	Growth	Growth
Strain	Temperature	Medium
Candidatus Pelagibacter ubique HTCC1062	20 °C	AMS1 ^a
Phaeobacter inhibens 2.10	28 °C	1/2 YTSS ^b
Phaeobacter inhibens DSM 17395	28 °C	1/2 YTSS ^b
Silicibacter pomeroyi ATCC 700808	30 °C	1/2 YTSS ^b
Trichodesmium Epibiont - 1B1	30 °C	MB 2216 ^b
Trichodesmium Epibiont - 3	30 °C	MB 2216 ^b
Trichodesmium Epibiont - 7	30 °C	MB 2216 ^b
Trichodesmium Epibiont - 11	30 °C	MB 2216 ^b
Vibrio alginolyticus DSM 2171	30 °C	MB 2216 ^b
Vibrio anguillarum 775	26 °C	L-Marine ^b
Vibrio anguillarum ATCC 14181	26 °C	L-Marine ^b
Vibrio fischeri ATCC 7744	26 °C	L-Marine ^b
Vibrio harveyi (campbellii) ATCC BAA-1116	30 °C	L-Marine ^b
Vibrio harveyi DSM 19623	30 °C	MB 2216 ^b
Vibrio orientalis DSM 19136	30 °C	MB 2216 ^b
Vibrio metschnikovii DSM 19132	30 °C	MB 2216 ^b
Vibrio splendidus DSM 19640	26 °C	MB 2216 ^b

Table S1. Pre-experimental conditions for cultures. ^a Cultures were maintained in liquid medium until mid-exponential phase. ^b Cultures were streaked onto agar plates of the listed medium and grown overnight. Pre-growth (~16 hours overnight) was then conducted in experimental liquid medium before the start of experiments (see methods). MB: marine broth.

	Final Concentration	C Contributed to	N Contributed to
Amino Acid	in Medium (M)	Medium (M)	Medium (M)
Essential AA			
Arginine	2.40 x 10 ⁻³	1.44 x 10 ⁻²	9.58 x 10 ⁻³
Cysteine	$4.00 \ge 10^{-4}$	$1.20 \ge 10^{-3}$	$4.00 \ge 10^{-4}$
Histidine	8.00 x 10 ⁻⁴	$4.80 \ge 10^{-3}$	2.40×10^{-3}
Isoleucine	1.60 x 10 ⁻³	9.60 x 10 ⁻³	$1.60 \ge 10^{-3}$
Leucine	1.60×10^{-3}	9.60×10^{-3}	$1.60 \ge 10^{-3}$
Lysine	1.58 x 10 ⁻³	9.51 x 10 ⁻³	3.17 x 10 ⁻³
Methionine	4.05×10^{-4}	2.03×10^{-3}	4.05×10^{-4}
Phenylalanine	8.00 x 10 ⁻⁴	7.20 x 10 ⁻³	8.00 x 10 ⁻⁴
Threonine	1.60 x 10 ⁻³	6.40 x 10 ⁻³	1.60 x 10 ⁻³
Tryptophan	2.00 x 10 ⁻⁴	2.20 x 10 ⁻³	4.00 x 10 ⁻⁴
Tyrosine	7.96 x 10 ⁻⁴	7.16×10^{-3}	7.96×10^{-4}
Valine	1.60×10^{-3}	8.00×10^{-3}	$1.60 \ge 10^{-3}$
Essential AA Total	1.38×10^{-2}	8.21×10^{-2}	2.44×10^{-2}
Non-Essential AA			
Alanine	$4.00 \ge 10^{-4}$	$1.20 \ge 10^{-3}$	$4.00 \ge 10^{-4}$
Asparagine	4.00 x 10 ⁻⁴	1.60 x 10 ⁻³	8.00 x 10 ⁻⁴
Aspartic Acid	$4.00 \ge 10^{-4}$	$1.60 \ge 10^{-3}$	$4.00 \ge 10^{-4}$
Glutamic Acid	$4.00 \ge 10^{-4}$	2.00×10^{-3}	$4.00 \ge 10^{-4}$
Glycine	$4.00 \ge 10^{-4}$	8.00 x 10 ⁻⁴	$4.00 \ge 10^{-4}$
Proline	$4.00 \ge 10^{-4}$	2.00×10^{-3}	$4.00 \ge 10^{-4}$
Serine	4.00 x 10 ⁻⁴	1.20 x 10 ⁻³	4.00 x 10 ⁻⁴
Non-Essential AA	2.80×10^{-3}	1.04 x 10 ⁻²	3.20×10^{-3}
Total (EAA & NEAA)	1.67 x 10 ⁻²	9.25 x 10 ⁻²	2.76 x 10 ⁻²

Table S2. Amino acid composition of MEM amino acids used in this study. Concentrations of individual amino acids as well as N and C contributions to the growth medium are listed. Liquid stock of essential (M5550) and non-essential (M7145) were purchased from Sigma.



Figure S1. Growth of SAR11 in AMS1 (black), AMS1 with Fe-replete H-Aquil trace metals (1 μ M Fe, red), AMS1 with Fe-deplete H-Aquil trace metals (no added Fe, green), and Fe-replete H-Aquil (1 μ M Fe, blue). Growth in H-Aquil was conducted as a negative control, and as expected, no growth was observed, most likely due to the lack of needed vitamins and an appropriate carbon source. No difference was observed between cultures grown with and without added Fe, suggesting that more rigorous Fe removal may be needed to achieve Fe limitation in this organism.



Figure S2. Growth of *Vibrios* and other marine heterotrophs in H-Aquil supplemented with 1µM Fe (black) and no added Fe (grey). Growth of *S. pomeroyi*, and all epibionts was conducted in a plate reader using 96-well microplates. All other experiments were conducted in Nunc flasks.



Figure S3. Growth of *V. harveyi* in metal-replete, Fe-deplete, and Cu-deplete H-Aquil. The top panel corresponds to H-Aquil utilizing fully synthetic salts. The bottom panel corresponds to H-Aquil made with Sargasso seawater. Metal-replete: blue, light blue; Cu-deplete: green, light green; Fe-deplete: red, orange.



Fig S4. Stability of *V. harveyi* growth rates in Fe-replete and deplete H-Aquil made with synthetic and natural (Sargasso) seawater. Cells were transferred every 24 hours to exhaust cellular stores of trace metals. Stable growth rates across transfers suggest that these reserves are negligible. Comparisons of synthetic and Sargasso seawater (SS) also demonstrate that salts are not the most significant source of metal contamination in our study. Experiments were conducted in biological duplicates.