Electronic Supplementary Material (ESI) for Environmental Science: Nano.

Primers

	Forward	Reverse	Product Size (bp)
GFP	(TAATACGACTCACTATAG)AGGGTGAAGGTGATGCAACA	(TAATACGACTCACTATAG)GCCGTTTCATATGATCTGGGT	219

Table S1 – Oligo sequence for in-vitro synthesis of dsRNA. T7 primer sequence is labeled in parenthesis.

Confirmation of GFP Knockdown

	Expression	Std. Error	95% CI	P
dsRNA	0.421	0.124 - 0.907	0.108 - 2.944	0.028
PNs	0.48	0.157 - 1.529	0.100 - 2.801	0.041

Table S2 - CGC4 *C. elegans* were exposed to GFP dsRNA and PNs (100 ng/µL) as described in Methods and GFP expression was quantitated. This data was previously published in the supplemental information of Lichtenberg, et.al.[1], and details of this experiment can be found therein.

FCS Data Plot Examples

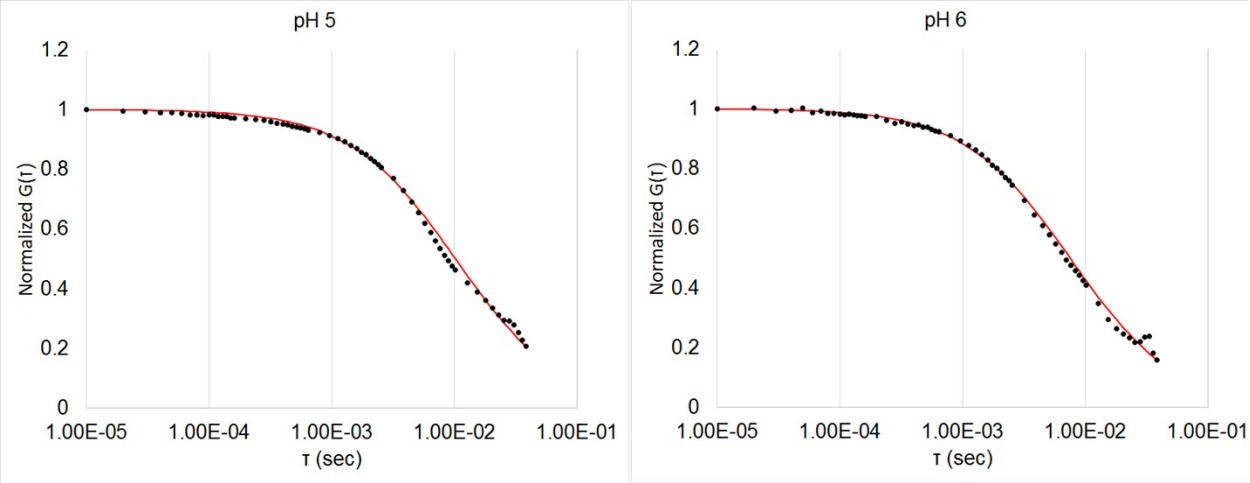

Figure S1 – Sample plots of FCS data generated in this study. Collected data is shown as black points, and the fit of the data to the autocorrelation function is show as a red line. In both cases, the data was normalized to the maximum G(τ) in each dataset.

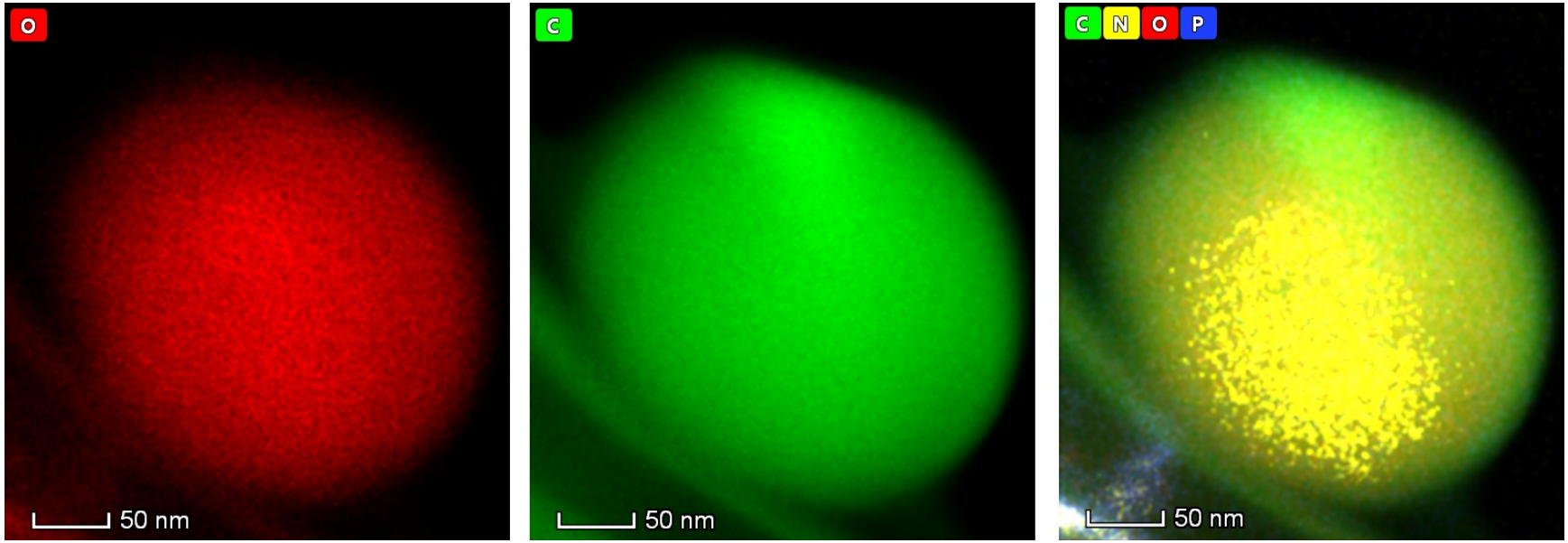

Figure S2 – EDS elemental maps for oxygen (left), carbon (center), and all measured elements (right).

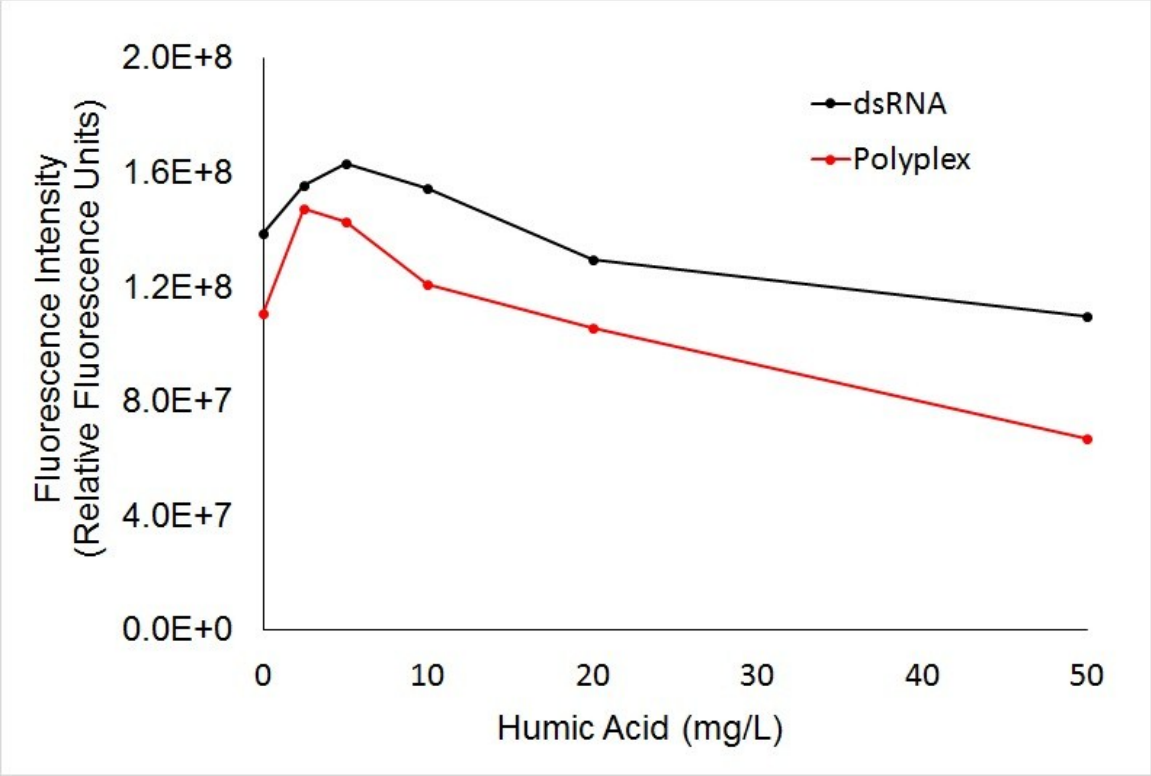

Figure S3 – Quenching of AlexaFluor 488 fluorescence by humic acid. AlexaFluor 488 labeled dsRNA and chitosan/dsRNA polyplexes were added to moderately hard reconstituted water (MHRW) amended with the indicated concentration of Pahokee peat humic acid, and fluorescence intensity was measured (Ex = 490 nm; Em = 525 nm). Background fluorescence from MHRW/humic acid blanks were subtracted from the corresponding measurement.

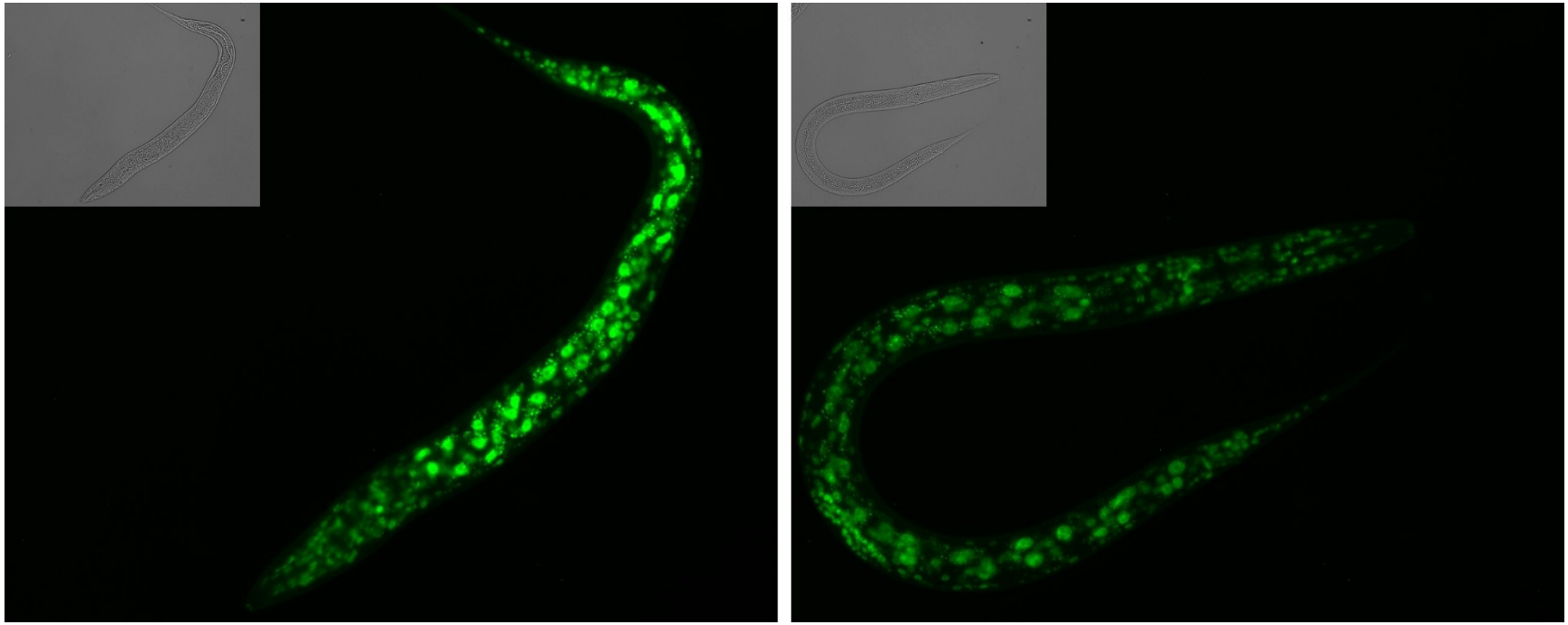

Figure S4 – Example images of control (left) and dsRNA (100 ng/µL) exposed CGC4 *Caenorhabditis elegans*. Exposure duration was 24 hrs, and imaging was performed as detailed in Methods.

1. S. S. Lichtenberg, O. V. Tsyusko, S. R. Palli and J. M. Unrine, *Environ. Sci. Technol.*, 2019, DOI: 10.1021/acs.est.8b06560.