

**Supporting Information for**

**Influence of nitrogen limitation on the bioaccumulation kinetics  
of hematite nanoparticles in the freshwater alga *Euglena*  
*intermedia***

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8 pages, including 3 Tables and 4 Figures

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23 **Table S1** Chemical components and their concentrations in HUT.

Components	Final concentration (mg L <sup>-1</sup> )	Components	Final concentration (mg L <sup>-1</sup> )
proteose peptone	600	CH <sub>3</sub> COONa	400
yeast extract	400	potassium citrate	40
KH <sub>2</sub> PO <sub>4</sub>	20	Vitamin B <sub>1</sub>	0.0004
MgSO <sub>4</sub> • 7H <sub>2</sub> O	25	Vitamin B <sub>12</sub>	0.0005

25 **Table S2** Chemical components and their concentrations in AF-6.

Components	Final concentration (mg L <sup>-1</sup> )	Components	Final concentration (mg L <sup>-1</sup> )
NH <sub>4</sub> NO <sub>3</sub>	198.1	Na <sub>2</sub> EDTA	0.75
CaCl <sub>2</sub> ·2H <sub>2</sub> O	147	FeCl <sub>3</sub> ·6H <sub>2</sub> O	0.097
MgSO <sub>4</sub> ·7H <sub>2</sub> O	246.7	MnCl <sub>2</sub> ·4H <sub>2</sub> O	0.041
KH <sub>2</sub> PO <sub>4</sub>	34	ZnCl <sub>2</sub> ·7H <sub>2</sub> O	0.005
CaCO <sub>3</sub>	10	Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	0.004
Citric acid	2	CoCl <sub>2</sub> ·6H <sub>2</sub> O	0.002
Fe-citrate	2		

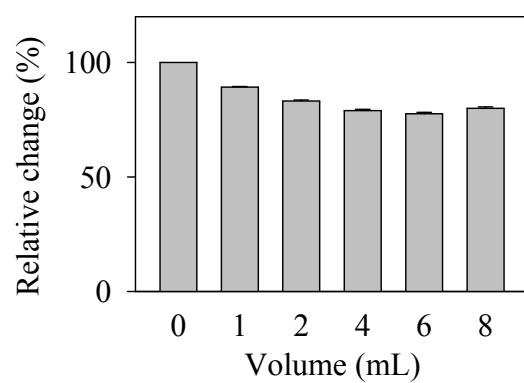
27 **Table S3** Full names of the abbreviations used throughout the text.

Abbreviation	Full name
HemNPs	polyacrylate-coated hematite ( $\alpha$ -Fe <sub>2</sub> O <sub>3</sub> ) nanoparticles
QDs	quantum dots
-N	nitrogen-depleted
NE	nutrient-enriched
TEM	transmission electron microscopy
DLS	dynamic light scattering particle sizer
RCF	relative centrifugal force
[HemNPs] <sub>cell</sub>	HemNP concentration in the cells (pg-Fe $\mu\text{m}^{-3}$ )
[HemNPs] <sub>med</sub>	HemNP concentration in the experimental medium (mg-Fe L <sup>-1</sup> )
[HemNPs] <sub>ads</sub>	cell-surface-adsorbed concentration of HemNPs (pg-Fe $\mu\text{m}^{-3}$ )
[HemNPs] <sub>intra</sub>	intracellular concentration of HemNPs (pg-Fe $\mu\text{m}^{-3}$ )
$k_u$	uptake rate constant of HemNPs (L $\mu\text{m}^{-3}$ h <sup>-1</sup> )
$k_e$	efflux rate constant of HemNPs (h <sup>-1</sup> )
$\mu$	cell-specific growth rate (h <sup>-1</sup> )
$F_v/F_m$	maximum photosynthetic system II quantum yield
Water-PAM	Water Pulse Amplitude Modulated Fluorometer
GF/F membrane	glass fiber/fine membrane
SSC	side scatter
[PS] <sub>surf</sub>	amount of polysaccharide attached on the cell surface (fg-Xanthan <sub>eq</sub> $\mu\text{m}^{-3}$ )
[CHO] <sub>cell</sub>	cellular carbohydrate concentration ( $\mu\text{mol } \mu\text{m}^{-3}$ )
[PS] <sub>dis</sub>	amount of polysaccharides released into the culture medium ( $\mu\text{M}$ )
ANOVA	analysis of variance
EDX	energy dispersive X-ray spectrometry

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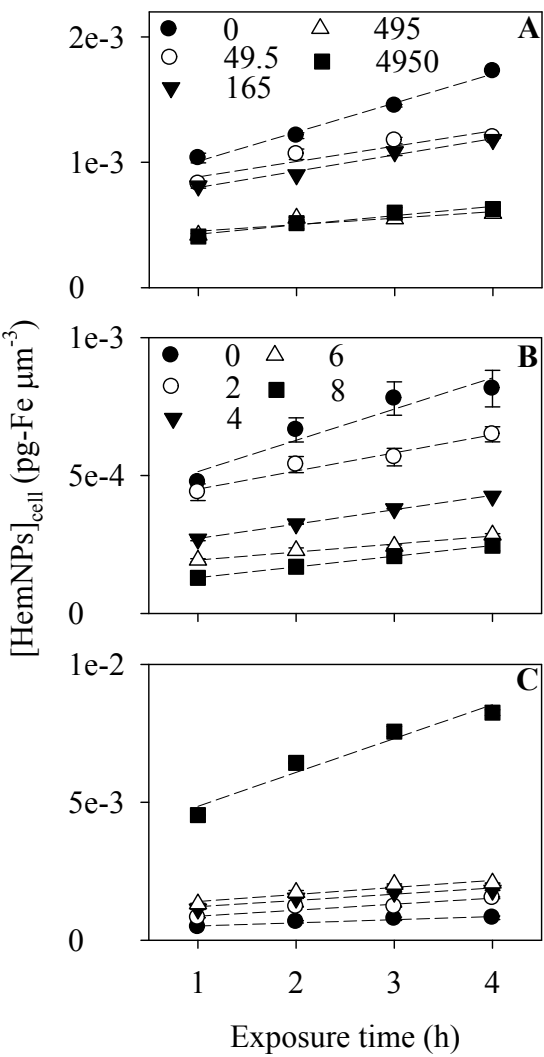
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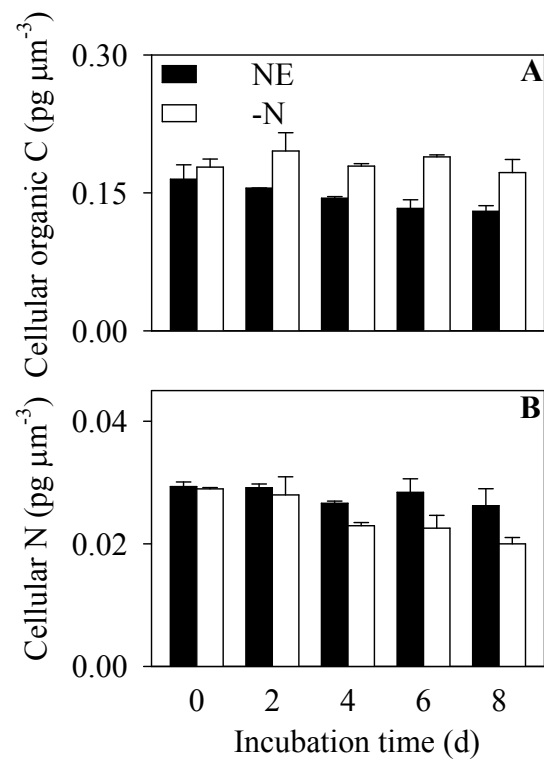
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32 **Fig. S1** Relative change of the cellular HemNP concentration ( $[\text{HemNPs}]_{\text{cell}}$ ) with increasing  
33 volume of rinsing solution (i.e., AF-6).



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36 **Fig. S2** The increase in the cellular HemNP concentration ( $[\text{HemNPs}]_{\text{cell}}$ ) with increasing  
37 exposure time in *Euglena intermedia* pre-cultured (A) in the experimental medium with  
38 different concentrations of nitrogen (0, 49.5, 165, 495, and 4950  $\mu\text{M}$ ) for 2 days as well as in  
39 (B) the nutrient-enriched and (C) nitrogen-depleted media for 0, 2, 4, 6, and 8 days before the  
40 4-h uptake experiment. Dashed lines are the linear regression between  $[\text{HemNPs}]_{\text{cell}}$  and  
41 exposure time. The data are reported as the mean  $\pm$  standard deviation ( $n = 3$ ).



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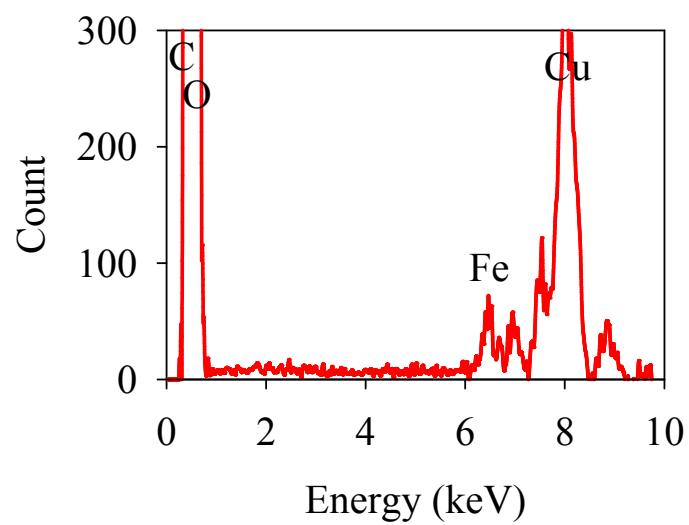
44 **Fig. S3** The cellular (A) organic carbon and (B) nitrogen concentrations in *Euglena intermedia*

45 pre-cultured in the nutrient-enriched (NE) and nitrogen-depleted (-N) media for 0, 2, 4, 6, and

46 8 days. The data are reported as the mean  $\pm$  standard deviation (n = 3).

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49 **Fig. S4** Representative elemental composition of the arrowed spots in the TEM images of Fig.

50 2B and C, as investigated by energy dispersive X-ray (EDX) spectrometry.