2411 JA 288

Plant and Soil 78, 401–408 (1984). Ms. 5600

Effects of temperature and moisture on urease activity in semi-arid tropical soils*

K.L. SAHRAWAT**
International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), ICRISAT Patancheru P.O., Andhra Pradesh 502 324, India

Received 26 July 1983. Revised October 1983

Key words Alfisol Buffer and non-buffer methods Relative humidity Soil urease protection Urease assay Urea hydrolysis Vertisol

Summary Studies of urease activity in an Indian Vertisol and Alfisol using both buffer (THAM pH 9.0) and non-buffer methods for assay of the urease activity showed that activity increased with increase in temperature from 10°C to a maximum at 60°C (Vertisol) and 70°C (Alfisol). Further increase in temperature decreased urease activity which was nearly totally inhibited at 100°C. Urease activity was not detected in soil samples collected in late summer when the soil moisture content was far below −15 bar pressure. Urease activity increased with increase in moisture content up to field capacity and remained constant with further increase in moisture content. The relevance of these findings to the ICRISAT improved management practices for Vertisols, which involve seeding of crops into dry soil just before the onset of rains is discussed.

Introduction

Detailed studies have been made of the effects of temperature and moisture on urease activity in temperate soils[2]. However, tropical soils have received especially little attention with regard to the effects of temperature and moisture on urease activity over the full range of these factors, despite the dominance of urea as the main nitrogenous fertilizer[11], and the wide variation in soil temperature and moisture regimes in the seasonally-dry tropics.

In the semi-arid climate, temperature and moisture content of the soil undergo marked changes during the year. In India, some of the most marked changes in soil moisture occur at the time of the onset of the Southwest monsoon; before its arrival, the surface soil is usually extremely dry because of an intensely dry and hot period of several months. At ICRISAT Center, temperatures of the surface soil (0–5 cm) can be as high as 55°C or more during the two summer months (April–May) before the onset of rains but, with the arrival of the rains, the

* Approved as Journal Article No. 288 by the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT).
**Address till July 1984: Department of Soil Science, University of Wisconsin, Madison, WI 53706, USA.

surface soil temperatures decrease considerably, and may range from 20°C to 35°C during the rainy season (mid-June through October)[7].

The interaction of temperature and moisture are particularly important for new agronomic practices proposed for some deep Vertisols in India. Where the rainfall during the rainy season is assured on these soils, the establishment of the rainy-season crop is much improved by sowing seed into a dry seed-bed just before the arrival of the monsoonal rains instead of waiting until after the rains[8].

However, the effect of temperatures in excess of 40°C on urease activity has received little attention, and existing results are conflicting. For example, Zantua and Bremner[20] indicated an optimum temperature of 70°C for maximum soil urease activity, with a rapid decrease with further increase in temperature to 80°C. In contrast, Dash *et al.*[6] reported an optimum temperature of 47°C. There is no apparent reason for these markedly different optima, but different assays were used for soil urease. Dash *et al.*[6] used a buffer method[16] (THAM buffer pH 9.0) for urease assay whereas Zantua and Bremner[20] used a non-buffer method[18].

Little is known of the effect of especially low moisture contents on soil urease activity in tropical soils. Dalal[5] found that urease activity in Trinidad soils increased with moisture content from 25% to 50% water holding capacity (WHC) beyond which it decreased slightly in one soil and considerably in another soil. On the other hand, Zantua and Bremner[20] observed that urease activity in Iowa soils was not significantly affected by soil moisture content.

The objective of this study was to characterize the effects of soil moisture and temperature on urease activity in the ranges of importance to the semi-arid tropics on soils that are representative of the two main soils at ICRISAT Center, namely Vertisols and Alfisols.

Materials and methods

Soils

The soils used were Vertisols and Alfisols. Surface (0–15 cm) samples were collected from the Kasireddipalli series (very fine, montmorillonitic, isohyperthermic Typic Pellusterts) and the Patancheru series (clayey-skeletal, mixed isohyperthermic Udic Rhodustalfs). The soils had been used for growing sorghum and maize for the past two years. The soil samples were air-dried, ground and sieved to pass through a 2 mm screen before use. Some characteristics of the soils are given in Table 1; for these analyses pH was measured with a glass electrode using soil to water ratio of 1:2, Organic C and total N were determined as described by Walkley and Black[17] and Bremner[1] respectively. Particle size analysis was made by the hydrometer method[4].

Effect of temperature on urease activity

The effect of temperature on soil urease activity was determined by incubating the soil samples at 60% WHC moisture and at temperature ranging from 10°C to 100°C using both

Table 1. Some characteristics of the soils

Characteristic	Vertisol	Alfisol
pH (1:2 water)	7.5	5.3
Organic C (%)	0.47	0.37
Total N (%)	0.057	0.053
Clay (%)	53	29
Silt (%)	26	8
Sand (%)	21	63
Water holding capacity (WHC) (%, W/W)	52	34
Moisture content at field capacity (%, W/W)*	40	24
Moisture content at −15 bar pressure (%, W/W)	19	5
Urease activity**	14.8	5.1

* This corresponds to moisture content at −0.10 bar and −0.33 bar pressure respectively for Alfisol and Vertisol.
**Urease activity expressed as μg urea N hydrolysed per g of soil per h at 37°C at 60% WHC moisture content.

buffer and non-buffer methods. The non-buffer method used was that described by Zantua and Bremner[18]. The buffer method (THAM buffer, pH 9.0) of Tabatabai and Bremner[16] was modified in that the NH_4^+-N produced after incubation of the soil samples was measured in the filtered extracts rather than by distillation of the soil suspension with MgO[12].

Effect of moisture on urease activity

For studying the effect of moisture on urease activity in soils, the non-buffer method[18] was used. The non-buffer method was selected because the method is more precise and it provides better index of urea hydrolysis in soils under natural conditions[18] than the buffer method. In another experiment, the effect on urea hydrolysis of urea on absorption of moisture from a humid atmosphere by initially air-dry soil samples was studied by incubating the samples at 37°C for 7 days in humid (approximately 100% RH) and non-humid environments (20–30% RH). The humid environment was created by placing a small beaker containing water in the incubation vessel. For the non-humid environment incubation was carried out in dry containers but with the atmosphere at ambient humidity (20–30% RH). Urea in powder form was mixed with the soil samples at a rate of 1000 μg N/g soil without changing the water content of the samples. Soil samples were analyzed for the urea remaining unhydrolyzed after 1, 2, 3, 5 and 7 days of incubation[9].

All the experiments reported were done at least in duplicate.

Results and discussion

Effect of temperature on soil urease

Urease activity of soil samples for both soils was always higher when measured with the buffer method, for the entire temperature range (Fig. 1 and 2). These results are in agreement with those reported by Zantua and Bremner[18] who found that the levels of soil urease activity in Iowa soils were higher when measured using the buffer method. The buffer used (THAM, pH 9.0) brings pH to the optimum range for urease activity.

Urease activity of the Alfisol increased with increase in temperature

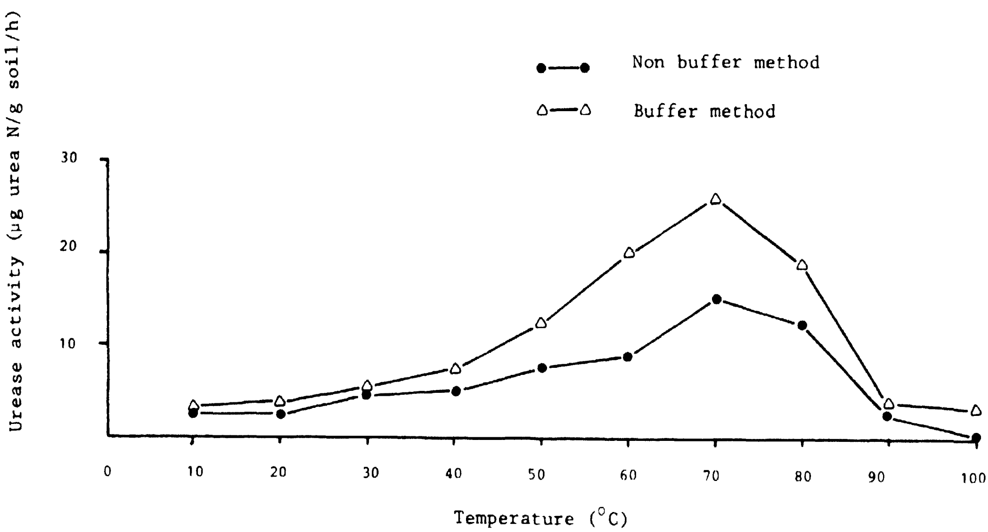

Fig. 1. Effect of temperature on urease activity in Alfisol.

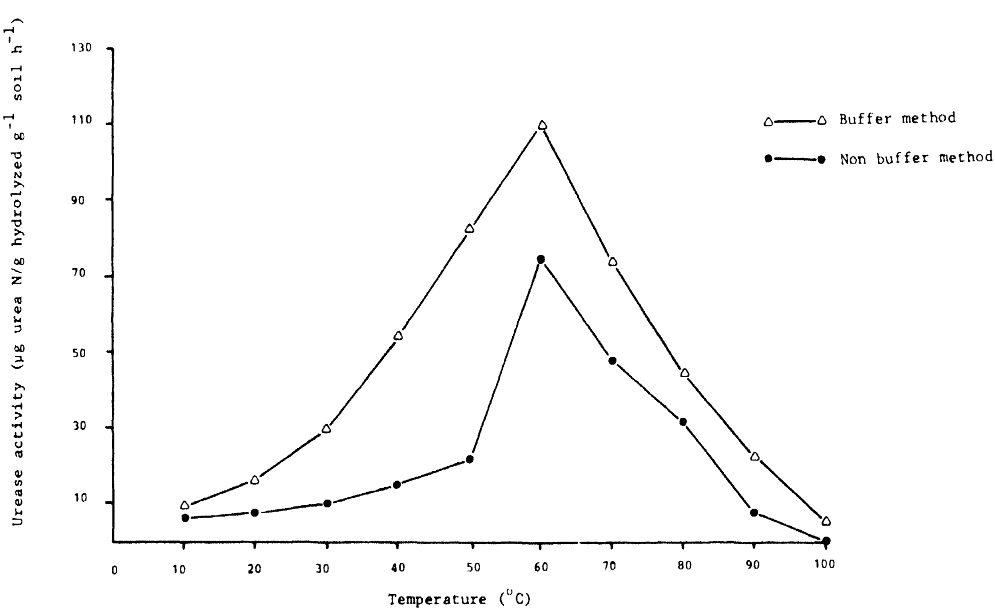

Fig. 2. Effect of temperature on urease activity in Vertisol.

from 10°C to 70°C and then decreased with further increase in temperature to 100°C (Fig. 1). Urease activity of the Vertisol also showed a similar trend with increase in temperature, but the maximal activity occurred at 60°C (Fig. 2). Common to both soils urease activity at 100°C was close to zero when the non-buffer method was used; this was not the case when the buffer method was used for assaying soil urease activity. The results are in agreement with those reported by Zantua and Bremner[20] who found that urease activity in Iowa soils increased with increase in temperature from 10° to 70°C and then decreased sharply with further increase in temperature to 80°C. These authors used the non-buffer method for assaying urease activity. This study provides evidence to show that temperature response to soil urease activity in the soils used is similar whether the buffer or non-buffer method is used for assay.

Results of this study differ from those recently reported for some Indian soils by Dash *et al.*[6], who found that the urease activity increased with increase in temperature from 17° to 47°C and then decreased with increase in temperature from 47° to 57°C. My results along with evidence presented by Zantua and Bremner[20] indicate that urease activity in soil has a much higher optimum of 60°–70°C and suggest that soil urease is protected from inactivation by some mechanism even at high temperatures. The role of clay and organic matter is usually implicated in the stability and protection of soil urease from degradation[3, 10, 14, 19]. It is difficult to explain why the results obtained by Dash *et al.*[6] are divergent. However, the content of organic matter and clay in the soils used by them were similar to those of Alfisol used in the present study. The difference also cannot be explained on the basis of different methods used for urease assay, because the present results have shown that the temperature optima for the soils was nearly the same by both the buffer and non-buffer methods.

Effect of moisture on urease activity

Urease activity could not be detected in air-dried Vertisol and Alfisol soil samples collected from the field during the summer months of April and May, 1981. The moisture content of the Vertisol (6.0% w/w) and Alfisol (0.9%) in the air-dried state was far below −15 bar pressure. In these assays, urea (dried) was applied in solid form without affecting the moisture regime of the field-dried soil samples.

Urease activity in both soils increased with increase in moisture level from the air-dried state to field capacity (0.1 bar for Alfisol and 0.33 bar for Vertisol) and then remained constant with further increase in moisture content (Fig. 3). Previous reports on the effects of moisture on soil urease activity have shown both increases and decreases in urease with increase in water level[2]. For example, with Trinidad soils it was found that urease activity initially increased in moisture content up to 50% WHC, and then decreased with further increase in moisture level[5]; on the other hand, a recent study with Iowa soils, showed that urease activity was not significantly influenced by the water level[20]. Results of the present study agree with those obtained by Dalal[5] to the extent that urease activity increased with increase in moisture content of soils to about field capacity; however, the results differ in that they

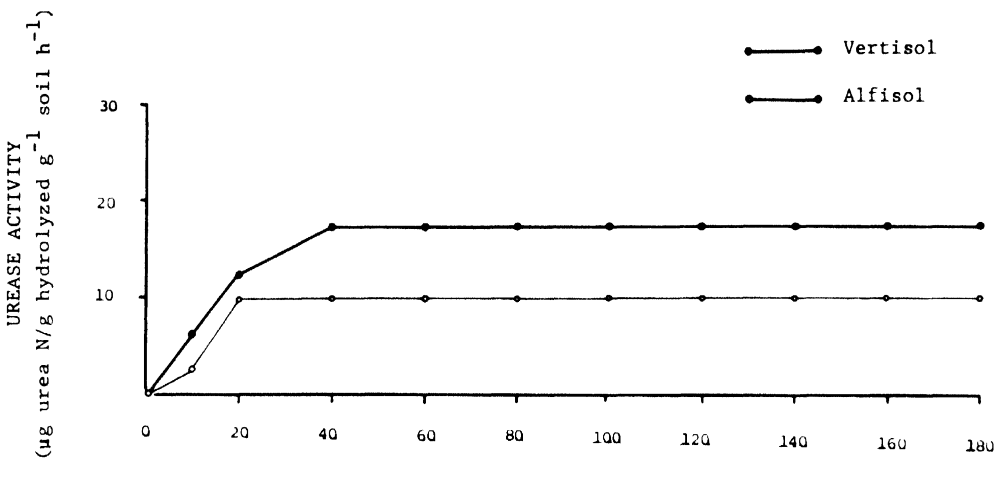

Fig. 3. Effect of moisture on soil urease activity.

showed that an increase in moisture content beyond field capacity did not affect urease activity while Dalal found that urease activity decreased to varying degrees in this range. In the present study, the effect of soil moisture content on urease activity was examined over a much wider range of initial moisture levels than those used in earlier studies[5, 20].

Skujins and McLaren[13, 15] showed that air-dried soil samples equilibrated at 80–100% relative humidity (RH) showed significant levels of urease activity. My results showed that urea hydrolysis, though quite slow, took place in air-dried soil samples when they were incubated in a humid (approximately 100% RH) environment (Table 2).

The rate of urea hydrolysis in air-dried soils in humid environment was slow. For example, when soil samples (5 g) were incubated at 50% WHC moisture content at 37°C (urea N applied at the rate of 1000 μg urea N g^{-1} soil), more than 95% of urea N was hydrolyzed in 2 days while only 2.7 to 3.6% of urea was hydrolyzed during the same period in air-dried soil samples incubated in a humid environment.

A finding from this study with practical significance is the fact that there is little urease activity in both Versitol and Alfisol in the naturally air-dried state prevalent in the summer months of April and May. This implies that urea applied to those soils in air-dried state will not be hydrolyzed until the onset of monsoon (in early June of the year) and there may be little loss until arrival of the monsoon. The relative humidity in the summer months of April and May before the onset of rains is very low; it usually ranges from 20 to 40%[7] and this may not be sufficient to initiate urea hydrolysis in soils having very low moisture content. Improved management practices for Vertisols recommend seeding of crops in dry soil just before the onset of rains[8]. Application

Table 2. Urea hydrolysis in air-dried soil samples as influenced by relative humidity (RH) of the incubation environment*

Soil	Initial soil moisture (%, W/W)	Incubation environment	Urea hydrolysed (μg/g) after days 1	2	3	5	7
Alfisol	0.9	Dry	0	0	0	0	0
		Humid**	0	36	44	129	164
Vertisol	6.0	Dry	0	0	0	0	0
		Humid**	0	27	44	84	119

* Soil samples (5 g) were mixed with urea powder at a rate of 1000 μg urea N g^{-1} soil and incubated at 37°C.

** Humid environment was created by placing inside the incubation vessel a small beaker containing water. RH in the dry and humid environment was approximately 20–30% and 100% respectively.

of urea at seeding, even to the soil surface, may not result in loss of N, because it appears that hydrolysis to ammonium will not occur (due to lack of soil moisture) until the arrival of the opening rains. Supporting field data are needed to verify this because it has relevance to fertilizer N management practices in the seasonally dry, semi-arid tropical regions.

Acknowledgement I thank Mrs. Jayashree Shanker for excellent technical assistance.

References

1 Bremner J M 1965 Total nitrogen. *In* Methods of Soil Analysis, Ed. C A Black, Agronomy 9, pp 1149–1178. Am. Soc. Agron; Madison, Winconsin.

2 Bremner J M and Mulvaney R L 1978 Urease activity in soils. *In* Soil Enzymes. Ed. R G Burns, pp 149–196. Academic Press, London.

3 Conrad J P 1940 The nature of the catalyst causing the hydrolysis of urea in soils. Soil Sci. 50, 119–134.

4 Day P R 1965 Hydrometer method of particle size analysis. *In* Methods of Soil Analysis. Ed. C A Black. Agronomy 9, pp 562–566. Am. Soc. Agron, Madison, Wisconsin.

5 Dalal R C 1975 Urease activity in some Trinidad soils. Soil Biol. Biochem. 7, 5–8.

6 Dash M C, Mishra P C, Mohanty R K and Bhatt N 1981 Effects of specific conductance and temperature on urease activity in some Indian soils. Soil Biol. Biochem. 13, 73–74.

7 ICRISAT 1978 International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Annual Report 1977–1978, Hyderabad, India, 295 p.

8 Kanwar J S, Kampen J and Virmani S M 1982 Management of Vertisols for maximising crop production – ICRISAT experience. *In* Trans. 12th Intern. Congr. Soil Sci., Symposia papers II, 94–118. New Delhi, India, 8–16 February 1982.

9 Mulvaney R L and Bremner J M 1979 A modified diacetyl monoxime method for colorimetric determination of urea in soil extracts. Commun. Soil Sci. Plant Anal. 10, 1163–1170.

10 Pettit N M, Smith A R J Freedman R B and Burns R G 1976 Soil urease: activity, stability and kinetic properties. Soil Biol. Biochem. 8, 479–484.

11 Sahrawat K L 1980 Control of urea hydrolysis and nitrification by chemicals – Prospects and problems. Plant and Soil 57, 335–352.

12 Sahrawat K L and Ponnamperuma F N 1978 Measurement of exchangeable NH_4^+ in tropical rice soils. Soil Sci. Soc. Am. J. 42, 282–283.
13 Skujins J J and McLaren A D 1967 Enzyme reaction rates at limited water activities. Science 158, 1569–1570.
14 Skujins J J and McLaren A D 1968 Persistence of enzymatic activities in stored and geologically preserved soils. Enzymologia 34, 213–235.
15 Skujins J J and McLaren A D 1969 Assay of urease activity using 14C-urea in stored, geologically preserved and in irradiated soils. Soil Biol. Biochem. 1, 88–89.
16 Tabatabai M A and Bremner J M 1972 Assay of urease activity in soils. Soil Biol. Biochem. 4, 479–487.
17 Walkley A and Black I A 1934 An examination of the Degtjareff method for determining soil organic matter and proposed modification of the chromic acid titration method. Soil Sci. 37, 29–38.
18 Zantua M I and Bremner J M 1975 Comparison of methods of assaying urease activity in soils. Soil Biol. Biochem. 7, 291–295.
19 Zantua M I and Bremner J M 1977 Stability of urease in soils. Soil Biol. Biochem. 9, 135–140.
20 Zantua M I and Bremner J M 1977 Unpublished data (cited by Bremner J M and Mulvaney R L 1978).