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Photosynthesis or planktonic respiration?

The nutrient-poor (oligotrophic) regions of the open ocean cover 30% of the Earth's surface. Microscopic plants (phytoplankton) living in this habitat account for about 10% of global CO_2 fixation[1]. Most of this organic production is rapidly respired within a microbial food web dominated by photosynthetic bacteria, heterotrophic bacteria and small (<5 μm diameter) protozoa and algae, with a small amount (about 1% of global CO_2 fixation) being exported from nutrient-poor upper ocean regions to the interior of the ocean[2,3]. Thus, on average, photosynthesis exceeds respiration in the sunlit 'euphotic' zone of the oligotrophic ocean. This view appears to have been challenged by del Giorgio and co-workers[4].

Although their conclusion that "…bacterial respiration is generally high, and tends to exceed phytoplankton net production in unproductive systems (less than 70 to 120 μg carbon per litre per day)" is likely to be valid for lakes, it is unlikely to hold in the oligotrophic ocean. An excess of net photosynthesis over respiration is evident in seasonal accumulation of O_2 in near-surface waters[5] at those times when nutrient supply from the interior of the ocean is cut off by vertical stratification. Net photosynthesis is also evident in downward transport (export) of particulate[2] and dissolved organic matter[3] from the euphotic zone to the interior of the ocean.

The conclusion of del Giorgio *et al.* that the oligotrophic ocean euphotic zone is net heterotrophic was based on analysis of estimates of bacterial respiration and primary production taken from the literature. But a systematic bias in methodology, possibly the consideration of phytoplankton respiration, may account for such a discrepancy.

Both bacterial biomass and bacterial respiration in oligotrophic ocean environments was probably overestimated by del Giorgio *et al.* Photosynthetic picoplankton can be mistaken for heterotrophic bacteria in conventional epifluorescence counts, leading to an overestimate of the abundance of heterotrophic bacteria in the most oligotrophic waters. For example, photosynthetic bacteria within the genus *Prochlorococcus* accounted for 31% of total bacterial counts (upper 200 m) in the oligotrophic North Pacific, and the biomass of photosynthetic picoplankton exceeds that of heterotrophic bacteria[6].

del Giorgio *et al.* relied on dark O_2 consumption in the <0.8–2-μm size fraction to estimate bacterial respiration. But in fact phytoplankton are likely to contribute significantly to dark oxygen consumption in this size fraction. Respiration is strongly correlated with growth rate in phytoplankton. For phytoplankton with a specific growth rate of 0.7 d^{-1} typical of open ocean[7], a specific respiration of 0.4 d^{-1} is anticipated[8]. This estimate of phytoplankton respiration can be compared with a calculation of the specific bacterial respiration rate of 0.4 d^{-1} based on a growth rate[9] of 0.1 d^{-1} and growth efficiency[4] of 0.2. Given equal biomass of heterotrophic bacteria and picophytoplankton[6], it is likely that picophytoplankton account for about 50% of oxygen consumption in the <0.8–2-μm size fraction of oligotrophic ocean waters. As a consequence, heterotrophic bacterial respiration may have been overestimated by 200% in such waters. del Giorgio *et al.* compared their estimate of "bacterial" respiration with net photosynthesis. As the contribution of phytoplankton to respiration is already included in the net photosynthesis measurement[10], del Giorgio *et al.* could have counted the contribution of picophytoplankton to respiration twice in their analysis.

In their paper, del Giorgio *et al.* assumed a respiratory quotient (RQ) of 1.0 CO_2 evolved per O_2 consumed to convert bacterial respiration from oxygen to carbon equivalents. This RQ is unconstrained by observations for open ocean bacteria, but it is likely to be an overestimate of CO_2 evolution from respiration of organic matter that includes proteins and lipids in addition to carbohydrates. Oceanographers typically use a photosynthetic quotient of 1.25 O_2 evolved per CO_2 assimilated to convert measurements of primary production from carbon to oxygen equivalents[11] for comparison with respiration rates measured as O_2 consumption. Multiplying the 200% overestimate of heterotrophic bacterial respiration by a photosynthetic quotient of 1.25 leads to an error of 250%.

Cross-system analyses, such as that undertaken by del Giorgio and co-workers, are necessary for clarifying our understanding of the relationship between heterotrophic and photosynthetic metabolism in aquatic systems. However, the conclusion of net heterotrophy of unproductive oceanic systems needs to be tempered, given uncertainty in phytoplankton respiration rates or other possible systematic errors in bacterial respiration and primary productivity measurements.

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del Giorgio and Cole reply — Geider raises two main issues: first, he points to substantial evidence suggesting net autotrophy in surface ocean waters; and second, he argues that the estimates of bacterial respiration we used in our paper[1], which are based largely on measurements of oxygen consumption in size-fractionated samples, could overestimate the contribution of bacteria.

The surface waters of the ocean, taken as a whole, are probably net autotrophic, for the reasons Geider states as well as others. We were careful to state our conclusions about open ocean systems to reflect this view. In fact, we suggested several mechanisms, including temporal lags and export of detrital organic carbon from more productive marine areas, that would be consistent with both our analysis and the idea of oceanic surface waters being net autotrophic. Thus, we do not disagree with Geider about the net autotrophy in oceanic surface waters.

The second issue raised by Geider is whether the estimates of respiration are representative of the metabolic rates of heterotrophic bacteria in the open seas. Geider argues that they are not, because of the possible contribution of minute autotrophs to the small-size planktonic fraction. From the point of view of the organic carbon balance, it does not matter if some of the respiration is due to the contribution of autotrophs. If one accepts the rate measurements, the destruction of organic matter by respiration is still larger than the production by photosynthesis. Geider estimates autotrophic and heterotrophic picoplankton respiration based on typical growth and specific respiration rates of these organisms in the sea. As part of this calculation, he

uses the 20% bacterial growth efficiency that we report[1]. We derived this efficiency precisely from the respiration measurements which Geider criticizes in his comment.

If one accepts this 20% growth efficiency, as many do[2–5], then bacterial respiration is four times larger than bacterial production. Let us combine, then, this value of bacterial growth efficiency with published data on bacterial production and primary production for the world's oceans to derive an independent figure for bacterial respiration. The most comprehensive review is that of Ducklow and Carlson[6]. Using these data, the resulting bacterial respiration represents on average 102 and 110% of net primary production in open ocean and coastal marine systems, respectively. Stated more broadly, if bacterial production is ~30% of net primary production, as has been widely reported[7], when bacterial growth efficiency is 23% or lower, bacterial respiration will exceed primary production. Possibly net heterotrophy is more widespread in the oceans than is currently accepted, or bacterial growth efficiency is much higher than 20%, or most measurements of bacterial production are serious overestimates, relative to primary production.

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Lifetime of plasma cells in the bone marrow

Immune protection is based on long-lived memory cells and effector cells, which are either cytotoxic or secrete antibodies. The lifespan of these effector cells has not so far been determined. Here we show that antibody-secreting plasma cells from bone marrow are as long-lived as memory B cells.

After immunization with T-cell-dependent antigens, specific antibody titres are often stable for long periods. As the half-life of antibodies is short[1], the observed antibody titre must be maintained by antibody-secreting plasma cells. In the bone marrow, antigen-specific plasma cells can be detected for at least one year after immunization[2], but whether these plasma cells are continuously generated by B cells that are activated by persisting antigens, or whether they are long-lived, is not known[3]. Analysis of proliferation of total plasma cells in bone marrow over a 10-day period has suggested that most plasma cells may have a lifespan of only a few weeks[4]. In splenic foci, plasma cells that are derived from B cells after primary immunization die by apoptosis after a few days[5].

We investigated the lifespan of murine bone-marrow plasma cells in a secondary immune response to ovalbumin. We used cellular-affinity matrix technology[6] to identify, isolate and analyse IgG1 antibody-secreting cells (Fig. 1a). Such cells secrete ovalbumin-specific IgG1 antibodies when isolated and cultured (about 0.3–0.5 ng per cell per day; data not shown), and could be identified by intracellular binding of ovalbumin and low expression of surface immunoglobulins.

After secondary immunization with ovalbumin, the absolute number of these plasma cells in the bone marrow (about 5×10^{-4}, ~75,000 per mouse), as determined by flow cytometry, and the specific antibody titre (>1 mg per ml serum) remain constant for more than 3 months, whereas in the spleen, the number of plasma cells drops from a peak of 150,000 per

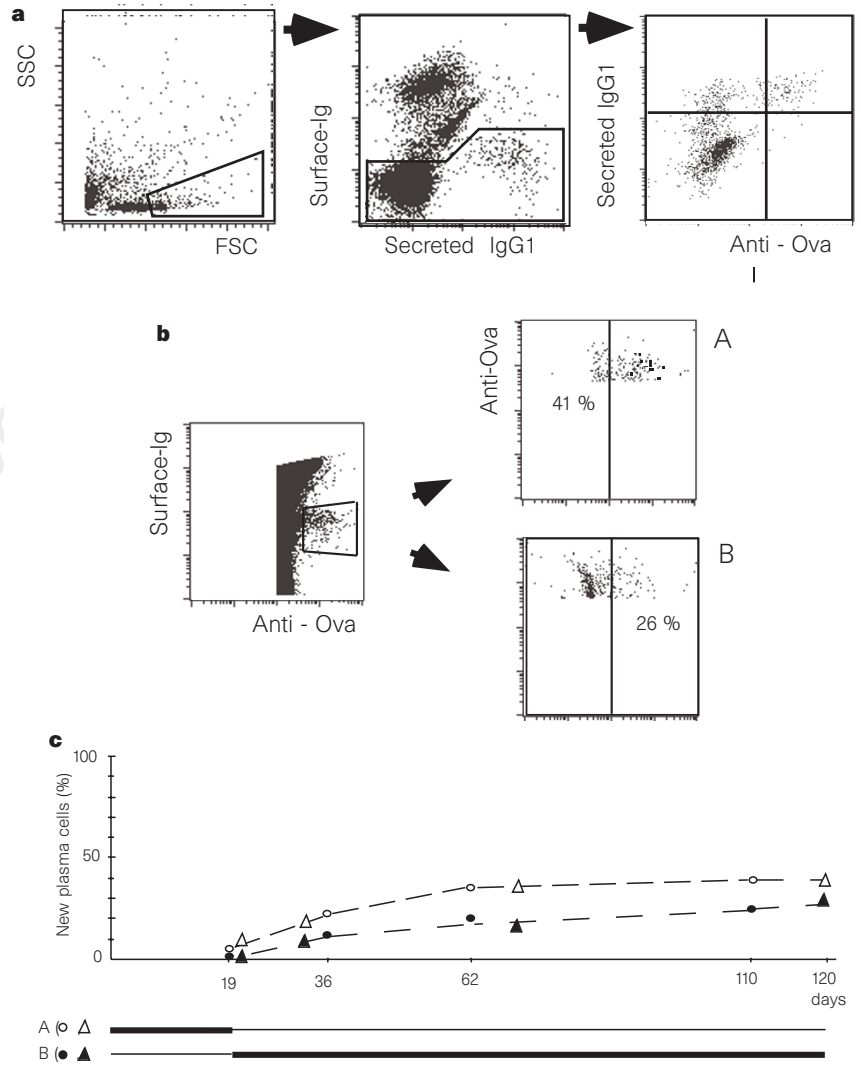

Figure 1 Lifetime analysis of antibody-secreting plasma cells. BALB/c mice were immunized with 0.1 mg alum-precipitated ovalbumin (Ova), and boosted 32 days later with 0.1 mg soluble ovalbumin. Femoral bone marrow of three mice was pooled for each stage of the analysis. **a,** Identification of IgG1-secreting, ovalbumin-specific plasma cells in the bone marrow as blast (forward scatter/side scatter, FSC/SSC), low surface immunoglobulin, high secreted IgG1, intracellular anti-ovalbumin-positive cells, 12 days after boost, with analysis gates as indicated. **b,** Analysis of BrdU incorporation as a measure of DNA synthesis by these bone-marrow plasma cells, 110 days after secondary immunization. BrdU incorporation was determined for group A and B plasma cells (see **c**), by staining with fluorescein-labelled anti-BrdU, with evaluation gates set according to controls (not shown). **c,** Replacement kinetics of ovalbumin-specific bone-marrow plasma cells after ovalbumin boost (day 0). BrdU feeding periods (1 mg per ml drinking water) are indicated by thickened bars. Frequencies of BrdU-negative (A) and BrdU-positive (B) bone-marrow plasma cells. Two experiments are indicated by circles and triangles. (Details of methodology are available from the authors on request).