Supplementary Materials for

Unicellular Cyanobacterium Symbiotic with a Single-Celled Eukaryotic Alga

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Supplementary Materials:

Figure S1: Flow cytograms and target cell populations for all samples used in this study. Red fluorescence is measured at 692-40 nm after excitation with a 488 nm laser and is a proxy for chlorophyll *a* content and forward scatter (FSC) is a proxy for cell size. All samples are unpreserved seawater except for (F), which is seawater concentrated 100X by vacuum filtration then preserved by freezing in liquid nitrogen for storage before analysis by flow cytometry. 3µm beads (B) were used for reference in some samples. Note the appearance of the dislodged UCYN-A (U) in the flow cytogram from concentrated seawater (F) and the absence of the UCYN-A population in un-concentrated water from the same sample (E). Other cell populations indicated are photosynthetic picoeukaryotes (PPE), *Synechococcus* (*Syn.*), and *Prochlorococcus* (*Pro.*). Light blue coloring indicates a greater density of events of a cell population. See Supplementary Table 1 for additional information on each sample and use in the experiments described in this study.

Figure S2: Presence of UCYN-A genome fragments in metagenomes prepared from sorted picoeukaryotes from three samples (T39, T41, and T60) from two stations (STB7 and STB11) from the South East Pacific Ocean (*14, 42*). Using BLASTN, numerous reads matching the UCYN-A genome at 99-100% similarity were found only at sample T60 (STB11) (Table S2). Notably, the partner 18S rRNA gene sequences were absent at Station STB7 and hits to the UCYN-A genome were relatively low (90-95% similar) with best hits to distantly related organisms such as *Cyanothece* and *Prochlorococcus* (Table S2).

Table S1: Sources, experiments conducted, and UCYN-A *nifH* quantification for samples utilized in this study. Experiments conducted: (A) UCYN-A *nifH* screens of sorted cell populations, (B) ^{15}N and ^{13}C incubations and HISH-SIMS, (C) 18S universal rRNA gene PCR from single and entire-population picoeukaryotes. Population abbreviations are *Prochlorococcus* (*Pro*) and photosynthetic picoeukaryotes (PPE).

Table S2: UCYN-A representation in metagenomes from sorted photosynthetic picoeukaryotes from two BIOSOPE stations.

Table S3: BLASTn hits of18S and 16S rRNA gene sequences derived from sorts of the entire picoeukaryote population from samples KM1110 (Accession numbers JX291805 - JX291865) (A) and HOT234 (Accession numbers JX291866 - JX291959) (B).

Table S4: Numbers and best BLASTn identity of 18S rRNA gene sequences derived from nested PCR of single *nifH*-positive picoeukaryotes from HOT234 and KM1110. Cruises are KM1110 (5 m March 2011, accession numbers JX291679 - JX291804) and HOT234 (79 m August 2011, accession numbers JX291547 - JX291678). Asterisk (*) indicates the only marine species amplified. All others are suspected contaminants from terrestrial sources. Full species names are *Chrysochromulina acantha, Pinus armandii, Pinus luchuensis, Pinus morrisonicola, Lithocarpus rufovillosus, Ralstonia solanacearum, and Cunninghamia lanceolata.*

Table S5: The UCYN-732 and Helpers A and B oligonucleotide (5' to 3') shown with other closely related free-living and symbiotic cyanobacterial sequences. Mismatches are highlight in red.

Table S6: Summary of cell dimensions and nanoSIMS analyses for UCYN-A and partner cells measured by nanoSIMS.

Supplementary Methods and Materials

1. *Environmental sampling:* Seawater samples were collected by 24-Niskin bottle CTD rosette at the hydrographic Station ALOHA in the North Pacific Ocean (*35*) during cruises KM1110 in March 2011 (25 meter depth), HOT234 (79 meter depth) in August 2011, and HOT239 (25 meter depth) in January 2012 and at a coastal site off the island of Hawai'i (20 2.047' N 155 57.394' W) on cruise KOK 11-15 (40 meter depth) in December 2011 (Table S1).

2. *Cell sorting:* All cell sorting was performed with a BD Biosciences Influx Cell Sorter equipped with a 488 nm laser (Sapphire Coherent), 70 µm diameter nozzle, and using BioSure Sheath fluid (BioSure, Grass Valley, CA USA) at 1X concentration. Sorting took place in a laboratory van equipped for flow cytometry either on board the ship (HOT234), on land directly following the cruise (KM1110), or on land following incubation in an on-deck flow-through seawater incubator and expedited shipping to California (HOT239 and KOK 11-15). All seawater samples were pre-filtered with a 50 µm filter (Partec Celltrics, Swedesboro, NJ USA) prior to sorting to prevent large particles from clogging the nozzle. Cells were sorted by gating on red fluorescence (692-40 nm) and forward scatter (FSC) using the BD cytometry software programs Spigot and FACS Sortware. The sort mode of "1.0 Drop Purity" was employed to ensure pure sorting of target populations. FlowJo (Tree Star, Ashland OR USA) was used to analyze cell counts and create dot plots (Fig. 1, Fig. S1).

3. *UCYN-A nifH screening of sorted cell populations:* Sorted cells were screened for UCYN-A nitrogenase (*nifH*) by qPCR using the UCYN-A-specific *nifH* primer/probe Taqman assay (Applied Biosystems, Carlsbad, CA USA) (*36*) for 45 cycles in 25 µL reactions. Three cell populations were screened for UCYN-A *nifH* including photosynthetic picoeukaryotes (PPE) (50-200 cells per sort replicate), *Prochlorococcus* (*Pro.*) (5,000 cells per sort replicate), and cells not PPE and not *Prochlorococcus* (at least 1,000 cells per replicate). At least four replicate sorts were analyzed by qPCR for each population and sample. Cells were sorted directly into Fast Step qPCR strips (Applied Biosystems) with 10 µL of 5 kD filtered water for direct use in qPCR following addition of the qPCR reagents on a StepOne qPCR machine (Applied Biosystems). No amplification of *nifH* in *Prochlorococcus* cell sorts indicated no *nifH* contamination of reagents or materials used. We relied on high temperatures in the first stages of qPCR to lyse cells rather than a DNA extraction step, which would have resulted in loss of material.

4. *Entire picoeukaryote population PCR with universal 18S/16S rRNA gene primers:* Sorts of the entire picoeukaryote population were carried out for each sample to assess the 18S and 16S rRNA gene diversity of the picoeukaryote population that was targeted in this study. Duplicate samples of 2500 cells (HOT234) and 500 cells (KM1110) were sorted into 10 µL 5 kD filtered water and amplified with 18S rRNA gene universal primers Ek555F and Ek1269R (*37*) in 50 µL reactions for 35 cycles using Platinum Taq reagents (Invitrogen, Grand Island, NY USA). PCR products were run on a 1% agarose gel for 90 minutes at 90V. Gel bands from approximately 500 bp and 700 bp positions were excised from the gel extracted using the QiaQuick Gel Extraction Kit (Qiagen).

   Purified PCR products were cloned using the pGEM-T Easy Kit (Promega, Madison, WI USA) following manufacturer's protocols and plasmid preps were performed using the Montage Plasmid MiniprepHTS Kit (Millipore, Billerica, MA USA). The UC Berkeley DNA Sequencing Facility (http://mcb.berkeley.edu/barker/dnaseq/) carried out Sanger sequencing. Sequences were checked for quality in Sequence Scanner (Applied Biosystems) and BLASTn was run against BLAST database nr (includes all GenBank, EMBL, DDBJ, and PDB sequences) in April 2011 to identify the sequences. Numerous 16S rRNA sequences (~500 bp) were picked up in addition to the 18S rRNA gene sequences (~730 bp) targeted by the primer set and these were included in analysis and confirmed the presence of UCYN-A in the photosynthetic picoeukaryote population (Table S3).

5. *UCYN-A nifH screening and nested PCR of single picoeukaryotes with 18S rRNA gene universal primers:* To identify the specific partner cell of UCYN-A (from the diverse picoeukaryote population) single picoeukaryotes from KM1110 (5 m) and HOT234 (79 m) were sorted into 10 µL of 5 kD water in 72

wells of 96-well qPCR plates. 24 wells were left empty for qPCR standards and no template controls (NTC). Plates were covered with AluminaSeal (Diversified Biotech, Dedham, MA USA) and stored at -80°C until processing. Whole plates were thawed at room temperature then screened for UCYN-A *nifH* as above.

Thirty single cell sorts with UCYN-A *nifH* gene copy of approximately 1 were selected for use in the nested PCR with 18S rRNA gene universal primers. The entire volume of the qPCR *nifH*-positive wells were used as template in 100 µL reactions with Platinum Taq reagents (Invitrogen). First, universal primers EukA/EukB (*38*) were applied for 35 cycles then 1 µL of the EukA/B PCR product used as template in 25 µL reactions with internal 18S rRNA gene primers 555F/1269R (*37*). Gel bands were extracted, purified, cloned (20-30 clones were picked for each single cell), and sequenced as above. For KM1110 samples, universal 16S rRNA gene primers (27F/1492R) and an internal unicellular diazotroph specific primer set (cya359F/nitro821R) (*39*) were applied to a subset of UCYN-A *nifH* positive single cells. All cya359F/nitro821R amplicons were identical to the UCYN-A 16S rRNA gene.

Sequences were compared to BLAST database nr (April 2011) by BLASTn for identification (Table S4). The great majority of sequences matched either the environmental prymnesiophyte clone from BIOSOPE T60.34 or *Pinus armandii* (Chinese White Pine). A few sequences matched other terrestrial tree species (Table S4). We believe the tree sequences are contaminants that are amplified when PCR template is in very low concentration (as in these single cell sorts) as no such tree sequences were derived from 18S rRNA gene clones libraries that were made from hundreds of sorted cells to assess the diversity of the entire picoeukaryote population (described above and Figure 2). Thus, we do not think they are an abundant sequence in our samples, but are due to reagent contamination acquired during sorting or PCR. To test this further, nested PCR with 18S rRNA gene universal primers (EukA/B) was applied to 5 kD-filtered water or *nifH*-negative cell sorts. *P. armandii* sequences were present only in the reactions made from the *nifH*-negative cell sorts, indicating that the pine sequences are a contaminant from sorting (likely in the sheath fluid or dust) rather than from PCR reagents. The only other marine sequence found in the 18S rRNA gene nested PCR sequences besides the BIOSOPE T60.34 sequence is from the prymnesiophyte *Chrysochromulina acantha.* This single sequence came from the clone library of one single cell sort from HOT234 that also contained *P.armandii* and BIOSOPE T60.34 sequences (sort ID 11D12,Table S4). Because we did not find this sequence in any other of the 30 single cell sorts, we presume that it is DNA contamination from the seawater sample amplified when template is in low concentration, rather than a specific associate of UCYN-A, as it did not appear in the clone library for the entire picoeukaryote population from samples HOT234 (Figure 2, Table S3).

6. *Prymnesiophyte 18S rRNA gene phylogeny*: Determining the phylogeny of the UCYN-A partner sequence was accomplished by constructing a tree of the full length 18S rRNA gene sequences of the UCYN-A partner best nucleotide hit (BIOSOPE T60.34, FJ537341, Uncultured *Chrysochromulina* clone) and selected cultured and environmental prymnesiophyte sequences (Fig. 2). Sequence alignments were created using SINA (v1.2.9) online with the SILVA SEED for the reference alignment (*40*). PhyML 3.0 was used to construct a maximum likelihood tree (*41*) using the HKY85 substitution model and bootstrapped with 100 replicates. Outgroups were chosen as in Cuvelier et al. (*16*) and included *Thalassiosira weissflogii* (AY485445), *Chlamydomonas reinhardtii* (AY665726), *Rhodomonas salina* (EU926158), *Compsopogon coeruleus* (AF342748), and *Chondrus crispus* (Z14140). Other species included in the tree (Fig. 2) are: *Pavlova sp.* CCMP1416 (AJ243369), *Phaeocystis globosa* (EU077556), *Phaeocystis pouchetii* isolate P360 (AF182114), *Phaeocystis antarctica* Karsten SK23 (X77481), *Phaeocystis jahnii* (AF163148), *Pleurochrysis sp.* CCMP 875 (AJ246265), *Reticulosphaera socialis* (X90992), Coccoid haptophyte CCMP 625 (U40924), *Isochrysis galbana* (ZJ246266), *Emiliania huxleyi* (AF184167), *Chrysochromulina campanulifera* strain J10 (AJ246273), *Chrysochromulina strobilus* (FN599060), *Chrysochromulina cymbium* (AM491018), *Chrysochromulina leadbeateri* (AM491017), *Chrysochromulina scutellum* (AJ246274), *Chrysochromulina sp.* NIES-1333 (DQ980478), *Chrysochromulina simplex* (AM491021), *Chrysochromulina parva* (AB601109), *Prymnesiophyte symbiont* 1 (AF166377), *Chrysochromulina acantha* strain T20 (AJ246278), *Chrysochromulina*

trondsenii K11 (AJ246279), *Chrysochromulina sp.* MBIC10513 (AB199882), *Chrysochromulina sp.* LKM-2007-1 (AM491020), *Chrysochromulina rotalis* (AM491025*), Chrysochromulina spinifera* (AB601108), *Chrysochromulina sp.* CCMP1204 (AM491016), *Chrysochromulina brevifilum* strain Kawachi (AM490995), *Chrysochromulina brevifilum* MBIC10518 (AB058358), *Chrysochromulina parkeae* (AM490994) , *Haptolina hirta*‡ (A5246272), *Haptolina ericina*‡ (AM491030), *Haptolina fragaria*‡ (AM491013), *Haptolina herdlensis*‡ (AM491011), *Haptolina brevifila PML* (AM491012), *Prymnesium chiton*° (AM491029), *Prymnesium minor*° (AM491010), *Prymnesium kappa*° (AJ246271), *Prymnesium polylepis*° (AJ004866), *Prymnesium* sp. UIO133, *Prymnesium patelliferum* (L34671)*, Braarudosphaera bigelowii*, Funahama-T3 (AB478412), *Braarudosphaera bigelowii*, TP05-6-b (AB250785), *Braarudosphaera bigelowii*, TP05-6-a (AB250784), *Braarudosphaera bigelowii* Furue-15 (AB478413), *Braarudosphaera bigelowii* Yatsushiro-1 (AB478414), Genus names are changed to *Prymnesium* for species marked with (°) and to *Haptolina* for species marked with (‡) as in Edvardsen *et al.* (15) .

7. *Whole genome amplification of BIOSOPE sorted picoeukaryotes and comparison to UCYN-A genome:* Samples from Station B7 (STB7 - samples T39 and T40) and Station B11 (STB11- sample T60) were collected by tangential flow concentration, which is much less disruptive than other concentration methods, and cell sorting and DNA was extracted as described previously (*14*). Whole genome amplification was performed as described before (*42*). Each sample (T39, T40, and T60) was sequenced on a single 454 Titanium run (*34*). Raw reads were trimmed as described (*34*) and BLASTed (BLASTN) against the genome of UCYN-A (NC_013771) using a maximum e-value of e^{-100}. Raw reads were also mapped against the genome of UCYN-A using the software Geneious (http://www.geneious.com/) with the Medium-Low Sensitivity (Table S2).

8. *^{15}N and ^{13}C incubations, HISH-SIMS, calculations:* Seawater for isotope incubations was collected from 25 m during cruise KM1110 at Station ALOHA (22° 45'N, 158° 00'W). Water was pre-filtered through 10μm nylon mesh (Nitex 03-10/2) to remove larger diazotrophs (i.e. *Trichodesmium*, diatom-*Richelia* symbioses). *Crocosphaera nifH* and UCYN-A *nifH* gene abundances measured by qPCR were 5,752 and 183,569 copies L^{-1}, respectively. Water was dispensed into 4 acid-cleaned and sterile 500 mL polycarbonate bottles (Nalgene). Two bottles were left un-amended as controls and two bottles were amended with 125 μL of 500 mM ^{13}C-bicarbonate (Cambridge Isotopes, Andover, MA USA) and 1.5 mL $^{15}N_2$ (Cambridge Isotopes) at 21:00 hours. All bottles were shaken to dissolve the $^{15}N_2$ bubbles then were immediately placed in an on-deck incubator temperature-regulated by continuously flowing surface seawater and shaded to approximately 30% of photosynthetically active radiation (PAR) for 36-hours before processing by flow cytometry for HISH-SIMS measurements and filtration for bulk isotope measurements.

For HISH-SIMS analysis, 15,403 cells from the picoeukaryote sort region (Fig. S1) were sorted into separate tubes containing 2% paraformaldehyde (PFA) prepared in sterile-filtered seawater. After fixation for at least one hour at room temperature, sorted samples were divided into triplicates and applied by pipette to a small area (5 mm) marked by an ink circle in triplicate on gold-palladium sputtered 0.2 μm filters. Gentle vacuum pressure (<5 psi) was used to draw the cells onto the filter. Cells on the filter were then rinsed with 1X Phosphate Buffered Saline (PBS) applied by pipette and gentle vacuum pressure, and allowed to dry. Filters were folded gently and placed into sterile microfuge tubes and stored at -20°C until further processing for the HISH-nanoSIMS.

A previously described HISH assay was applied to the PFA-fixed flow sorted picoeukaryote population samples with minor modifications (*28*). A highly specific oligonucleotide probe mixture and assay was developed based on the 16S rRNA gene of UCYN-A (CP001842 FJ170277). A mixture of a 5′-horseradish peroxidase (HRP)-labelled oligonucleotide UCYN-732 probe and helper A and B oligonucleotides were used and followed by the deposition of fluorine-containing tyramides (Oregon Green 488 dissolved in dimethylformamide containing 20 mg mL^{-1} 4-iodophenyl boronic acid) (Table

S6). Helper probes A and B (Table S5) were used to increase the accessibility and subsequent intensity of the UCYN-732 probe. A non-probe (NON338) (*43*) was used as a negative control.

The 5 mm ink-marked circles were excised with a knife and the cells were embedded in 0.1% low gelling point agarose to avoid cell loss. The cell wall was permeabilized at 37°C with a lysozyme solution (10 mg mL^{-1} in 0.05 M EDTA, pH 8.01, 0.1 M Tris-HCl, pH 7.5; Fluka, Taufkirchen, Germany). After permeabilization, filter samples were washed with ultrapure water (MQ, Millipore) and transferred to a 0.01 M HCl solution for 10 min at room temperature (RT) in order to bleach endogenous peroxidases. Hybridization conditions were as follows: 8.5 h at 35°C in a hybridization buffer containing: 0.9 M NaCl, 40 mM Tris-HCl (pH 7.5), 10 % dextran sulfate (wt/vol), 0.01 % (wt/vol) sodium dodecyl sulfate (SDS), 50% formamide (Fluka) (vol/vol), 10 % (wt/vol) Blocking Reagent (Boehringer, Mannheim, Germany), 1X Denhard's reagent, 0.26 mg mL^{-1} sheared salmon sperm DNA (Ambion), 0.2 mg mL^{-1} yeast RNA (Ambion). Post-hybridization, samples were incubated at 37°C for 15 min in pre-warmed (37°C) washing buffer containing 20 mM NaCl, 5 mM EDTA (pH 8.0), 20 mM Tris-HCl (pH 7.5), and 0.01 % SDS and subsequently transferred to 1X PBS (pH 7.6) for 20 min. Under dark conditions and at 46°C, a tyramide signal amplification (TSA) solution (5 % 20 x PBS; 2 M NaCl; 0.1 % blocking reagent; 10 % dextran sulfate; 0.0015 % (vol/vol) H_2O_2, 1 % Oregon Green 488 tyramide (Molecular Probes, Leiden, The Netherlands), 1 µL of tyramide solution) was applied to the samples for 20 min. Samples were rinsed twice at RT in 1X PBS (pH 7.6) and MQ water for 15 min each rinse. Cells were air dried, and stained with 1 µg mL^{-1} 4',6'-diamidino-2-phenylindole (DAPI) for 10 min at RT in the dark, then washed three times in MQ water and air dried. The total number of UCYN-A cells was enumerated on 2 of the 3 replicate samples prior to nanoSIMS analysis. All hybridized UCYN-A cells were counted as associated with partner cells or as dislodged. In addition, the partner cells, which were associated with 1, or 2 UCYN-A cells were further enumerated. For sample 1, 259 total UCYN-A cells were counted, with 83 (32%) UCYN-A cells dislodged, 166 (64%) UCYN-A cells were in association with 1 partner cell, and 5 partner cells (or 10 (4%) UCYN-A cells) were found with 2 UCYN-A cells attached. For sample two 278 UCYN-A cells were counted, and 107 (38 %) UCYN-A cells were dislodged, 163 (59 %) UCYN-A cells were in association with 1 partner cell, and 4 partner cells (or 8 (3%) UCYN-A cells) were found with 2 UCYN-A cells.

After enumeration, areas on the filter sections with UCYN-A cells were marked with arrows and numbers using a Laser Microdissection (LMD) Microscope 6500 (Leica, Berlin, Germany) fitted with appropriate filter set for the Oregon Green 488 tyramides (excitation max 498nm). Subsequently, the filters were mounted on a new glass slide coated with a 4:1 (v/v) embedding solution (low fluorescence glycerol mountant (Citifluor AF1, Citifluor Ldt, London, United Kingdom) and mounting fluid Vecta Shield (Vecta Laboratories, Burlingame, CA USA) and examined with an Axioplan II microscope (Carl Zeiss, Jena, Germany) fitted with the appropriate filter sets for the Oregon Green 488 tyramides and for DAPI (excitation max 390 nm). Microscopic pictures were taken and used for orientation purposes during subsequent nanoSIMS analysis and for post-processing using *look@nanosims* software (*44*).

NanoSIMS analysis was performed using a Cameca NanoSIMS 50L instrument. Ten individuals cells were analysed which were found in association with 1-2 UCYN-A cells, and 6 unattached UCYN-A cells were also measured. Analysis time, including tuning of detectors, was equivalent to 150h. Carbon (C), fluorine (F), nitrogen (as CN) and sulfur (S) isotopes (^{12}C, ^{13}C, ^{19}F, $^{12}C^{14}N$, $^{12}C^{15}N$ and ^{32}S) were measured simultaneously in raster imaging mode. Sample surfaces were rastered with a 16 keV Cesium (Cs^+) beam and a current between 25-35 pA. Primary ions were focused into a nominal ~50 nm spot diameter. The primary ion beam was used to raster the analyzed area with 2000 counts per pixel over the chosen raster size and a dwelling time of 1 or 3 ms per pixel. Areas ranged in size from 5 x 5 to 20 x 20 μm^2 depending on the distribution of the targeted cells (most areas were 10 x 10 μm^2). Negative secondary ions were collected simultaneously in electron multiplier detectors. Prior to analysis, the area was pre-sputtered for 1-2 min with a high-current Cs^+ beam to implant Cs and remove surface contaminants.

All scans were corrected for drift of the beam and sample stage after acquisition. Isotope ratio images were created as the ratio of a sum of total counts for each pixel over all recorded planes (40-100

planes) of the investigated isotope and the main isotope. Regions of interest (ROIs) around cell structures and cell diameter were manually circled and calculated using *look@nanosims* software (*44*).

The biovolume (V) of the UCYN-A and the associated cells were calculated according to the volume of a sphere:

$$V = (\pi/6) \times Ø^3 \quad (1)$$

where Ø is the cell diameter, and π is 3.14. The cell diameter was determined by the length of the ROI using the look@nanoSIMS software. As previously published (*45*), the carbon (C) content per cell was estimated by:

$$\text{Log } [C] = -0.363 + (0.863 \times \text{Log } (V) \quad (2)$$

The C content per cell (C_{con}) was converted into N content per cell (N_{con}) based on conversion factors provided by Tuit et al. (*46*) assuming a modified Redfield ratio (C:N) of 8.6. The C_{con} and N_{con} represent the initial C and N content. The isotopic ratios ($R_C = {}^{13}C/{}^{12}C$ and $R_N = {}^{15}N/{}^{14}N$) based on ROI selections and nanoSIMS analysis were used to calculate atom percent (AT %) enrichment of ^{13}C or ^{15}N by:

$$A_C = R_C/(1+R_C) \times 100 \quad (3)$$

$$A_N = R_N/(1+R_N) \times 100 \quad (4)$$

where A_C is the atom (AT) % ^{13}C and A_N is the AT % ^{15}N. The cell specific C and N_2 assimilation (F_C or F_N) was calculated for by:

$$F_C = ({}^{13}C_{ex} \times C_{con})/C_{SR} \quad (5)$$

$$F_N = ({}^{15}N_{ex} \times N_{con})/N_{SR} \quad (6)$$

where ${}^{13}C_{ex}$ and ${}^{15}N_{ex}$ are the ${}^{13}C/{}^{12}C$ and ${}^{15}N/{}^{14}N$ ratio of the individual ROIs corrected for by the mean ${}^{13}C/{}^{12}C$ and ${}^{15}N/{}^{14}N$ ratios in time zero samples divided by 100. The initial time zero ratios (${}^{13}C/{}^{12}C$ and ${}^{15}N/{}^{14}N$) were measured on bulk particulate samples (seawater collected onto a combusted GFF from the same depth of experiment) by a standard PDZ Europa ANCA-GSL elemental analyzer interfaced to a PDZ Europa 20-20 isotope ratio mass spectrometer (Sercon Ltd., Cheshire, UK). The bulk measurements were made at the stable isotope facility of University of California, Davis. The C_{SR} and N_{SR} are the calculated AT % of ^{13}C or ^{15}N in the experimental bottle and the C_{con} and N_{con} are described above (equation 2). The assimilated N or C was then divided by incubation time to determine cell-specific C and N_2 fixation rates. The percentage of fixed N transferred to the eukaryotic partners was determined by dividing the N assimilated into the associated cell as calculated above by the sum of N assimilated into the UCYN-A and the associated cell and multiplying by 100. Similarly, the percentage of C fixed and transferred to the UCYN-A cell was determined by dividing the C assimilated by UCYN-A as described above by the sum of C assimilated by the UCYN-A and the associated cell and multiplying by 100.

Table S1. Sample sources, experiments conducted, and UCYN-A *nifH* quantification for samples utilized in this study. Experiments conducted: (A) UCYN-A *nifH* screens of sorted cell populations, (B) ^{15}N and ^{13}C incubations and HISH-SIMS, (C) 18S universal rRNA gene PCR from single and entire-population picoeukaryotes. Population abbreviations are *Prochlorococcus* (Pro) and photosynthetic picoeukaryotes (PPE). 'NA' indicates the absence of quantitative measurements required for calculations.

Cruise	Date	Location	Depth	Experiments conducted	UCYN-A *nifH* detected in PPE?	%PPE with UCYN-A *nifH*	UCYN-A *nifH* gene copies per mL (PPE)	UCYN-A *nifH* gene copies per mL (*Pro*)	UCYN-A *nifH* gene copies per mL (not *Pro,* not PPE)	% Total UCYN-A *nifH* from PPE
KM1110	Mar-11	Station ALOHA	5m	(A), (C)	Yes	20.6±4.5	NA	NA	NA	NA
KM1110	Mar-11	Station ALOHA	25m	(A), (B)	Yes	NA	NA	NA	NA	NA
HOT234	Aug-11	Station ALOHA	79m	(A), (C)	Yes	109.8±34.0	1267.7(±392.1)	0	469.6(±354.6)	73.0
KOK 11-15	Dec-11	Coastal Hawai'i	40m	(A)	Yes	39.4±8.6	403.8(±88.4)	0	229.6(±81.0)	63.8
HOT239	Jan-12	Station ALOHA	25m	(A)	Yes	23.5±11.5	256.2(±125.8)	0	14.2(±14.0)	94.7

Table S2 : UCYN-A representation in metagenomes from sorted photosynthetic picoeukaryotes from two BIOSOPE stations.			
Sample code	**T39**	**T41**	**T60**
Station	STB7	STB7	STB11
Longitude	-120.38	-120.38	-107.29
Latitude	-22.05	-22.05	-27.77
Depth	175	40	0
Number of reads	1,054,607	1,381,023	1,154,137
Number of reads matching UCYN-A genome			
- BLASTN hits P < e-100 and > 95% ID	0	1	730
Reads assembling to UCYN-A genome			
- Number of reads assembled	4	10	980
- % identity	90.8%	94.5%	99.7%
- genome coverage bp	444	820	178,405
- genome coverage %	0.0%	0.1%	12.4%

Table S3. BLASTn hits of18S and 16S rRNA gene sequences derived from sorts of the entire picoeukaryote population from samples KM1110 (Accession numbers JX291805 - JX291865) (A) and HOT234 (Accession numbers JX291866 - JX291959) (B).

(A) 18S rRNA gene hits

Cruise ID	Hit ID	Hit Description	# Hits	Category
KM1110	gb|AF290085.2|	Uncultured marine diatom 48-5-EKD54 18S ribosomal RNA gene partial sequence	4	Bacillariophyta
KM1110	gb|L33449.1	Mytilus californianus 18S small subunit ribosomal RNA gene complete sequence	1	Bivalvia
KM1110	gb|AY749516.1|	Uncultured eukaryote clone H30.5 18S ribosomal RNA gene partial sequence	1	Centrohelea
KM1110	gb|EF622558.1|	Chlorarachniophyceae sp. RCC337 small subunit ribosomal RNA gene partial sequence; nucleomorph	4	Chlorarachniophyceae
KM1110	gb|EF172998.1|	Uncultured eukaryote clone SSRPE02 18S ribosomal RNA gene partial sequence	2	Chrysophyceae
KM1110	gb|AY046860.1|	Uncultured eukaryote isolate C3_E031 18S ribosomal RNA gene partial sequence	4	Chrysophyceae
KM1110	gb|FJ537322.1|	Uncultured marine chrysophyte clone Biosope_T39.120 18S ribosomal RNA gene partial sequence	3	Chrysophyceae
KM1110	gb|FJ537350.1|	Uncultured marine chrysophyte clone Biosope_T65.146 18S ribosomal RNA gene partial sequence	2	Chrysophyceae
KM1110	gb|FJ537356.1|	Uncultured marine chrysophyte clone Biosope_T84.071 18S ribosomal RNA gene partial sequence	1	Chrysophyceae
KM1110	gb|AY129063.1|	Uncultured marine eukaryote clone UEPAC48p3 18S small subunit ribosomal RNA gene partial sequence	1	Chrysophyceae
KM1110	gb|EU247836.1|	Pedinellales sp. CCMP2098 18S ribosomal RNA gene partial sequence	3	Dictyochophyceae
KM1110	gb|EU500103.1|	Uncultured eukaryote clone hotxp1a10 18S ribosomal RNA gene partial sequence	1	Dictyochophyceae
KM1110	gb|AY426832.1|	Uncultured marine eukaryote clone BL000921.5 18S ribosomal RNA gene partial sequence	1	Dictyochophyceae
KM1110	gb|GQ344723.1|	Uncultured marine eukaryote clone cRFM1.39 18S ribosomal RNA gene partial sequence	**3**	Dictyochophyceae
KM1110	gb|GQ382424.1|	Uncultured marine eukaryote clone MO010_1.00340 18S ribosomal RNA gene partial sequence	1	Dictyochophyceae
KM1110	gb|EU780594.1|	Uncultured eukaryote clone AMT15_1B_25 18S ribosomal RNA gene partial sequence	1	Dinophyceae
KM1110	gb|EU106739.1|	Stramenopile sp. RCC853 18S ribosomal RNA gene partial sequence	1	Pinguiophyceae
KM1110	gb|GQ913175.1|	Uncultured eukaryote clone 111.2.81 18S ribosomal RNA gene partial sequence	1	Pinguiophyceae
KM1110	emb|AM491010.2|	Chrysochromulina minor partial 18S rRNA gene strain PLY 304	2	Prymnesiophyceae
KM1110	emb|AJ402351.1|	eukaryote clone OLI11056 18S rRNA gene	1	Prymnesiophyceae
KM1110	gb|FJ537341.1|	Uncultured Chrysochromulina clone Biosope_T60.034 18S ribosomal RNA gene partial sequence	37	Prymnesiophyceae
KM1110	gb|EF172993.1|	Uncultured eukaryote clone SSRPD92 18S ribosomal RNA gene partial sequence	1	Prymnesiophyceae
KM1110	gb|HM581630.1|	Uncultured marine eukaryote clone EN351CTD040_09Apr01_20m 18S small subunit ribosomal RNA gene partial sequence	2	Prymnesiophyceae
KM1110	gb|HM581628.1|	Uncultured marine eukaryote clone EN351CTD040_16_09Apr01_4m 18S small subunit ribosomal RNA gene partial sequence	5	Prymnesiophyceae
KM1110	gb|HM581603.1|	Uncultured marine eukaryote clone FS04R13_10_27Feb07_75m_sort 18S small subunit ribosomal RNA gene partial sequence	1	Prymnesiophyceae
KM1110	gb|EF539131.1|	Uncultured marine eukaryote clone MB07.26 18S ribosomal RNA gene partial sequence	1	Prymnesiophyceae
KM1110	gb|HM581615.1|	Uncultured marine eukaryote clone OC413BATS_P053_15m 18S small subunit ribosomal RNA gene partial sequence	1	Prymnesiophyceae

(A) 16S rRNA gene hits

Cruise ID	Hit ID	Hit Description	# Hits	Category
KM1110	gb|EF574323.1|	Uncultured bacterium clone S25_667 16S ribosomal RNA gene partial sequence	1	plastid (Bacillariophyceae)
KM1110	gb|EU394568.1|	Uncultured diatom clone PEACE2006/111_P3 16S ribosomal RNA gene partial sequence; chloroplast	1	plastid (Bacillariophyceae)
KM1110	gb|AY702151.1|	Dictyochophyte sp. RCC332 16S ribosomal RNA gene partial sequence; plastid	1	plastid (Dictyochophyceae)
KM1110	gb|GQ863844.1|	Uncultured eukaryote clone Ellett_IB4_32m_260 16S ribosomal RNA gene partial sequence; chloroplast	2	plastid (Dictyochophyceae)
KM1110	gb|FJ649290.1|	Uncultured phototrophic eukaryote clone STB11_25m_F8 16S ribosomal RNA gene partial sequence; chloroplast	2	plastid (Dictyochophyceae)
KM1110	gb|FJ649286.1|	Uncultured phototrophic eukaryote clone STB11_25m_E12 16S ribosomal RNA gene partial sequence; chloroplast	6	plastid (Dinophyceae)
KM1110	gb|GU119713.1|	Uncultured organism clone Gven_I11 16S ribosomal RNA gene partial sequence; chloroplast	4	plastid (Eustigmatophyceae)
KM1110	gb|HQ672102.1|	Uncultured bacterium clone F9P2610_S_H04 16S ribosomal RNA gene partial sequence	1	plastid (Pelagophyceae)
KM1110	gb|GQ863828.1|	Uncultured eukaryote clone Ellett_IB4_32m_102 16S ribosomal RNA gene partial sequence; chloroplast	1	plastid (Pelagophyceae)
KM1110	gb|FJ649259.1|	Uncultured phototrophic eukaryote clone UPW1_5m_B9 16S ribosomal RNA gene partial sequence; chloroplast	1	plastid (Prasinophyceae)
KM1110	gb|HQ672120.1|	Uncultured bacterium clone F9P2610_S_I09 16S ribosomal RNA gene partial sequence	1	plastid (Prymnesiophyceae)
KM1110	gb|EF574915.1|	Uncultured bacterium clone S25_1259 16S ribosomal RNA gene partial sequence	1	plastid (Prymnesiophyceae)
KM1110	gb|EF574086.1|	Uncultured bacterium clone S25_430 16S ribosomal RNA gene partial sequence	2	plastid (Prymnesiophyceae)
KM1110	gb|EF574441.1|	Uncultured bacterium clone S25_785 16S ribosomal RNA gene partial sequence	1	plastid (Prymnesiophyceae)
KM1110	gb|EF052026.1|	Uncultured haptophyte clone 250304-27 16S ribosomal RNA gene partial sequence; plastid	1	plastid (Prymnesiophyceae)
KM1110	gb|EF052003.1|	Uncultured haptophyte clone 250304-3 16S ribosomal RNA gene partial sequence; plastid	2	plastid (Prymnesiophyceae)
KM1110	gb|CP001068.1|	Ralstonia pickettii 12J chromosome 1 complete sequence	6	*Ralstonia pickettii*
KM1110	gb|CP001842.1|	Cyanobacterium UCYN-A complete genome	11	UCYN-A

Table S3. BLASTn hits of18S and 16S rRNA gene sequences derived from sorts of the entire picoeukaryote population from samples KM1110 (Accession numbers JX291805 - JX291865) (A) and HOT234 (Accession numbers JX291866 - JX291959) (B).

(B) 18S rRNA gene hits

Cruise ID	Hit ID	Hit Description	# Hits	Category
HOT234	gb|AY429070.1|	Uncultured dinoflagellate clone W159G6 18S ribosomal RNA gene partial sequence	1	Dinophyceae
HOT234	gb|AY664893.1|	Uncultured eukaryote clone SCM28C40 18S ribosomal RNA gene partial sequence	6	Dinophyceae
HOT234	gb|AY664957.1|	Uncultured eukaryote clone SCM15C83 18S ribosomal RNA gene partial sequence	1	Dinophyceae
HOT234	gb|AY664961.1|	Uncultured eukaryote clone SCM28C60 18S ribosomal RNA gene partial sequence	1	Dinophyceae
HOT234	gb|AY665023.1|	Uncultured eukaryote clone SCM27C47 18S ribosomal RNA gene partial sequence	1	Dinophyceae
HOT234	gb|DQ499645.1|	Lepidodinium viride 18S ribosomal RNA gene internal transcribed spacer	4	Dinophyceae
HOT234	gb|DQ504314.1|	Uncultured alveolate clone LC22_4EP_19 18S ribosomal RNA gene partial sequence	1	Dinophyceae
HOT234	gb|EU418969.1|	Dinophyceae sp. GD1590bp26 18S ribosomal RNA gene partial sequence	17	Dinophyceae
HOT234	gb|EU818044.1|	Uncultured marine alveolate clone ZZ0053262 18S ribosomal RNA gene partial sequence	1	Dinophyceae
HOT234	gb|EU818520.1|	Uncultured marine alveolate clone ZZ0053129 18S ribosomal RNA gene partial sequence	1	Dinophyceae
HOT234	gb|FJ914456.1|	Uncultured marine dinoflagellate clone S7 18S ribosomal RNA gene partial sequence	1	Dinophyceae
HOT234	gb|GQ382900.1|	Uncultured marine eukaryote clone MO010_42.00029 18S ribosomal RNA gene partial sequence	2	Dinophyceae
HOT234	gb|AY665124.1|	Uncultured eukaryote clone SCM27C27 18S ribosomal RNA gene partial sequence	6	Maxillopoda
HOT234	gb|AY749516.1|	Uncultured eukaryote clone H30.5 18S ribosomal RNA gene partial sequence	1	Maxillopoda
HOT234	gb|GU969200.1|	Clausocalanus furcatus 18S ribosomal RNA gene partial sequence	1	Maxillopoda
HOT234	gb|U40927.1|	Coccoid pelagophyte CCMP1395 nuclear 18S ribosomal RNA gene	2	Pelagophyceae
HOT234	gb|EU287795.1|	Uncultured marine Polycystinea clone OLI011-75m.50 18S ribosomal RNA gene partial sequence	1	Polycystinea
HOT234	gb|FJ537305.1|	Uncultured Prasinophyceae clone Biosope_T19.017 18S ribosomal RNA gene partial sequence	1	Prasinophyceae
HOT234	gb|FJ537318.1|	Uncultured Prasinophyceae clone Biosope_T39.095 18S ribosomal RNA gene partial sequence	2	Prasinophyceae
HOT234	gb|FJ537324.1|	Uncultured Prasinophyceae clone Biosope_T41.030 18S ribosomal RNA gene partial sequence	4	Prasinophyceae
HOT234	gb|FJ537325.1|	Uncultured Prasinophyceae clone Biosope_T41.051 18S ribosomal RNA gene partial sequence	1	Prasinophyceae
HOT234	gb|FJ537354.1|	Uncultured Prasinophyceae clone Biosope_T84.034 18S ribosomal RNA gene partial sequence	1	Prasinophyceae
HOT234	gb|HM474512.1|	Uncultured Prasinophyceae clone T41_W01D.032 18S ribosomal RNA gene partial sequence	1	Prasinophyceae
HOT234	gb|U40921.1|	Coccoid green alga CCMP1205 nuclear 18S ribosomal RNA gene	1	Prasinophyceae
HOT234	dbj|AB183618.1|	Emiliania sp. MBIC10582 gene for 18S rRNA partial sequence strain: MBIC10582	7	Prymnesiophyceae
HOT234	emb|AM490987.2|	Syracosphaera pulchra partial 18S rRNA gene strain ALGO GK 17	1	Prymnesiophyceae
HOT234	gb|FJ537341.1|	Uncultured Chrysochromulina clone Biosope_T60.034 18S ribosomal RNA gene partial sequence	4	Prymnesiophyceae
HOT234	gb|DQ314809.1|	Uncultured marine eukaryote clone NOR26.10 18S ribosomal RNA gene partial sequence	1	Sarcomonadea
HOT234	gb|EF172987.1|	Uncultured eukaryote clone SSRPD85 18S ribosomal RNA gene partial sequence	1	Syndiniophyceae
HOT234	gb|EF172989.1|	Uncultured eukaryote clone SSRPD87 18S ribosomal RNA gene partial sequence	1	Syndiniophyceae
HOT234	gb|EU793392.1|	Uncultured syndiniales clone PROSOPE.E5-55m.20 18S ribosomal RNA gene partial sequence	2	Syndiniophyceae
HOT234	gb|EU793689.1|	Uncultured syndiniales clone PROSOPE.ED-50m.193 18S ribosomal RNA gene partial sequence	1	Syndiniophyceae
HOT234	gb|EU793933.1|	Uncultured syndiniales clone PROSOPE.EM-5m.190 18S ribosomal RNA gene partial sequence	1	Syndiniophyceae

(B) 16S rRNA gene hits

Cruise ID	Hit ID	Hit Description	# Hits	Category
HOT234	gb|EF572495.1|	Uncultured bacterium clone S23_594 16S ribosomal RNA gene partial sequence	1	*Actinobacteria*
HOT234	gb|GU061498.1|	Uncultured bacterium clone CE1-DCM-27 16S ribosomal RNA gene partial sequence	1	*Actinobacteria*
HOT234	gb|DQ438491.1|	Uncultured bacterium clone ECS-P7-D55 16S ribosomal RNA gene partial sequence	2	plastid (Prasinophyceae)
HOT234	gb|AY702115.1|	Emiliania huxleyi strain CCMP625 16S ribosomal RNA gene partial sequence; plastid	1	plastid (Prymnesiophyceae)
HOT234	gb|EF052031.1|	Uncultured haptophyte clone 250304-32 16S ribosomal RNA gene partial sequence; plastid	1	plastid (Prymnesiophyceae)
HOT234	gb|EU802928.1|	Uncultured bacterium clone 4C230321 16S ribosomal RNA gene partial sequence	1	*Prochlorococcus*
HOT234	gb|EU804095.1|	Uncultured bacterium clone 6C231987 16S ribosomal RNA gene partial sequence	1	*Prochlorococcus*
HOT234	gb|EU805328.1|	Uncultured bacterium clone 6C233331 16S ribosomal RNA gene partial sequence	11	*Prochlorococcus*
HOT234	gb|GU061741.1|	Uncultured bacterium clone CEP-5m-43 16S ribosomal RNA gene partial sequence	1	*Prochlorococcus*
HOT234	gb|CP001842.1|	Cyanobacterium UCYN-A complete genome	1	UCYN-A

Table S4: Numbers and best BLASTn identity of 18S rRNA gene sequences derived from nested PCR of single *nifH*-positive picoeukaryotes from HOT234 and KM1110. Cruises are KM1110 (5 m March 2011, accession numbers JX291679 - JX291804) and HOT234 (79 m August 2011, accession numbers JX291547 - JX291678). Asterisk (*****) indicates the only marine species amplified. All others are suspected contaminants from terrestrial sources. Full species names are *Chrysochromulina acantha, Pinus armandii, Pinus luchuensis, Pinus morrisonicola, Lithocarpus rufovillosus, Ralstonia solanacearum, and Cunninghamia lanceolata*.

		Marine species		***Terrestrial species and suspected contaminants***								
Cruise	**Single Cell ID**	**BIOSOPE T60.34***	***C. acantha****	***P. armandii***	***P. luchuensis***	***P. morrisonicola***	***L. rufovillosus***	**environmental embryophyte**	***R. solanacearum***	***C. lanceolata***	**TOTAL sequences**	**Majority of sequences from each single cell**
KM1110	3	0	0	13	0	0	0	0	1	0	14	*P. armandii*
KM1110	4	0	0	14	0	0	0	0	0	0	14	*P. armandii*
KM1110	5	0	0	14	0	0	0	0	0	0	14	*P. armandii*
KM1110	6	12	0	1	1	0	0	0	0	0	14	Biosope_T60.034
KM1110	7	1	0	11	0	1	0	0	0	1	14	*P. armandii*
KM1110	8	0	0	14	0	0	0	0	0	0	14	*P. armandii*
KM1110	9	0	0	12	0	0	1	0	0	0	13	*P. armandii*
KM1110	10	0	0	12	0	1	1	0	0	0	14	*P. armandii*
KM1110	11	0	0	13	0	0	0	0	0	0	13	*P. armandii*
KM1110	12	17	0	0	0	0	0	0	0	0	17	Biosope_T60.034
KM1110	13	0	0	17	1	0	0	0	0	0	18	*P. armandii*
KM1110	14	0	0	16	1	1	0	0	0	0	18	*P. armandii*
KM1110	15	17	0	2	0	0	0	0	0	0	19	Biosope_T60.034
KM1110	16	18	0	1	0	0	0	0	0	0	19	Biosope_T60.034
KM1110	17	16	0	2	0	0	0	0	0	0	18	Biosope_T60.034
KM1110	18	18	0	0	0	0	0	0	0	0	18	Biosope_T60.034
KM1110	19	19	0	0	0	0	0	0	0	0	19	Biosope_T60.034
KM1110	20	0	0	19	0	0	0	0	0	0	19	*P. armandii*
KM1110	21	0	0	18	0	0	0	2	0	0	20	*P. armandii*
KM1110	22	20	0	0	0	0	0	0	0	0	20	Biosope_T60.034
HOT234	16E7	0	0	14	1	0	0	0	0	0	15	*P. armandii*
HOT234	11D12	5	1	11	0	0	0	0	0	0	17	*P. armandii*
HOT234	11D6	0	0	16	0	0	0	0	0	0	16	*P. armandii*
HOT234	16C10	7	0	3	0	1	0	0	0	0	11	Biosope_T60.034
HOT234	16C9	10	0	10	0	0	0	0	0	0	20	Biosope_T60.034
HOT234	16D11	0	0	8	0	0	0	0	0	0	8	*P. armandii*
HOT234	16F10	11	0	5	0	0	0	0	0	0	16	Biosope_T60.034
HOT234	11A9	0	0	15	1	1	1	0	0	0	18	*P. armandii*
HOT234	15F1	17	0	1	0	0	0	0	0	0	18	Biosope_T60.034
HOT234	16B2	13	0	14	0	0	0	0	0	0	27	*P. armandii*

Table S5. The UCYN-732 and Helpers A and B oligonucleotide (5’ to 3’) shown with other closely related free-living and symbiotic cyanobacterial sequences. Mismatches are highlighted in red.

	Helper A	UCYN-732	Helper B	
UCYN-A	5’ AGCTTTCGTCCCTGAGTGTCA	GTTACGGTCCAGTAGCAC	GCCTTCGCCACCGATGTT	3’
Cyanothece ATCC 51142	5’ AGCTTTCGTCCCTGAGTGTCA	GTTCCGGTCCAGTAGCGC	GCCTTCGCCACCGATGTT	3’
endosymbiont of *Climacodium frauenfeldianum*	5’ AGCTTTCGTCCCTGAGTGTCA	GTTTCGGTCCAGTAGCGC	GCCTTCGCCACCGATGTT	3’
endosymbiont of *Rhopalodia gibba*	5’ AGCTTTCGTCCCTGAGTGTCA	GTTCCGGCCCAGTAGCGC	GCTTTCGCCACCGATGTT	3’

Table S6. Summary of cell dimensions and nanoSIMS analyses for UCYN-A and partner cells measured by nanoSIMS.

Cell Type	Cell diameter (μm)	AT% ^{13}C	AT % ^{15}N
UCYN-A and partner			
Partner 1	1.47	2.8853	1.7218
UCYN-A	0.60	1.9108	1.0577
Partner 2	1.34	2.2014	0.6538
UCYN-A	0.71	1.8723	0.5854
Partner 3	1.35	2.4951	1.8019
UCYN-A	0.92	1.8752	1.2248
Partner 4	1.12	2.0990	1.5448
UCYN-A	0.52	2.0501	1.3155
Partner 5	1.22	2.8060	1.5893
UCYN-A	0.52	2.1220	1.2336
Partner 6	1.47	2.7436	1.4769
UCYN-A	0.79	2.3495	1.2287
Partner 7	1.65	2.8107	1.6822
UCYN-A	0.75	2.1861	1.2258
Partner 8	1.23	1.8838	1.5322
UCYN-A	0.31	1.2862	1.4322
UCYN-A	0.40	1.5864	1.3271
Partner 9	0.99	1.8501	1.3067
UCYN-A	0.45	1.2316	1.0430
UCYN-A	0.38	1.5583	1.2236
Partner 10	1.76	2.8268	1.9983
UCYN-A	0.76	1.9954	1.3398
UCYN-A	0.65	2.0280	1.4642
Dislodged UCYN-A			
UCYN-A	0.60	1.8954	1.3914
UCYN-A	0.69	1.6464	1.1809
UCYN-A	0.68	2.0098	1.4992
UCYN-A	0.86	2.0549	1.3330
UCYN-A	0.57	1.2560	0.7777
UCYN-A	0.56	1.5506	1.0606

Figure S1

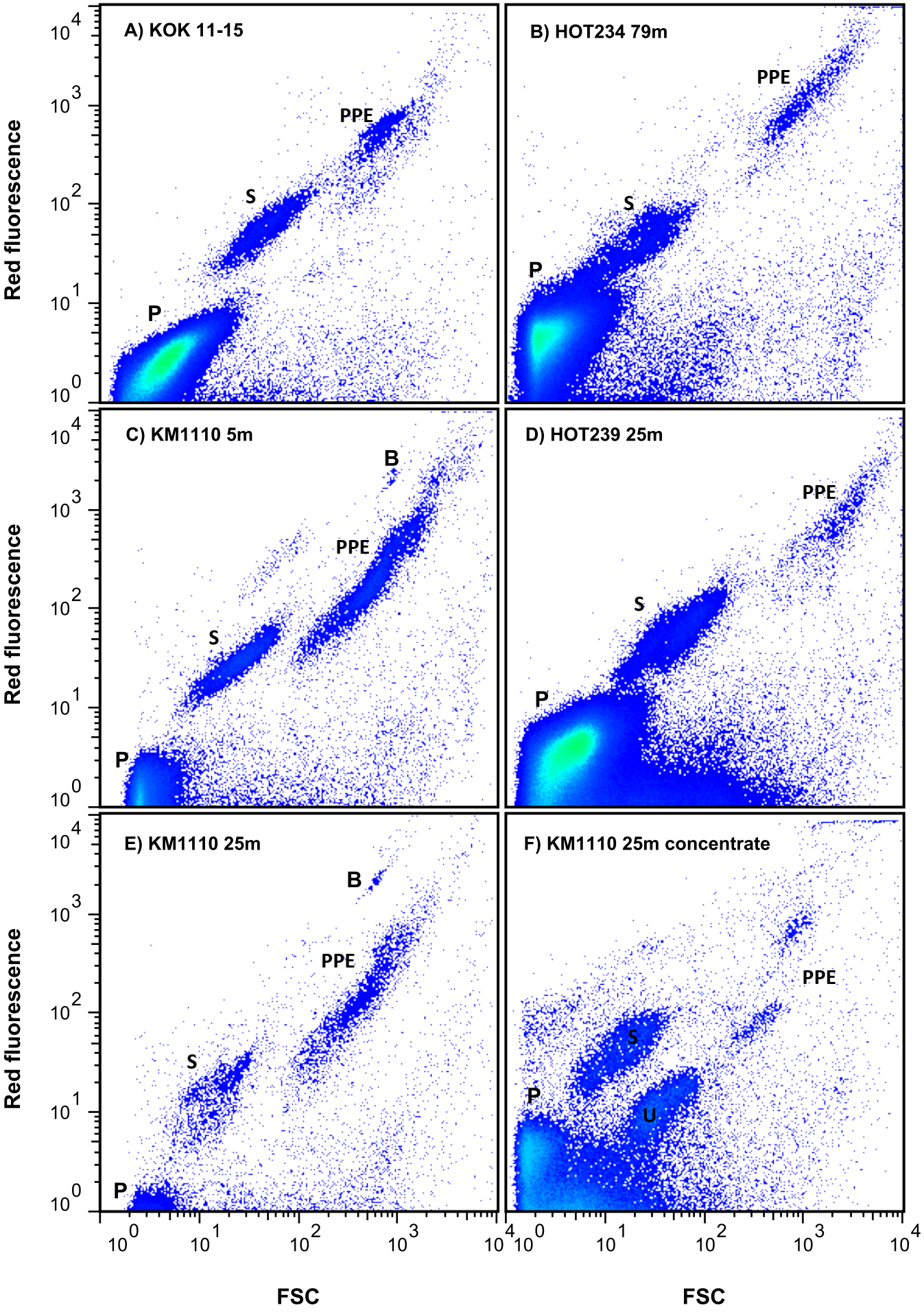
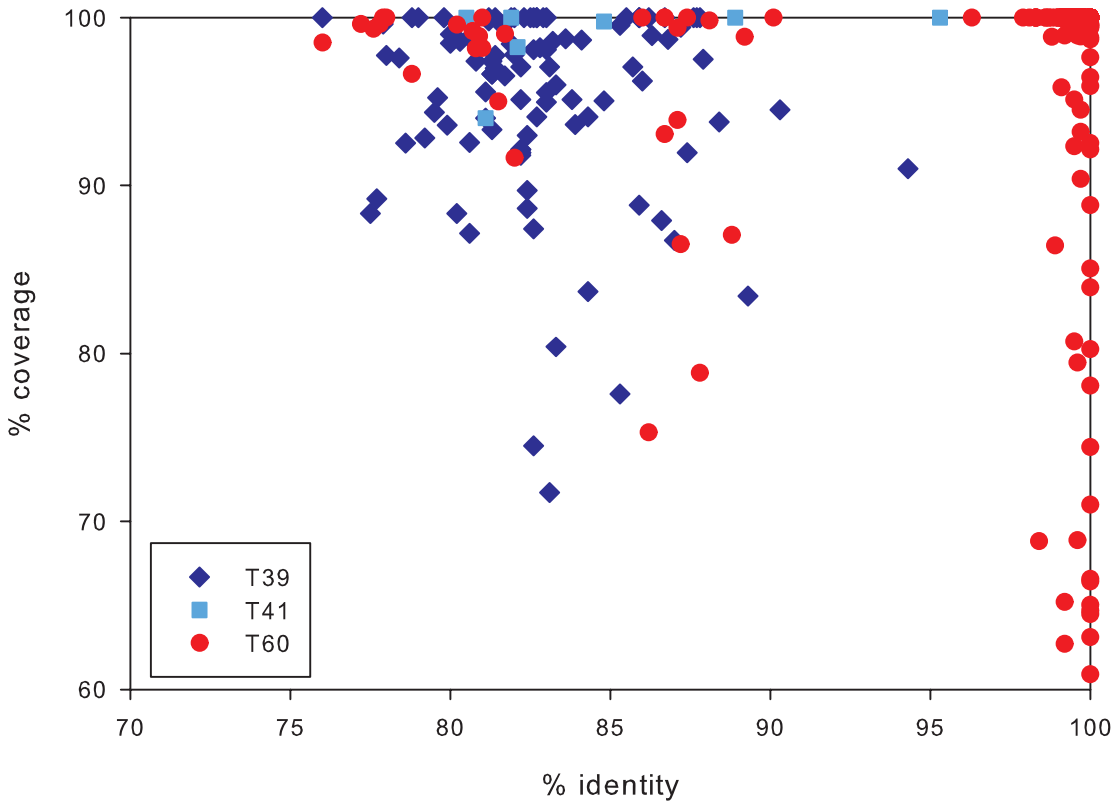

A) KOK 11-15
B) HOT234 79m
C) KM1110 5m
D) HOT239 25m
E) KM1110 25m
F) KM1110 25m concentrate
P
S
B
PPE
U
Red fluorescence
FSC
10^0
10^1
10^2
10^3
10^4

Figure S2


100
90
80
70
60
% coverage
70
75
80
85
90
95
100
% identity
T39
T41
T60

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