## **SUPPLEMENT**

## Dead in the water: the vicious cycle of blanks during natural level <sup>14</sup>C manipulation of marine algal cultures

Stephanie Kusch, Albert Benthien, Klaus-Uwe Richter, Björn Rost, Gesine Mollenhauer

**Table S1**. Radiocarbon ( $\Delta^{14}$ C) and stable carbon isotope ( $\delta^{13}$ C) results of purified chlorophyll *a*, alkenones, and LMW alkanoic acids. Errors denote 1 $\sigma$  analytical uncertainties. Reproducibility is  $<2\sigma$  analytical uncertainties.

Sample	MODERN		IN	INTERMEDIATE			FOSSIL		
	δ <sup>13</sup> C [‰]	$\Delta^{14}$ C [‰]	ID <sup>#</sup>	δ <sup>13</sup> C [‰]	Δ <sup>14</sup> C [‰]	ID <sup>#</sup>	δ <sup>13</sup> C [‰]	Δ <sup>14</sup> C [‰]	ID <sup>#</sup>
E. huxleyi									
DIC pre-growth	$-1.0{\pm}0.1$	60.0±3.4	100689	$-14.5 \pm 0.1$	$-380.1\pm2.4$	100691	$-22.1\pm0.1$	-711.1±1.6	100693
DIC post-growth	$4.2{\pm}0.1$	83.0±3.5	100690	$-12.1\pm0.1$	$-437.1\pm2.0$	100692	$-13.1\pm0.1$	$-519.4\pm2.1$	100694
chlorophyll a	$-15.4{\pm}0.1$	24.5±10.6	89070	$-27.6\pm0.1$	$-463.9\pm2.0$	89071	$-22.5\pm0.1$	$-277.6\pm2.5$	89072
alkenones	$-20.2 \pm 0.1$	$77.0{\pm}3.9$	89073	$-35.0\pm0.1$	$-451.3\pm2.1$	89074	$-30.2\pm0.1$	$-280.3 \pm 2.5$	89075
T. pseudonana									
DIC pre-growth	$-14.8 \pm 0.1$	$-43.9\pm2.9$	100695	$-8.8\pm0.1$	$-135.7 \pm 3.0$	100697	$-16.5\pm0.1$	$-439.8\pm2.2$	100699
DIC post-growth	$-11.0\pm0.1$	$-505.2\pm2.5$	100696	$-2.4{\pm}0.1$	$-172.8 \pm 2.6$	100698	$-23.6\pm0.1$	$-843.1\pm1.3$	100700
chlorophyll a	$-27.8 \pm 0.1$	-452.3±1.9	89076	n.d.	$-357.0\pm42.8$	ETH 44554	$-34.7\pm0.1$	$-706.5 \pm 2.3$	89078
C14:0 alkanoic acid	$-30.2\pm0.1$	$-348.8 \pm 3.1$	89079	n.d.	$-490.5\pm37.2$	ETH 44552	n.a.	n.a.	
C16:0 alkanoic acid	n.a.	n.a.		n.d.	$-332.9\pm25.5$	ETH 44553	$-42.6\pm0.1$	$-653.3\pm2.0$	89082
C16:1 alkanoic acid	n.a.	n.a.		n.a.	n.a.		n.d.	$-689.6\pm23.7$	ETH 44555

<sup>#</sup> NOSAMS ID number; IDs with ETH prefix represent ultra-small samples analyzed at ETH Zürich.

n.a. Not analyzed.

n.d. Not determined.

Sample	MODERN	INTERMEDIATE	FOSSIL
E. huxleyi			
DIC pre-growth	2.04	2.05	1.82
DIC post-growth	0.82	1.25	0.81
biomass	735,300	366,900	584,600
T. pseudonana			
DIC pre-growth	2.04	2.14	2.24
DIC post-growth	1.34	1.36	1.82
biomass	n.d.*	n.d. *	n.d. *

Table S2. DIC concentrations (mmol/kg) and final cell biomass (cells/ml) in the *E. huxleyi* and *T. pseudonana* cultures.

\* Could not be determined with the Coulter Counter.



Fig S1. Chromatogram of the purified 'Modern'  $C_{14:0}$  alkanoic acid fraction. GC-based compound purity is 98.6%.

3



**Fig S2**. Chromatogram of the purified 'Modern' alkenones fraction. GC-based compound-fraction purity is 98.3%.



**Fig S3**. Chromatogram of the 'Modern' chlorophyll *a* fraction obtained from *E*. *huxleyi* after the first LC purification step.



Fig S4. Chromatogram of the purified 'Intermediate'  $C_{14:0}$  alkanoic acid fraction. GC-based compound purity is 99.9%.



**Fig S5**. Chromatogram of the purified 'Intermediate'  $C_{16:0}$  alkanoic acid fraction. GC-based compound purity is 99.9%. Note that the signal at approximately 35 min is not a peak, but an artefact caused by a detector fault.



Fig S6. Chromatogram of the purified 'Intermediate' alkenones fraction. GC-based compound-fraction purity is 99.8%.



**Fig S7**. Chromatogram of the 'Intermediate' chlorophyll *a* fraction obtained from *E. huxleyi* after the first LC purification step.



Fig S8. Chromatogram of the purified 'Fossil'  $C_{16:0}$  alkanoic acid fraction. GC-based compound purity is 97.3%.



Fig S9. Chromatogram of the purified 'Fossil'  $C_{16:1}$  alkanoic acid fraction. GC-based compound purity is 99.4%.



**Fig S10**. Chromatogram of the purified 'Fossil' alkenones fraction. GC-based compound-fraction purity is 99.8%.



**Fig S11**. Chromatogram of the 'Fossil chlorophyll *a* fraction obtained from *E*. *huxleyi* after the first LC purification step.