## SHORT COMMUNICATION

# Effects of mercury compounds on soil microbes

### Summary

Even low concentrations of mercury compounds in agar media strongly decreased the number of soil microbes that could be isolated on these media. Mercury compounds added to the soil in very high concentrations inhibited CO<sub>2</sub>-evolution, dehydrogenase activity, and nitrification. In contrast, the number of microbes increased somewhat in a clay soil treated with HgCl<sub>2</sub>. Phenylmercury acetate had a stronger inhibiting effect than HgCl<sub>2</sub>. In sandy soil the microbial processes were inhibited more strongly than in clay soil.

Mercury compounds present in the soil or added in low concentrations are not expected to seriously disturb organic-matter breakdown, nitrogen mineralization, or the soil-microbe numbers.

#### Introduction

Local heavy pollution of the soil with mercury in the Netherlands and alarming reports from Sweden and elsewhere about the formation of highly toxic methyl mercury compounds in under-water sediments, prompted us to start investigations into the extent of mercury methylation by soil microbes. We also investigated the effects of some mercury compounds on soil-microbe numbers and general metabolic activities. A report of the latter is given below.

### Methods and materials

Numbers of microbes were counted by means of the soil-dilution plate method on soil extract agar media.

Dehydrogenase activity was measured as the amount of formazan formed during 24 h incubation at 37°C with and about glucose added. This was achieved by the addition of tetrazoliumchloride to waterlogged soil samples plus CaCO<sub>3</sub>. The method used was that of Casida <sup>1</sup>, slightly modified.

The CO<sub>2</sub> liberated from the soil during incubation at 29°C was captured in 1 N NaOH and titrated; the sum of the amounts of ammonia and nitrate in the soil was determined in 1 N NaCl extracts from the amount of NH<sub>3</sub> distilled, using the method of Cotte and Kahane<sup>2</sup>. Ammonia was determined by distillation following the addition of MgO; nitrate was found by difference.

Some properties of the soils used in the investigation are given in Table 1. The two clay soil samples were taken from a polder in the area of the river Rhine estuary. The dune sand was taken from a bulb field where mercury

TABLE 1
Properties of the soils used in the experiments

| Soil                | Organic<br>matter,<br>% | Mercury,<br>ppm | Fraction<br><16 µm | pH-KCl     |
|---------------------|-------------------------|-----------------|--------------------|------------|
| Clay 1              | 6.5                     | 1.1             | 45                 | 7.7        |
| Clay 2<br>Dune sand | 5.6<br>1.5              | 0,45<br>0.04    | 52<br>—            | 7.7<br>7.5 |
| Diluvial sand       | 3.1                     |                 | _                  | 4.9        |

compounds had been used for disinfection and the sample of the diluvial sand soil came from the grounds of the Institute for Soil Fertility. Samples used in the experiments were usually air dried and rewetted with deionized water or solutions of mercury compounds.

### Results

- (a) Effect on the number of microbes. Addition of HgCl<sub>2</sub> (100 ppm) or Hg (NO<sub>3</sub>)<sub>2</sub> (150 ppm )to clay soil 1 gave somewhat higher numbers of microbes than were present in the untreated control. However, when mercury compounds were added to the plating media there was a strong decrease in the numbers of microbes: addition of HgCl<sub>2</sub> (10 ppm), Hg(NO<sub>3</sub>)<sub>2</sub> (12.5 ppm), phenylmercury acetate (PMA, 1.3 ppm) or methylmercury chloride (0.5 ppm) reduced - in this order - the number to between 10 per cent and less than 0.01 per cent of the colonies as counted on the plates without mercury. (b) Effect on dehydrogenase activity. Dehydrogenase activity was inhibited by 70 per cent in the case of the clay soil and by 90 to 100 per cent for the sandy soil, immediately after addition of HgCl<sub>2</sub> (100 ppm). Addition of PMA (100 ppm) similarly gave 97 to 100 per cent inhibition in all samples. The diluvial sand showed strong inhibition by 5 ppm HgCl<sub>2</sub> or PMA. After ten days incubation, however, the activity was already partially restored; simultaneous addition of glucose (200 ppm) to the soil appeared to give a more rapid recovery of the relative activity.
- (c) Effect on the carbon and nitrogen mineralisation. The CO<sub>2</sub> evolution from both the clay soil 2 and the dune sand, incubated during six weeks, was lowered by 20 per cent when HgCl<sub>2</sub> (100 ppm) had been added. The ammonification and nitrification during six weeks incubation of three soils treated with HgCl<sub>2</sub> or PMA (10 and 100 ppm) as compared with the controls is shown in Table 2.

# Discussion and conclusion

Mercury compounds, used as pesticides, normally enter the soil in amounts of 1 g/ha up to 200 g/ha (0.0005–0.1 ppm), which is far below the level where, according to the results mentioned above, a serious disturbance of organic-matter breakdown or nitrogen mineralization may be expected. In the Rhine-

TABLE 2

Effect of mercury compounds on nitrogen conversions in the soil as percentages of the controls

| Mercury compound            | Ammonification, % |              | Nitrification, % |              |
|-----------------------------|-------------------|--------------|------------------|--------------|
| added to the soil           | Clay<br>soils     | Dune<br>sand | Clay<br>soils    | Dune<br>sand |
| HgCl <sub>2</sub> (10 ppm)  | 107               | 95           | 108              | 95           |
| HgCl <sub>2</sub> (100 ppm) | 85                | 76           | 60               | 5            |
| PMA (10 ppm)                | 105               | 81           | 106              | 43           |
| PMA (100 ppm)               | 58                | 81           | 6                | 5            |

delta area much heavier pollution of the clay soil with mercury is found, namely up to about 10 ppm. The form is which mercury is present here is not known but, since organomercury compounds such as PMA are rather quickly 'mineralized', it is likely that most of it is in an inorganic form. Thus no serious reduction in the number of microbes or in carbon and nitrogen mineralization is to be expected.

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#### References

- 1 Casida, L. E., Soil Sci, 98, 371-377 (1964).
- 2 Cotte, J. and Kahane, E., Bull. Soc. Chim. France, 542-544 (1946).