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Biogeosciences Discussions

Interactive comment on "Determination of the metabolically active fraction of benthic foraminifera by means of Fluorescent in situ Hybridization (FISH)" *by* C. Borrelli et al.

Anonymous Referee #4

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Much has been written over the past several decades on methods used to distinguish live from dead foraminifera in environmental samples. Borrelli et al. propose the application of FISH as a more accurate tool for this, but several concerns need to be addressed. First, I am particularly concerned about the small sample sizes used in the study. Overall, too few individuals were used in each treatment to make the results meaningful. For example, only 5 individuals with agglutinated tests were used, and it appears that these data are repeated on Tables 1 & 3. On Table 3, it appears that only a few specimens were assayed using the S17 probe (7 Ammonia, 6 miliolids – species not distinguished). Only 62 individuals (Table 1) or 69 individuals (Table 3) were used for all treatments and controls. This is clearly not sufficient given the range of variabil-

ity, including the intensity in fluorescence. Second, the discrepancy in Table 1 between the number of individuals tabulated as live/dead using optical examination (which I assume is the search for pseudopodia and cytoplasmic coloration) and the live/dead tally using FISH is troublesome. To show that this method is truly valid, only foraminifers that were verifiably alive should have been used. There is no way to tell whether individuals were counted as alive using optical methods that were actually dead (or the other way around). Again, this discrepancy is particularly troublesome given the small sample size. Third, the results on fed v. starved individuals is interesting, and in a general way, they are in agreement with the findings of Parfrey and Katz (2010, Genome Biol. Evol., 2:678-685). However, again, the small number of individuals examined precludes drawing any solid conclusions. Overall, the results only demonstrate the potential application of FISH as a vital assay, but the superiority of this method over others was not adequately demonstrated for the reasons cited above.

Interactive comment on Biogeosciences Discuss., 7, 7475, 2010.