

The study to evaluate 6 chelating compounds—NTA, EDTA, tartaric acid, citric acid, oxalic acid and thiourea—for their ability to inhibit urease activity in a sandy clay loam showed that these compounds had little, if any, effect on the urea hydrolysis when used at 50 $\mu\text{g/g}$ of soil concentration. The percentage inhibition of urease activity by the chelating compounds tested ranged from 0 to 10% in a 5 hr incubation test.

Evaluation of Some Chelating Compounds for Retardation of Urea Hydrolysis in Soil

By K. L. SAHRAWAT*

*Division of Agricultural Chemicals,
Indian Agricultural Research Institute,
New Delhi 110 012*

Introduction

Urea is the most important nitrogen fertilizer in the world¹ and is fast replacing ammonium sulfate. Most of the problems encountered in the use of this fertilizer like damage to seeds and the growing seedlings, ammonium toxicity, nitrite toxicity and loss due to ammonia volatilization are caused by the rapid hydrolysis of urea by soil urease.^{2-5,8}

There is an obvious need for finding solution to these problems caused by rapid hydrolysis of urea for promoting the efficient use of this fertilizer. Perhaps solution to most of these problems could be obtained at least partly if the soil urease is inhibited to retard the rate of urea hydrolysis in soil. Attempts have been made in the past for inhibiting the soil urease activity by the use of different chemical agents.^{10,11,13,14} Recently Douglas and Bremer⁷ have developed a rapid method for evaluating different compounds for inhibiting urease activity in soils and this has facilitated testing of many compounds as urease inhibitors, which was earlier hindered by a lack of suitable technique for this purpose. Using this technique, Bremner and Douglas¹ made studies to evaluate more than 100 compounds as inhibitors of urease activity in soils and concluded that the possibility that the various problems encountered owing to rapid hydrolysis of urea can be reduced by application of urease

inhibitors to soils and feedlots deserves attention.

The objective of the present study was to test some chelating compounds for their ability to retard urea hydrolysis in soils by inhibiting urease activity. The rationale behind evaluation of chelating compounds for inhibiting soil urease activity was that the chelating compounds are known to interfere with the functioning of enzymes involving sulphahydral (-SH) group like urease by chelating the functions having -SH groups. Also there are evidences to show that the chelating compounds have the ability to chelate with the metallic ions in enzymatic activities responsible for functioning of nitrifiers, mainly *Nitrosomonas*.⁹ The effects of chelating compounds on soil urease have not been investigated and thus prompted evaluation of some chelating compounds for the purpose.

Experimental

The soil used in the study was a sandy clay loam alluvial soil from the farm of the Indian Agricultural Research Institute, New Delhi. The surface (0-15 cm) soil sample collected was air-dried and ground to pass a 2-mm sieve before use. The soil tested: pH, 7.6; total N, 0.07%; organic C, 0.62%; CEC, 11.6 me/100 g soil. Soil analyses were done as described earlier.¹² The soil has urease activity of 8.2 μg urea hydrolyzed per g of soil per hr at 30°C.

Analytical reagent grades of NTA (nitrilotriacetic acid), EDTA (ethylene diamine tetra acetic acid), tartaric acid, citric acid, oxalic acid and thiourea were used for

*Present address: Soil Chemist, International Cross Research Institute for the Semi-Arid Tropics (ICRISAT), 1-11-256, Begumpet, Hyderabad 500 016, A.P., India.

testing their abilities to retard soil urease activity. All the compounds were added at a concentration of 50 ppm of soil, phenylmercuric acetate was used as a standard urease inhibitor for comparison purposes.

The following 5 hr incubation test described by Douglas and Bremner⁷ was adopted to study the effects of different chelating compounds on the soil urease activity. Soil samples (10 g) were incubated with 1 ml of toluene and 10 ml of urea solution in water containing 10 mg of urea-N at 30°C for 5 hr to determine the effect of the test compounds on urea hydrolysis at a concentration of 50 ppm of soil. Urea hydrolysis by soil urease was measured by determining the amounts of urea remaining unhydrolyzed (from the total added) by colorimetric method after extraction of the incubated soil samples with 2 M KCl containing phenyl acetate urease inhibitor (Douglas and Bremner⁶). All the compounds tested were applied in their aqueous solutions. For NTA and EDTA, their corresponding sodium salts viz Na₃NTA and Na₃EDTA were used and their concentrations were used on the basis of the free acids, viz. NTA and EDTA. All the determinations were made in duplicates and the percentage inhibition of soil urease activity by different compounds calculated.

Results and Discussion

The results of the actions of different chelating compounds and phenylmercuric acetate on inhibiting soil urease activity are presented in Table 1, which indicate that none of the chelating compounds tested were effective in retarding urease activity. Thiourea inhibited urease activity to the extent of 10 per cent only and other compounds showed little ability to inhibit urease activity. However, phenyl mercuric acetate was quite effective in inhibiting urease activity, resulting in 75 per cent retardation of the soil urease activity in the 5 hr incubation test. In another incubation experiment, it was observed that urea hydrolysis in soil samples was complete by one week both with or without the addition of the chelating compounds, as no urea could be detected after one week, thus confirming the ineffectiveness of these compounds in

TABLE 1—EFFECTS OF CHELATING COMPOUNDS ON SOIL UREASE ACTIVITY*

Compound	inhibition of urease activity, %
NTA	2
EDTA	1
Tartaric acid	0
Citric acid	0
Oxalic acid	0
Thiourea	10
Phenylmercuric acetate	75

*All the compounds were tested at a concentration of 50 ppm of soil. The urease activity of the soil without addition of the chelating compound was 8.2 µg urea hydrolyzed/g soil/h at 30°C.

inhibiting soil urease. However, phenyl mercuric acetate showed 62 per cent inhibition of the soil urease activity after one week under similar conditions.

The results of the present study suggest that the chelating compounds tested may be of little value in retarding urea hydrolysis in soils.

REFERENCES

1. Bremner, J. M. and Douglas, L. A., *Soil Biol. Biochem.* 3 (1971), 297.
2. Cooke, G. W., In *Phosphorus in Agriculture Bull. Doc. No. 53*, (1969), 1. International Superphosphate and Compound Association, Paris.
3. Court, M. N., Stephen, R. C. and Waid, J. S., *Nature, London* 194 (1962), 1263.
4. Court, M. N., Stephen, R. C., and Waid, J. S., *J. Soil Sci.* 15 (1964a), 42.
5. *Ibid.*, (1964b), 49.
6. Douglas, L. A. and Bremner, J. M., *Proc. Soil Sci. Soc. Amer.* 34 (1970), 859.
7. Douglas, L. A. and Bremner, J. M., *Soil Biol. Biochem.* 3 (1971), 309.
8. Gasser, J. K. R., *Soils Fertel.* 27 (1964), 175.
9. Lees, H., *Biochem. J.* 52 (1952), 134.
10. Pugh, K. B. and Waid, J. S., *Soil Biol. Biochem.* 1 (1969a), 195.
11. *Ibid.*, (1969b), 207.
12. Sahrawat, K. L., *Soil Biol. Biochem.* 9 (1977), 173.
13. Volk, G. M., *J. Agric. Fd. Chem.* 9 (1961), 280.
14. Waid, J. S. and Pugh, K. B., *Chem, Industr.* (1967), 71.