

## Supplementary information

# Dissolved organic matter controls of Arsenic bioavailability to bacteria

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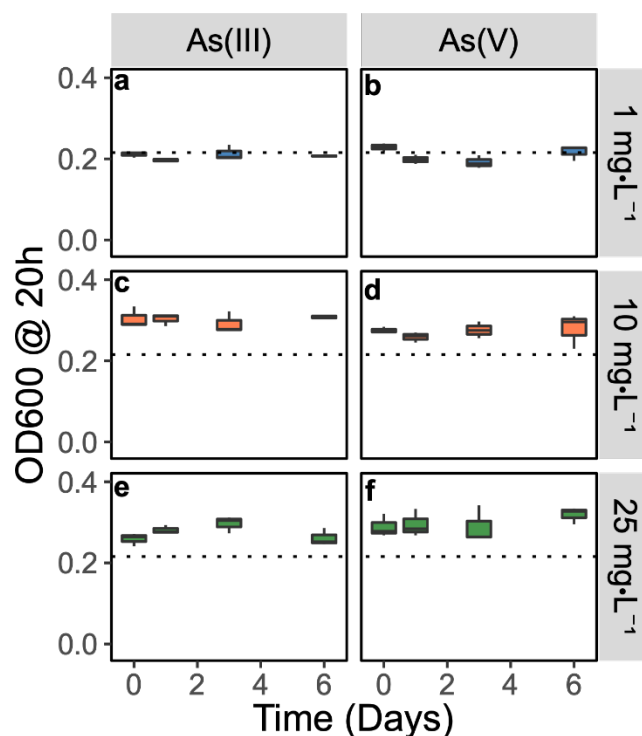
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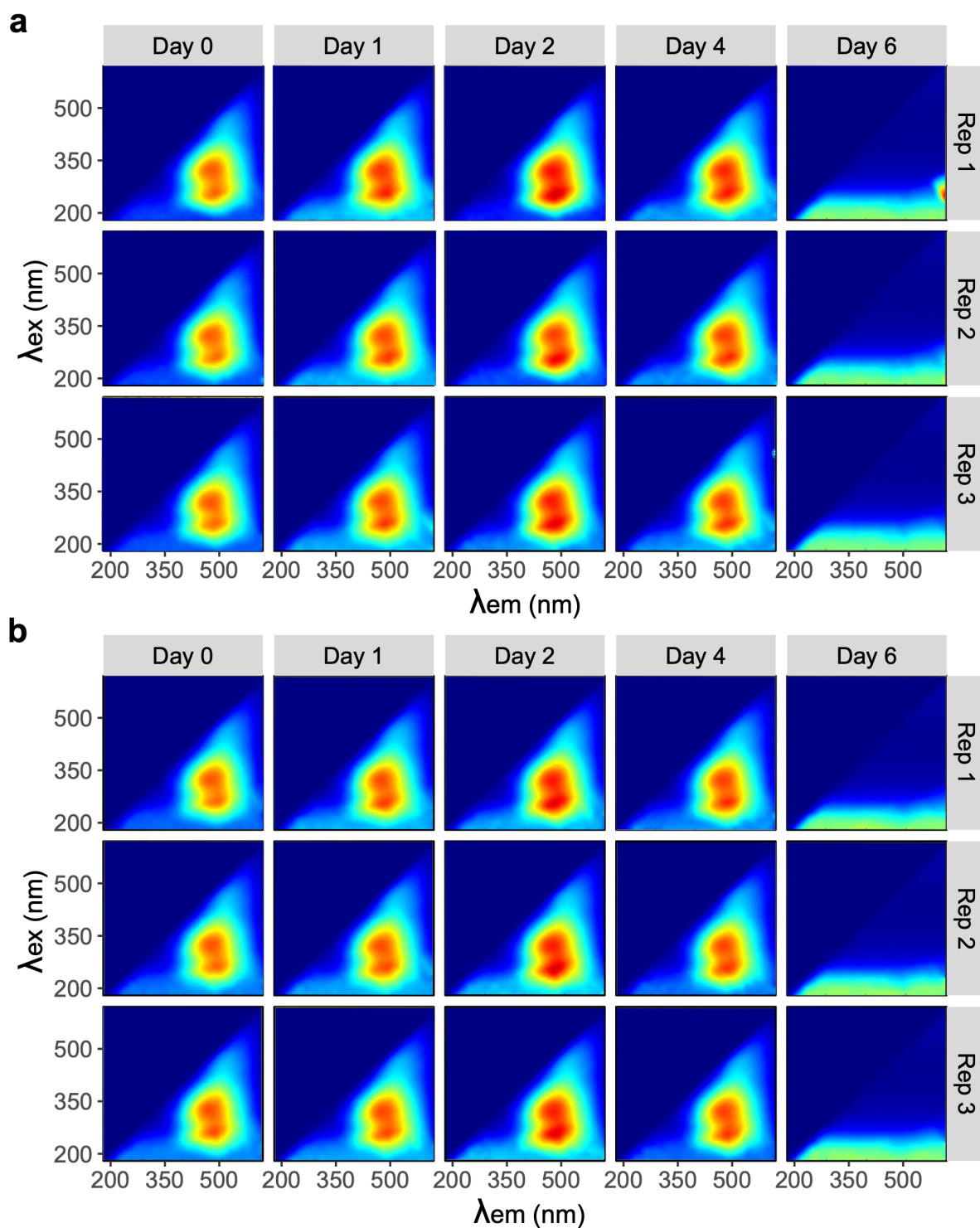
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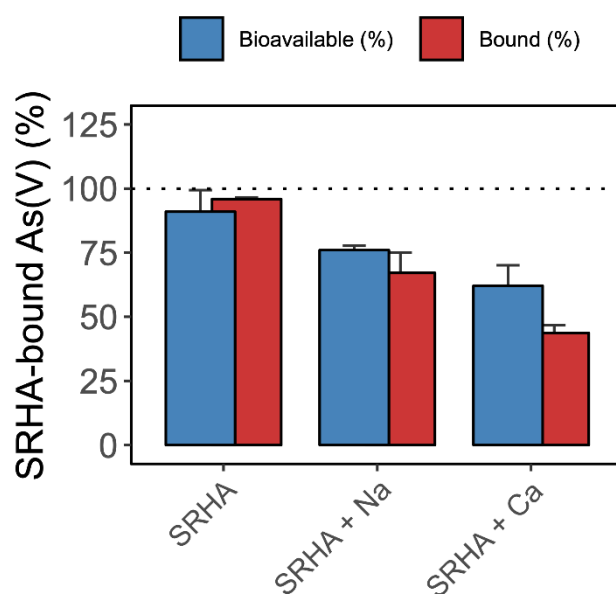
## Section A : Supplementary Figures



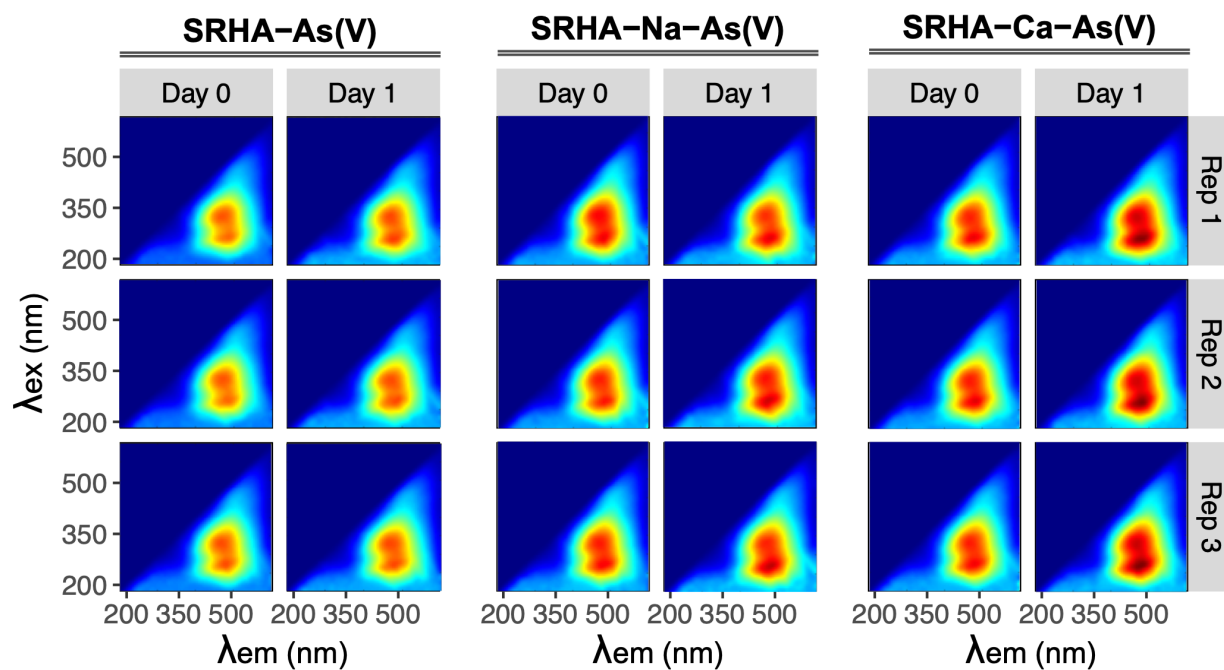
**Fig. S1 |** High DOM concentrations increase in the yield of biosensor cultures (Fig. 1). Boxplots represent triplicate cultures containing either 200 nM of As(III) (**a, c, e**) or 200 nM As(V) (**b, d, f**) in the presence of a DOM (SRHA) gradient (**vertical facets**). Axes include room-temperature pre-incubation time (X-axis) and output signals of treatments are indicated as percent deviation from our no-DOM controls (Y-axis). Dotted line represents biosensor yield in the no-DOM controls. Stars represent a significant decrease from the no-DOM control incubated for the same period of time determined using raw OD600 measurements and TukeyHSD post-hoc analysis on a one-way anova.



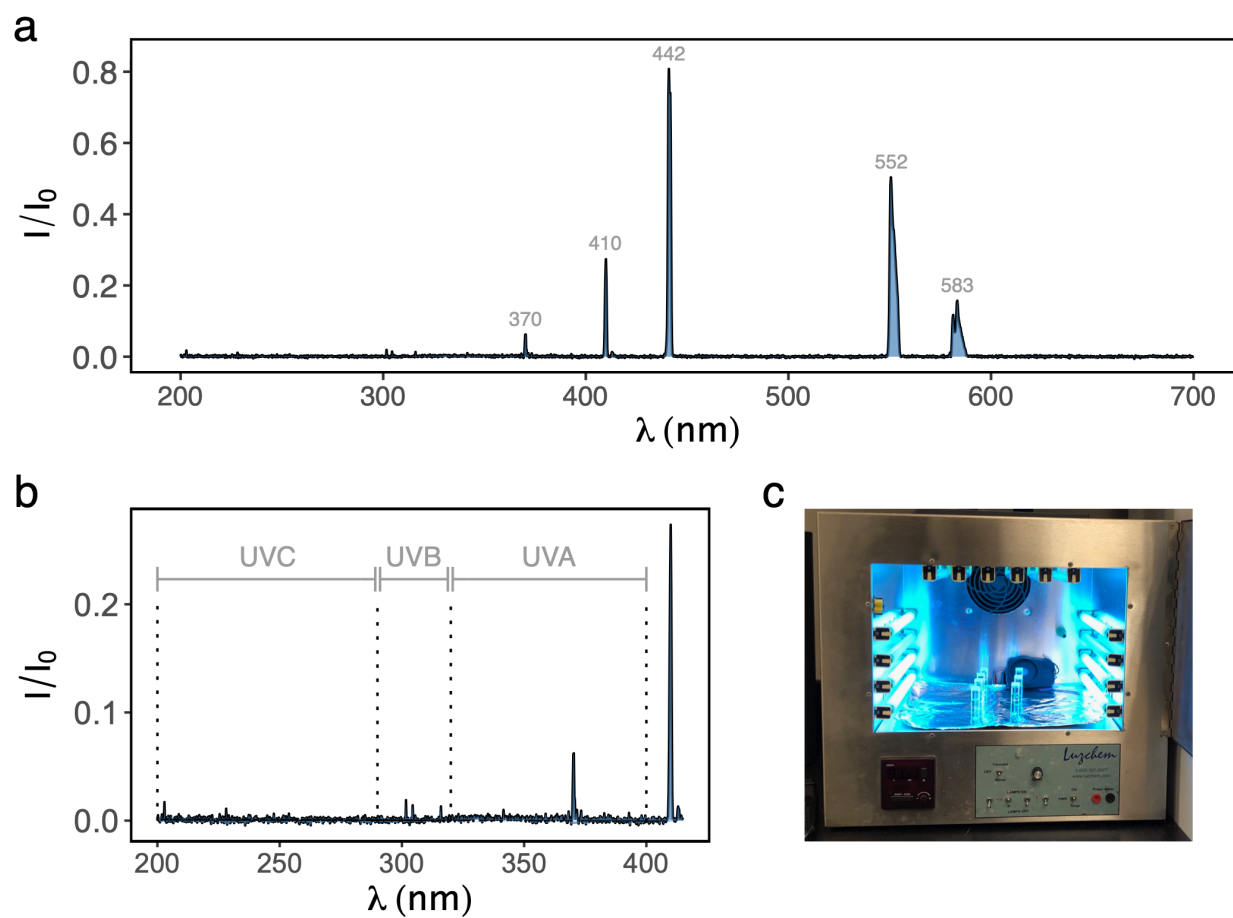
**Fig. S2** | Shift in the fluorescence spectrum/intensity of As-SRHA solutions over time. Triplicate samples are shown by the vertical facets, and pre-incubation time (in days) by the horizontal facets for **a)** As(III) + SRHA and for **b)** As(V) + SRHA.



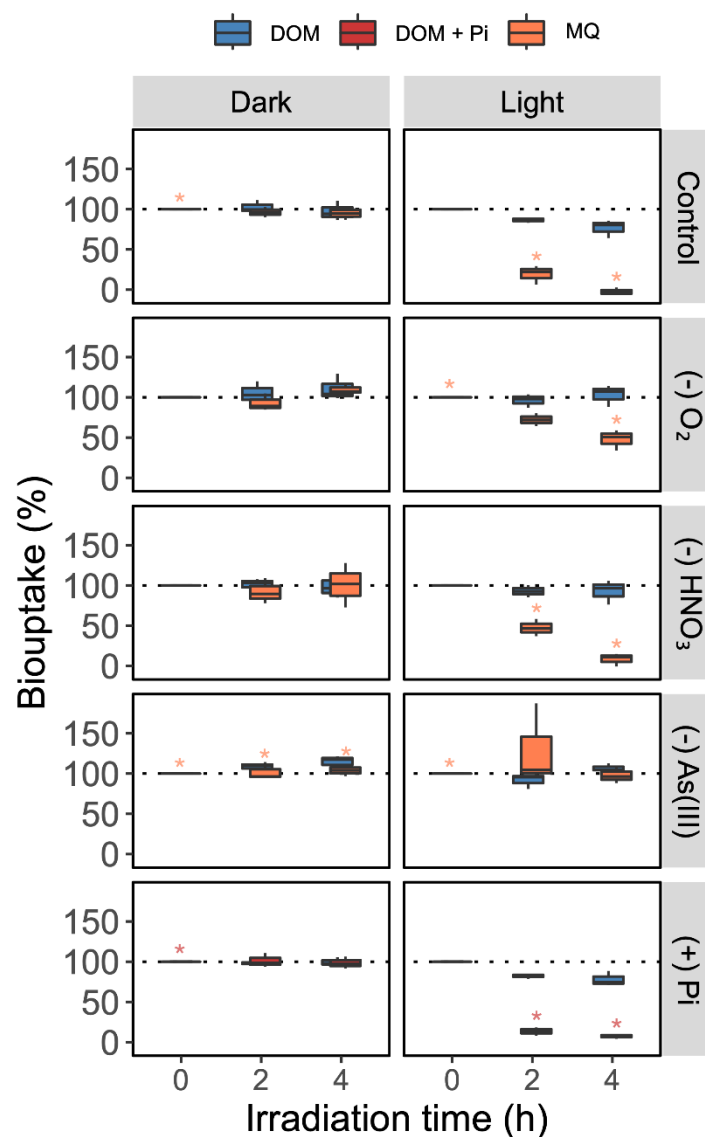
**Fig. S3 |** Cation influence on As(V) binding to SRHA and As(V) bioavailability. Total As(V) and SRHA concentrations were adjusted to 200 nM and 10 mg/L respectively. 10 mM Na or Ca were added both inside of a 500 Da dialysis cut-off membrane and in the external solution, at the same ionic strength. Error bars represent standard deviation of triplicate biological samples. Dotted line represents biosensor output signal of the no-DOM controls at the same ionic strength.



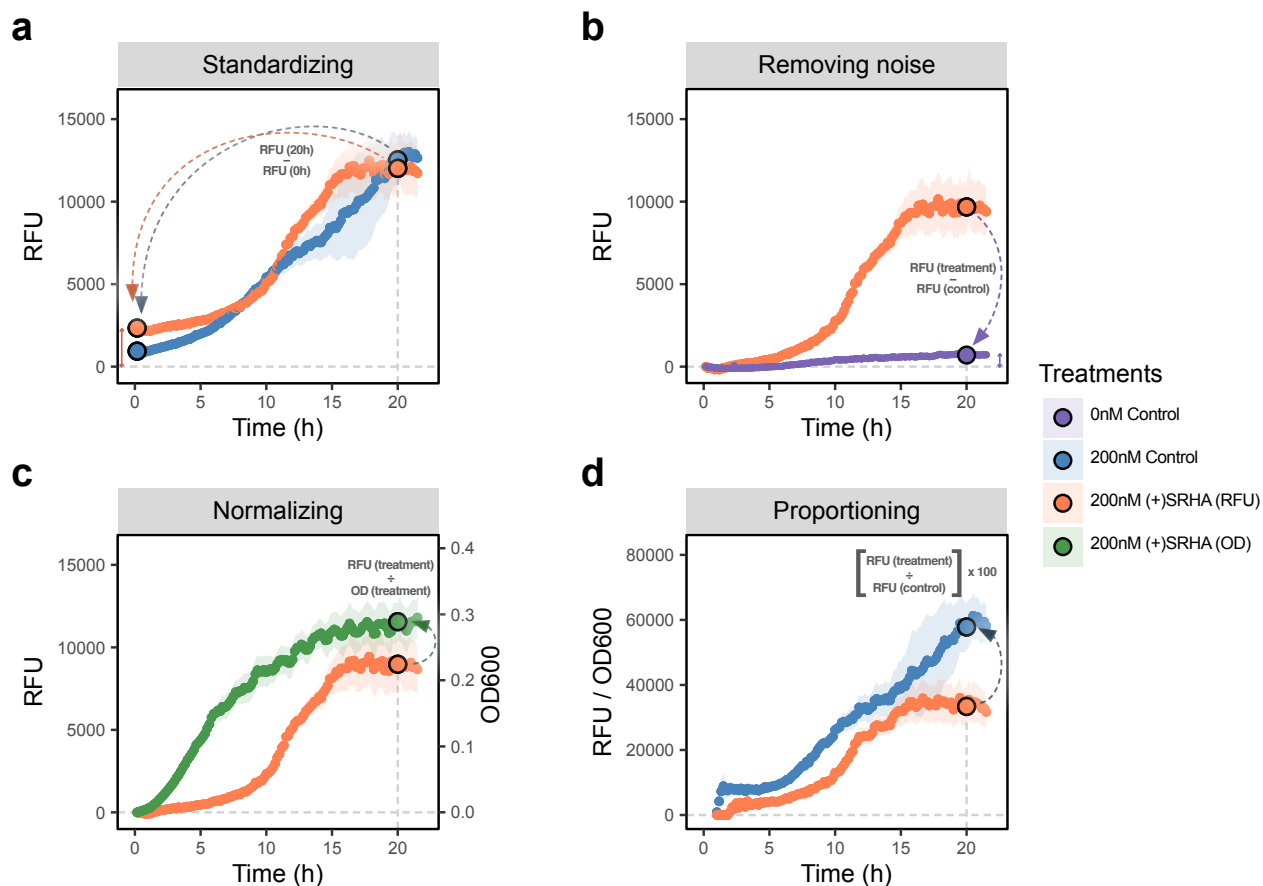
**Fig. S4 |** Cation increase As(V)-SRHA fluorescence intensity. Triplicate samples are shown by the vertical facets. 10 mM Na<sup>+</sup> or Ca<sup>2+</sup> was added to 200 nM As(V) and 10 mM SRHA concentrations



**Fig. S5 |** Photoreactor light spectrum extends to visible components. Spectral components of light are presented from **a)** 200 nm to 700 nm and from **b)** 200 nm to 415 nm. Wavelengths of the peaks and UV ranges are present. **c)** A photo of the Luzchem photoreactor showing the placement of the cuvettes inside.



**Fig. S6 |** Supplemental controls for Fig. 4. Each horizontal facet represents an independent assay reproduced on separate days. Here, a decrease in As(III) uptake over time suggests As(III) oxidation. Biosensor fluorescence was converted to percent biouptake (Y-axis) by comparing output signal of the irradiated treatments to the pre-irradiated (T0) controls. Biosensor endpoints present the mean and standard deviation of three independent biological triplicates. Fig. 4 findings (**Control**) were replicated absent oxygen (**- O<sub>2</sub>**), absent nitric acid in As(III) stock (**- HNO<sub>3</sub>**), and when PO<sub>3</sub><sup>2-</sup> was added to the reaction vessel (**+ Pi**). Addition of As(III) after photoirradiation DOM did not induce As(III) oxidation (**- As(III)**).



**Fig. S7 |** Example of data analysis steps applied to raw data to generate figures. The optical densities at 600 nm (OD600) including the mean (points) and standard deviation (ribbon) of independently grown triplicate biosensor cultures are shown. The output signal of the biosensor is presented in relative fluorescence units (RFU) during the first 22 h of growth immediately following exposure to arsenic. Controls shown here are those used in correction and normalization steps that generate the 25 mg/L T0 As(V) box shown in Fig. 1. Controls are figure-dependent. Step details are further described in the methods section.



**Section B : Supplementary Protocol****Supplementary Method | Anerobic assay controls**

To test whether these findings were an artifact of our experimental design, we performed the assay under anoxic conditions in one treatment and removed  $\text{HNO}_3^-$  from our As(III) stock solution in a separate treatment. In both cases, only in water treatments did we notice significant As(III) photooxidation (**Fig. S6**). We also tested whether As(III) oxidation in water alone was an artifact of the cuvette or of the water used in our laboratory by only introducing As(III) to the cuvettes after irradiation, 0.5h prior to exposing the microbes. As expected, we found no oxidation of As(III) for any of the biological triplicates (**Fig. S6**).

## Section C : Supplementary Table

**Supplementary Table |** Properties of DOM origins used to produce RDA (Fig. 3).

| Condition | Cation | S    | N    | Acetal | Aromatic | Aliphatic | Carboxyl | Phenolic | Q1    | Q2   | pKa  |
|-----------|--------|------|------|--------|----------|-----------|----------|----------|-------|------|------|
| ESFA      | No     | 1.43 | 1.43 | 1      | 30       | 22        | 13.24    | 2.27     | 14.12 | 0.74 | 3.67 |
| ESFA      | Na     | 1.43 | 1.43 | 1      | 30       | 22        | 13.24    | 2.27     | 14.12 | 0.74 | 3.67 |
| ESFA      | Ca     | 1.43 | 1.43 | 1      | 30       | 22        | 13.24    | 2.27     | 14.12 | 0.74 | 3.67 |
| ESFA      | Cu     | 1.43 | 1.43 | 1      | 30       | 22        | 13.24    | 2.27     | 14.12 | 0.74 | 3.67 |
| SRNOM     | No     | 1.78 | 1.78 | 7      | 23       | 27        | 11.21    | 2.47     | 11.2  | 1.6  | 4.16 |
| SRNOM     | Na     | 1.78 | 1.78 | 7      | 23       | 27        | 11.21    | 2.47     | 11.2  | 1.6  | 4.16 |
| SRNOM     | Ca     | 1.78 | 1.78 | 7      | 23       | 27        | 11.21    | 2.47     | 11.2  | 1.6  | 4.16 |
| SRNOM     | Cu     | 1.78 | 1.78 | 7      | 23       | 27        | 11.21    | 2.47     | 11.2  | 1.6  | 4.16 |
| SRFA      | No     | 0.41 | 0.66 | 6      | 22       | 35        | 11.17    | 2.84     | 11.66 | 2.05 | 3.76 |
| SRFA      | Na     | 0.41 | 0.66 | 6      | 22       | 35        | 11.17    | 2.84     | 11.66 | 2.05 | 3.76 |
| SRFA      | Ca     | 0.41 | 0.66 | 6      | 22       | 35        | 11.17    | 2.84     | 11.66 | 2.05 | 3.76 |
| SRFA      | Cu     | 0.41 | 0.66 | 6      | 22       | 35        | 11.17    | 2.84     | 11.66 | 2.05 | 3.76 |
| SRHA      | No     | 0.55 | 1.5  | 7      | 31       | 29        | 9.13     | 3.72     | 9.74  | 4.48 | 4.35 |
| SRHA      | Na     | 0.55 | 1.5  | 7      | 31       | 29        | 9.13     | 3.72     | 9.74  | 4.48 | 4.35 |
| SRHA      | Ca     | 0.55 | 1.5  | 7      | 31       | 29        | 9.13     | 3.72     | 9.74  | 4.48 | 4.35 |
| SRHA      | Cu     | 0.55 | 1.5  | 7      | 31       | 29        | 9.13     | 3.72     | 9.74  | 4.48 | 4.35 |