

Response to anonymous referee no. 2 (BGD 9, C2834-C2836, 2012)
From Buford Price, first author of manuscript BGD 9, 6535-6577, 2012

Changes are indicated in blue.

Response to comment 1:

At end of Introduction:

For a typical generation time of 1 day for *Pro*, *Syn*, and other common bacteria in warm oceans, ice cores offer the opportunity to study evolution over ~300 million generations by analyzing the genomes vs depth of those blown onto the oldest glacial ice and preserved without mutations at subzero temperature.

Response to comment 2 is at bottom of p. 3 of my attached Word document:

Because of the low cell concentration in ice from most sites, we used a DupontSorvall RC-5B centrifuge at 14000 rpm for 45 minutes at 10°C to achieve an order of magnitude increase in concentration that we estimated by comparing cell counts before and after centrifuging. We are aware that centrifuging introduces some mass-dependent bias, for example, by favoring *Synechococcus* over *Prochlorococcus*.

Response to comment 3 is on bottom of my p. 4 of my attached Word document:

The calculation of cell concentration in an ice sample took into account both the volume of melted ice collected in the flow cytometer during an FCM run and the increased concentration due to centrifuging. In order to obtain at least 10^2 cells ml^{-1} per FCM run, we filled a 4 ml FCM vial with melted ice that had been centrifuged twice to increase the cell concentration by a factor up to 25.

Indicated in blue under minor comments:

1. On p 4 of my attached Word document: I defined **FSC** and **SSC**.
2. At bottom of p. 4 of my attached Word document: I added the sentence in blue: In order to obtain at least 10^2 cells ml^{-1} per FCM run, we filled a 4 ml FCM vial with melted ice that had been centrifuged twice to increase the cell concentration by a factor up to 25.
3. In first paragraph on Results, I changed **BSF** to **BFS**.
4. In references, I changed the publication date of Marie et al. from 1990 to 1999.
5. I changed the text on p.6556, l. 28 to p. 6557, l. 8 to the following:
Furthermore, the following results of Tung et al. (2005) allow one to set an experimental upper limit much less than $1 \mu\text{m}$ per year on the vertical distances bacteria travel in veins: at depths of 2054 m, 3018 m, and 3036 m in GISP ice, they found a factor ~10 higher concentration of methanogens at depths within $20 \mu\text{m}$ of the depths of spikes of excess methane found by Ed Brook (unpublished results). They concluded that the excess methane had been generated by methanogens metabolizing during more than one hundred thousand years in the ice at a temperature of -10°C . Later Rohde et al. (2008) found spikes of excess Trp that they attributed at least in part to N_2O -producing microbes at exactly the same depths in GISP ice where Sowers et al. (2003) had found narrow spikes of excess N_2O . Random walk of such microbes in veins would have smeared out the sharp agreement between depth of microbes and depth of their gas production.

6. Bramall 2007: his Ph.D. thesis was never published.
 7. In the caption to Fig. 1a, I replaced BSF with BFS.
 8. In Fig. 2, I added the units for y-axis.
 9. I used a 10 um scale bar for Fig. 3A.
 10. I added a scale bar.
 11. I added a description of (I) to (L) in the caption to Fig. 6.
 12. I corrected Fig. 7.
- Note that I have attached the revised versions of Figs. 3, 6, and 7.

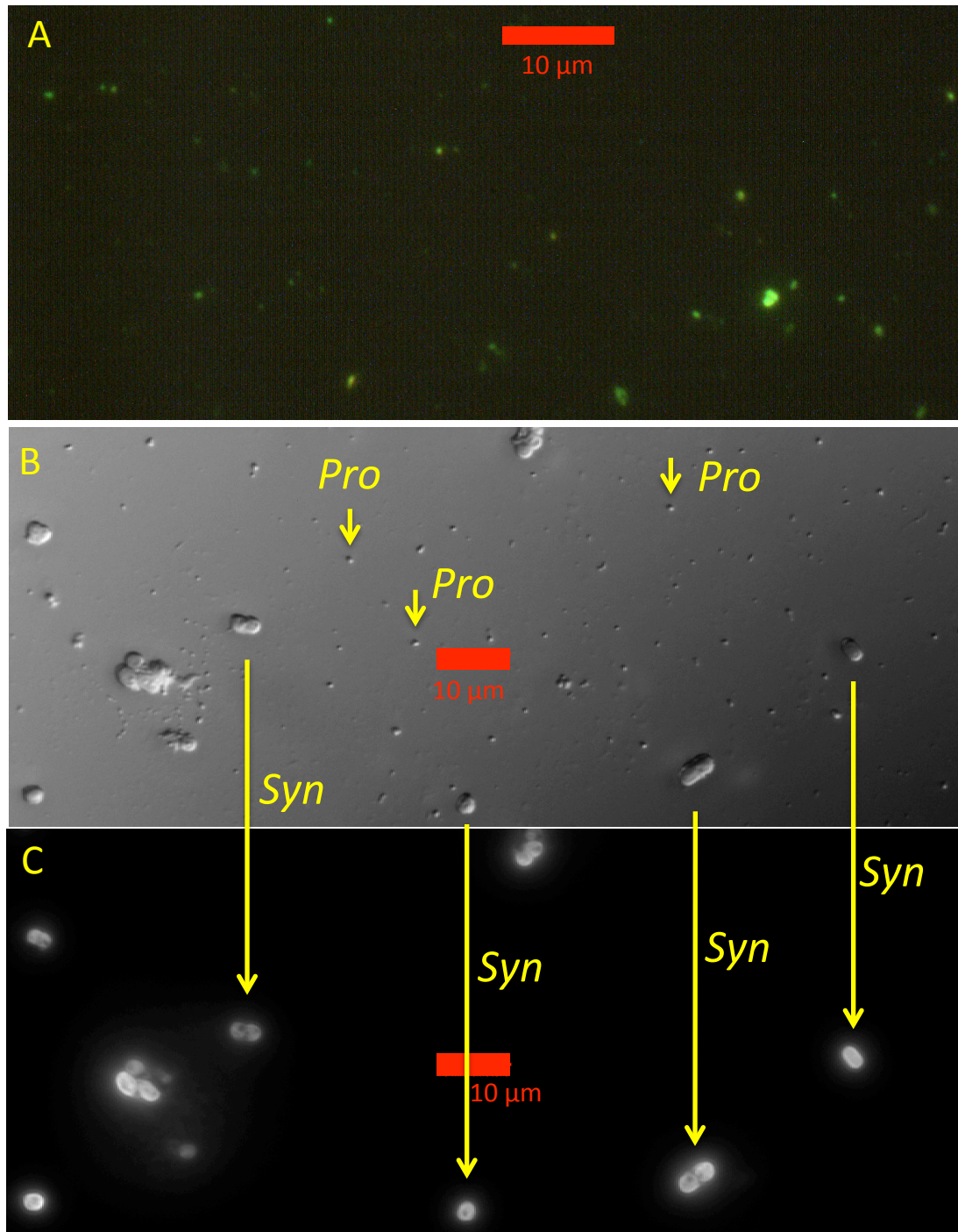


Fig. 3

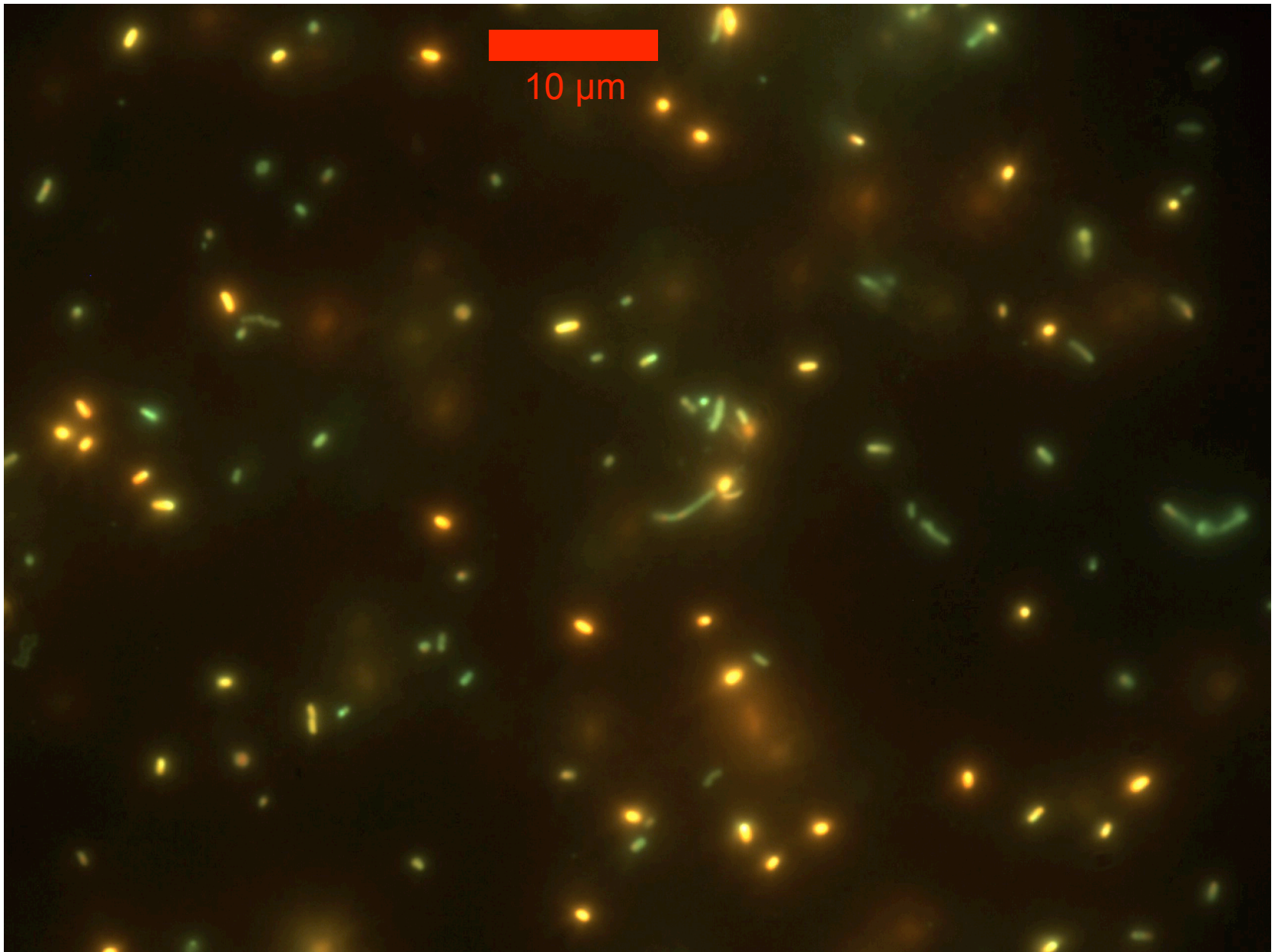


Figure 4

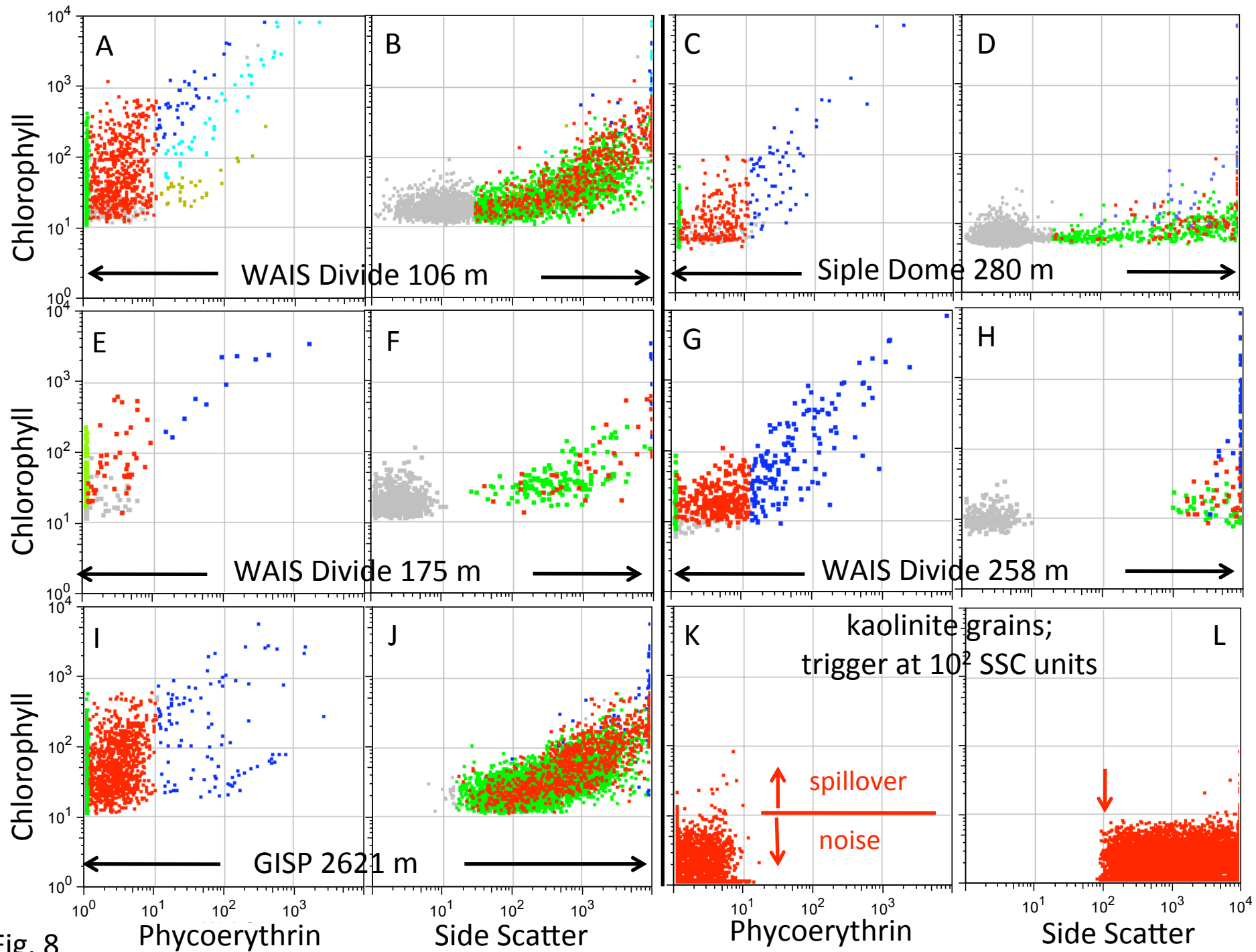


Fig. 8