

ISOLATION AND CHEMICAL CHARACTERIZATION OF HIGH-MOLECULAR WEIGHT DISSOLVED ORGANIC MATTER (HMW-DOM) FROM FRESHWATER LAKES IN JAPAN, BY USE OF TANGENTIAL FLOW ULTRAFILTRATION.

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Since Benner et al. (1992) succeeded in separation of high molecular weight dissolved organic matter (HMW-DOM) from a large volume of ocean water by use of tangential flow ultrafiltration technique (TFF), a great many works have been published on isolation and characterization of DOM in seawater and coastal waters (*e.g.*, Mannino and Harvey, 2000; Ogawa and Tanoue, 2002). However, only limited works have been done on freshwater regimes (Repeta et al., 2002). It led us to examine chemical characterization of HMW-DOM from freshwater lakes of different trophic status (eutrophic L.Suwa, mesotrophic L.Kizaki and L.Nakatsuna and oligotrophic L.Aoki).

Tens to hundreds mg of high molecular-weight dissolved organic matter (HMW-DOM) were isolated from 80-100 L of water samples from four freshwater lakes in Nagano Prefecture, Japan. Apparatus used was a Millipore Ltd. Pellicon Lab Cassette TFF equipped with a Millipore Ltd. PCAC and/or PLAC filter of nominal molecular cut-off of 1000 Dalton. Retention efficiency of TFF was assayed by filtrating aqueous solution (2 mg/L) of cyanocobalamine (MW=1355). It revealed that about 50 % of this size of molecules would be retained, when 100 L of sample water is condensed to 1 L (concentration factor = 100) (Yoshiyama et al., 2003). The DOM solution concentrated by TFF was further condensed by stirring ultrafiltration (Amicon YM1: MCO=1000 Dalton) and finally lyophilized.

The puffy powder accounted for 44-70% of the total dissolved organic carbon (DOC) in the sample waters. Chemical nature of the HMW-DOM was characterized by CN elemental, carbon and nitrogen stable isotopes, and lipid analyses.

Stable isotope ratios of carbon ($\delta^{13}\text{C} = -29.3 \sim -23.9 \text{ ‰}$ against PDB) and nitrogen ($\delta^{15}\text{N} = +0.5 \sim +8.6 \text{ ‰}$ against air) are heavier for HMW-DOM from a eutrophic lake (-24.1 ‰ and +8.6 ‰) and lighter for HMW-DOM from an oligotrophic lake (-29.3 ‰ and

+0.5 ‰). The data from two mesotrophic lakes (-27.4 ‰: +1.3 ‰ and -26.5 ‰: +4.1 ‰) fell between the two extremes. On the other hand, the C/N elemental ratio is low (11.1) for a eutrophic lake water and high for meso- to oligotrophic lake waters (12.6 to 17.4). Fig. 1 shows the correlation between $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$, where data dispersed on a straight line, suggesting a variable contribution of autochthonous microbial origin of organic matter to HMW-DOM according to the trophic status of the lake.

Fatty acids associated with HMW-DOM were compared with those of particulate matter (PM) and plankton collected in parallel. Fatty acids in HMW-DOM are mostly short-chain saturated and unsaturated homologues, being indicative of microbial origin. Relative proportion of branched bacterial fatty acids was also apparently large in HMW-DOM.

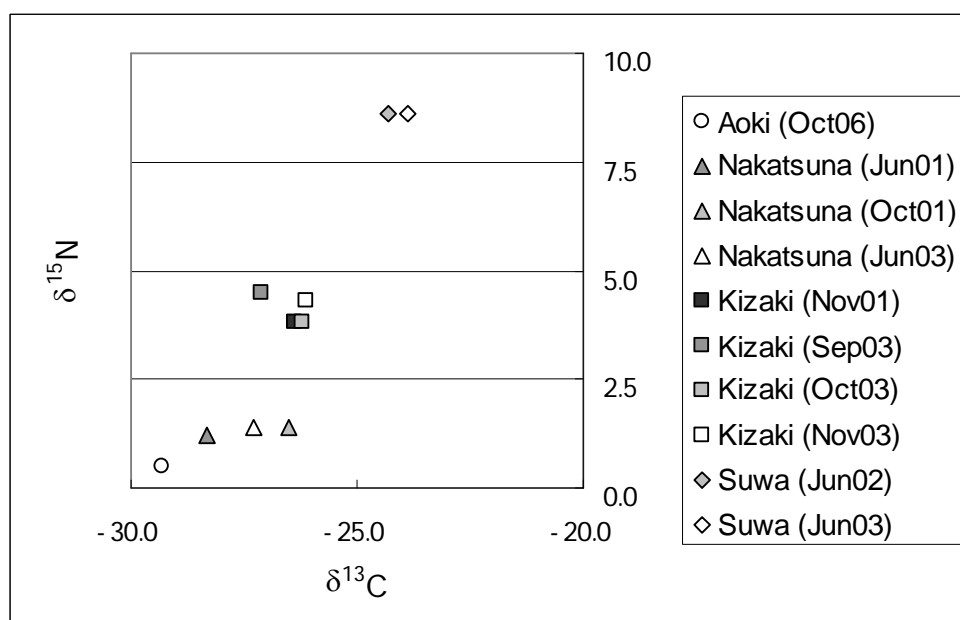


Figure 1. Correlation of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of HMW-DOM separated from freshwater lakes.

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