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Levels of Chemical Versus Biological Methylation of Mercury in Sediments

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Conversion to methylmercury in soils and sediments amplifies the environmental hazards of inorganic mercury (Hg) pollutants (National Academy of Sciences, 1978; Jeffries, 1982; Robinson and Tuovinen, 1984). The cited reviews imply that the Hg methylation process is biochemical or, at least, requires a biochemically produced methyl donor such as methylcobalamin. However, Rogers (1976; 1977) reported higher levels of methylmercury synthesis in Hg-spiked and steam-sterilized soils as compared to similarly spiked, but non-sterile soils. More recently, Nagase et al. (1982; 1984) reported on the ability of fulvic acid fractions from river sediment and leaf mould to methylate Hg and identified 2,6-di-tert-butyl-4-methyl-phenol as one of the naturally-occurring non-biochemical methyl donors. Although these pioneering publications have not resolved the environmental significance of abiotic versus biochemical Hg methylation, their superficial reading may create the impression that abiotic Hg methylation predominates. Our measurements reported here allow a direct quantitative comparison of the two processes.

For the purpose of this dicussion, chemical Hg methylation is defined as the portion of the total that occurs also in the absence of a metabolically active microbial community. Two approaches were used in comparing the contributions of biochemical versus chemical Hg methylation: its level in anoxic saltmarsh sediments incubated at favorable pH and temperature was compared to methylation at pH and temperature values hostile to life. Hg methylation in normal sediment was also compared to that in steam-sterilized sediment incubated either at normal or elevated temperatures.

MATERIALS AND METHODS

Low salinity (0.4%) sediment was collected form saltmarsh mudflats in Cheesequake State Park (New Jersey, U.S.A.). On dry weight basis the sediment contained 20% organic matter, had a pH of 6.8-7.0 and a redox potential of -220 mV (Compeau and Bartha, 1984).

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Samples were collected at low tide to a depth of 20 cm with a 5 cm diameter hand corer (Wildco Instruments, Saginaw, MI) equipped with plastic core liners. Immediately after sampling, the core liner was sealed at both ends and wrapped in plastic to maintain the redox potential of the sediment. Within one hour of collection, samples were placed in an anaerobic chamber (Model 6500 PACE, Labline Instruments, Melrose Park, IL), having a gas atmosphere of 3% H_2, 5% CO_2 and 92% N_2. Within the chamber, sediment samples were slurried in deoxygenated water and subsequently distributed into 75 ml test tubes sealed with teflon-lined screw caps. The tubes contained 60 ml sediment slurry (16-17 g dry sediment). Sediment pH was determined by an Accumet (Fisher Scientific Co., Springfield, NJ) instrument and, in some cases, adjusted to pH 2.0 or 14.0 using HCl or NaOH, respectively. Prior to spiking with 10 ppm $HgCl_2$ (calculated on dry sediment basis) some samples were steam-sterilized (121°C for 1 h), others were used without sterilization. Sterilized samples were treated aseptically throughout the incubation period. After 3 days at either 25 or 60°C, the methylmercury formed was extracted from the sediment according to the procedure of Longbottom et al. (1973) and determined by gas chromatography (Compeau and Bartha, 1983). The data represent the average of duplicate determinations.

RESULTS AND DISCUSSION

In unspiked sediment, methylmercury was below the detection limit (3 ng/g). In spiked sediment during a 3-day period, 288 ng methylmercury per g dry sediment was synthesized at 25°C and pH 7 (Table 1). These temperature and pH values were typical during the summer season and similar levels of methylmercury synthesis were routinely observed in this low-salinity anoxic saltmarsh sediment (Blum and Bartha, 1980; Compeau and Bartha, 1984). No methylmercury synthesis was detected when the sediment pH was adjusted to 14, regardless of the incubation temperature. Very low amounts of methylmercury were synthesized at pH 2.0 and in steam-sterilized sediment, the highest value (21 ng/g) being only 7% of the level in the biologically active sample. Incubation at 60°C and pH 7 yielded 47 ng/g methylmercury, but this set of conditions did not exclude thermophilic microbial activity (Doetsch and Cook, 1973). A pre-sterilized sample yielded only 9 ng/g methylmercury under the same incubation conditions.

The presented data show that chemical methylation definitely occurs in anoxic saltmarsh sediment, but the resulting methylmercury levels are approximately one order of magnitude lower than those formed by biochemical Hg methylation. The amounts of methylmercury formed by chemical methylation in our experiments are comparable to those reported by Rogers (1976). He found that up to 6.4 ng methylmercury per g steam-sterilized agricultural soil was produced from a 500 μg/g Hg(NO) spike. There is, however, a very large difference between the maximal methylmercury levels (4.3 ng/g) measured by Rogers (1976) in biologically active aerobic agricultural soils and those found by us in biologically active anaerobic saltmarsh sediment (228 ng/g). In this context,

Table 1. Synthesis of methylmercury in anoxic saltmarsh sediment spiked with 10 g/g HgCl and incubated for three days under conditions permitting or restricting biological activity.

Temperature °C	pH	Steam-sterilized	Methylmercury formed) (ng/g dry sediment)
25	7	no	288
25	7	yes	2
25	2	no	10
25	14	no	nd
60	7	no	47
60	7	yes	9
60	2	no	15
60	14	no	nd

The letters "nd" stand for "not detected." Detection limit was 3 ng/g.

it should be noted that while some microorganisms synthesize methylmercury, others are capable of destroying it (Robinson and Tuovinen, 1984), and the actual methylmercury levels measured are determined by the respective rates of the two opposing processes. These, in turn, are influenced by the prevailing environmental conditions. In saltmarsh sediments, aerobic conditions inhibited the synthesis and promoted the destruction of methylmercury, while anaerobic conditions had the opposite effect (Compeau and Bartha, 1984). This may explain the low levels of methylmercury found in biologically active aerobic agricultural soils (Rogers, 1976). Sterilization destroys not only the methylating but also the methylmercury-degrading microorganisms. Thus, the small amounts of methylmercury formed by chemical methylation in sterilized soils and sediments are protected from biodegradation. These considerations satisfactorily explain the results of Rogers (1976;1977) who found higher methylmercury levels in sterilized soils as compared to non-sterile controls. In most aquatic sediments, only the upper few mm are aerobic and the rest of the sediment is in an anaerobic state. Anaerobic conditions favor the formation and inhibit destruction of methylmercury, and in anaerobic sediments over 90% of the methylmercury is formed by biochemical mechanisms. In good agreement with our findings is an early report by Jensen and Jernelöv (1969). In $HgCl_2$-spiked, presumably anaerobic sediment samples from a freshwater aquarium, these authors observed the formation of 180 and 440 ng/g methylmercury in 5 and 10 days, respectively, but in steam-sterilized sediment, methylmercury concentration was below background level (40 ng/g).

In conclusion, our measurements on biologically active and inactive samples along with the analysis of previously published data indicate that in anaerobic sediments abiotic mechanisms may form up to 21 ppb of methylmercury, while biochemical methylation under similar conditons forms up to 288 ppb. As compared to

biochemical methylation, the environmental significance of abiotic Hg methylation is minor.

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