

Supplementary Materials

Text S1. Determination of Phenol

Reagents and solutions were prepared according to the following. A saturated Na₂CO₃ solution was prepared by dissolving 216 g in a liter of deionized (DI) water. Standard stock solution was prepared by dissolving 100 mg of 2-hydrobenzoic acid in 1 L of DI water. Working standards of 2.5, 5, 10, 20, 30, and 40 mg L⁻¹ of diluted 2-hydrobenzoic acid were prepared from stock solution. A 0.7 mL aliquot of the water-extractable DOM was mixed with 50 µL of Folin–Ciocalteu’s reagent in a 1.5 mL Eppendorf tube and allowed to sit for 3 min at room temperature. A 100 µL of saturated Na₂CO₃ solution and 150 µL of DI water were added to the solution and thoroughly mixed. If a precipitate formed, the solution was centrifuged for 2–3 min at 2000 × g and the absorbance was read immediately. Absorbance was determined at 725 nm against a blank. Samples developed a blue color when phenols were present. The blank was colorless. Calibration curves were prepared and phenol concentration calculated in a mg L⁻¹ 2-hydroxybenzoic acid equivalent. The standard solution was prepared using the same procedure as described for the sample solution. A blank was also prepared following the same procedure as the sample, but deionized water was used in place of the extracted DOM.

Text S2. Determination of hexoses

Reagents and solutions used were prepared according to the following. Anthrone–sulfuric acid reagent was prepared by dissolving 0.2 g of anthrone (analytical grade) in 100 mL of concentrated sulfuric acid. Afterwards, the solution was left to sit for 1 h at room temperature before use. This solution was prepared fresh daily. The standard stock solution was prepared by dissolving 100 mg of glucose in 1 L of DI water. Working standards of 2.5, 5, 10, 20, 30, and 50 mg L⁻¹ of diluted glucose were prepared from the stock solution. A 1 mL extracted DOM sample solution was mixed with 2 mL of anthrone–sulfuric acid reagent. The solution was vortexed and left to sit for 15 min at room temperature. Then, the standard or anthrone-treated sample was transferred to a glass cuvette and its absorbance read at 625 nm against the blank. A calibration curve was prepared and phenol concentration calculated in a mg L⁻¹ glucose equivalent. The standard solution was prepared using the same procedure as that described for the sample solution. A blank of the same volume as the sample was used.

Text S3. Determination of Free Amino Acids

Reagents and solutions were prepared according to the following. Acetate buffer (pH 5.5) was prepared by dissolving 54 g of Na acetate trihydrate in 40 mL of DI water and then adding 10 mL of glacial acetic acid. Afterwards, the solution pH was adjusted to 5.5 with NaOH. Ninhydrin reagent was prepared by dissolving 2 g of ninhydrin and 0.3 g of hydrindantin in 75 mL of 2-hydroxy ethanol. The solution was purged with N₂ for 30 min, after which 25 mL of acetate buffer (pH 5.5) was added. This solution was prepared fresh daily with limited air exposure. The dilutant was prepared by mixing equal amounts of 95% ethanol with DI water. Standard stock solution was prepared by mixing 1000 µmol leucine solution in 1 L of DI water. Working standards of 20, 40, 60, 80, and 100 µmol L⁻¹ of diluted 2-hydrobenzoic acid were

prepared by diluting the stock solution. Two milliliters of DOM sample solution was mixed with 1.25 mL of the ninhydrin reagent in 10 mL glass tubes. The tubes were capped with Teflon-lined screw caps and kept in a 95 °C water bath for 25 min. The tubes were cooled to room temperature in another water bath, and then, 4.5 mL of dilutant was mixed with the cooled solution. The standard or treated sample was transferred to a glass cuvette and its absorbance read at 570 nm against a blank. A calibration curve was prepared and amino acid concentration was calculated in a $\mu\text{mol L}^{-1}$ leucine equivalent. The standard solution was prepared with the same procedure described for the sample solution. Deionized water of the same volume as the sample was used as a blank.

Text S4. Protein Determination

Three reagents and solutions used in the analysis were prepared according to the following. Standard stock solution was prepared daily by dissolving 100 mg of bovine serum albumin (BSA) in 1 L of DI water. Working standards of 2.5, 5, 10, 15, 20, and 25 mg L^{-1} of diluted BSA were prepared by diluting the stock solution. Bradford protein reagent was purchased and stored under refrigeration until use. Subsequently, protein was quantified according to the following. Bradford protein reagent in the amount 0.5 mL was added into a spectrophotometer cuvette and mixed with 0.5 mL of DOM sample solution, standard solution, or blank solution. The mixture was left to sit for 5 min at room temperature and then absorbance was read at 620 nm against the blank. A calibration curve was prepared and protein concentration was calculated in a mg L^{-1} BSA equivalent.