

00:12:44,330 --> 00:12:41,970

then look for genes proximal to that

239

00:12:47,450 --> 00:12:44,340

that are predicted to be an outer

240

00:12:48,980 --> 00:12:47,460

membrane pore in so the ones that aren't

241

00:12:52,400 --> 00:12:48,990

homologous to any of the models but

242

00:12:55,000 --> 00:12:52,410

still seem to fit that same type of

243

00:12:59,560 --> 00:12:55,010

system with a poran and a cytochrome

244

00:13:02,060 --> 00:12:59,570

what about the EPI or GOP ly any

245

00:13:05,660 --> 00:13:02,070

considerations about yeah so that's

246

00:13:08,110 --> 00:13:05,670

something that I've considered and we've

247

00:13:10,250 --> 00:13:08,120

discussed as a lab quite a bit but

248

00:13:13,640 --> 00:13:10,260

something that I I still need to

249

00:13:16,940 --> 00:13:13,650

investigate with these systems yeah

250

00:13:18,830 --> 00:13:16,950

thanks yeah welcome thank you so we

251

00:13:23,490 --> 00:13:18,840

still have time for another question if