

## Turnover of $^{15}\text{N}$ ammonium sulfate with dicyandiamide under aerobic and anaerobic soil conditions

K. Vilsmeier<sup>†</sup>

*Institute of Plant Nutrition, Technical University of Munich, Weihenstephan, FRG*

Received 21 December 1990; accepted in revised form 15 April 1991

**Key words:** Nitrification, nitrification inhibitor,  $^{15}\text{N}$ -labelled N-fertilizer, non extractable N

### Abstract

The influence of the nitrification inhibitor dicyandiamide (DCD) on the turnover of  $^{15}\text{N}$ -labelled ammonium sulfate (AS) was investigated in two soils under aerobic and waterlogged conditions. Nitrification of ammonium sulfate was markedly inhibited by addition of DCD in both soils. Up to 45% of the supplied N was transformed into a non-extractable N form, which only slowly released nitrogen over 147 or 264 days. This immobilization was higher in the presence of DCD than without DCD. In all aerobic experiments, the recovery was  $100\% \pm \text{max. } 2.4\%$ , indicating that no gaseous losses of N occurred.

If aerobic preincubation of 28 or 42 days was followed by water-logging with  $\text{H}_2\text{O}$  or a solution of glucose, considerable N losses occurred only in presence of the carbohydrate. DCD retarded nitrification and thus reduced losses by denitrification from 61 to 15%.

DCD application resulted in an increased immobilization of labelled N into the non-exchangeable soil N fraction. This amounted to more than 50% of the applied N, compared to 39% without DCD.

### Introduction

With rising  $\text{NO}_3$  concentrations in ground and drinking water as well as for economic reasons the efficient use of nitrogen fertilizers is of increasing importance. Application of nitrification inhibitors offers one possibility to reduce losses of N by leaching and denitrification which can amount to 30% [8]. While the water soluble dicyandiamide (DCD) is completely decomposed to ammonium in soils the question of residues and practical applicability of nitrapyrine, the most widely known nitrification inhibitor, is still

problematic [3, 7, 9, 12]. The following investigations were aimed at evaluating turnover, distribution and balance of  $^{15}\text{N}$  labelled ammonium sulfate after addition of dicyandiamide under aerobic and anaerobic conditions. Such knowledge may further help in avoiding or minimizing unproductive and ecologically undesirable N losses.

### Materials and methods

**Fertilizers:** ammonium sulfate ( $^{15}\text{N}$  labelled), 15.7%  $^{15}\text{N}$  dicyandiamide (unlabelled, always 10% of the ammonium sulfate-N)

**Soils:** arable soils (Table 1)

<sup>†</sup> The late Dr. Klaus Vilsmeier, a very dedicated and talented young scientist, died before he was able to finish completely the revised version of this article. We will always keep him in our minds and kindly remember his kind personality as well as his sense of humour and justice. Prof. Dr. Heiner Goldbach on behalf of all members of the department.

Table 1. Properties of the studied soils

	Soil type	pH <sub>(CaCl<sub>2</sub>)</sub>	Total N %	Total C %	Fixed NH <sub>4</sub> mg N/100 g	Clay %	Silt %
Weihenstephan I	brown earth from	6.5	0.15	1.43	2	20	66
Weihenstephan II	loess	6.8	0.11	1.07	27	25	59

#### Experimental conditions:

100 g air-dry soil received 20 mg N as ammonium sulfate with or without 2 mg N as DCD and was incubated in 11 polyethylene flasks with screw caps, giving a soil depth of 3 cm. The N rate was equivalent to 100 kg N/ha. In the aerobic incubation trials the samples were adjusted to 60% of total water holding capacity (twc). Anaerobic conditions were provided by water-logging with up to 180% of twc after an aerobic preincubation over 28 or 42 days. The incubation temperature was kept constant at  $14^{\circ} \pm 1^{\circ}\text{C}$  for periods up to 264 days (corresponding to an average soil temperature during the vegetation period). Under aerobic conditions gas exchange was assured by weekly aeration of the incubated flasks. Eventual water losses were corrected for by adding distilled water.

Soluble N was extracted with 0.05 M K<sub>2</sub>SO<sub>4</sub> or 2 M KCl (because of the higher molarity), the exchangeable NH<sub>4</sub>-N was distilled by addition of NaOH, and for the determination of the exchangeable NO<sub>3</sub>-N the distillation was repeated after the addition of Devarda alloy (Modified after 4).

The immobilized-N fraction was obtained by digesting freeze-dried and ground soil after K<sub>2</sub>SO<sub>4</sub> extraction acc. to Kjeldahl. Fixed NH<sub>4</sub>-N was liberated by boiling with KOBr and digestion with conc. H<sub>2</sub>SO<sub>4</sub> [6]. For <sup>15</sup>N-analysis the distillate were titrated with 0.005 M H<sub>2</sub>SO<sub>4</sub>, evaporated; NH<sub>4</sub> was reduced with LiOBr under high vacuum, sealed in gas ionization tubes and measured by emission spectroscopy with a <sup>15</sup>N analyzer (Jasco, Japan) [15].

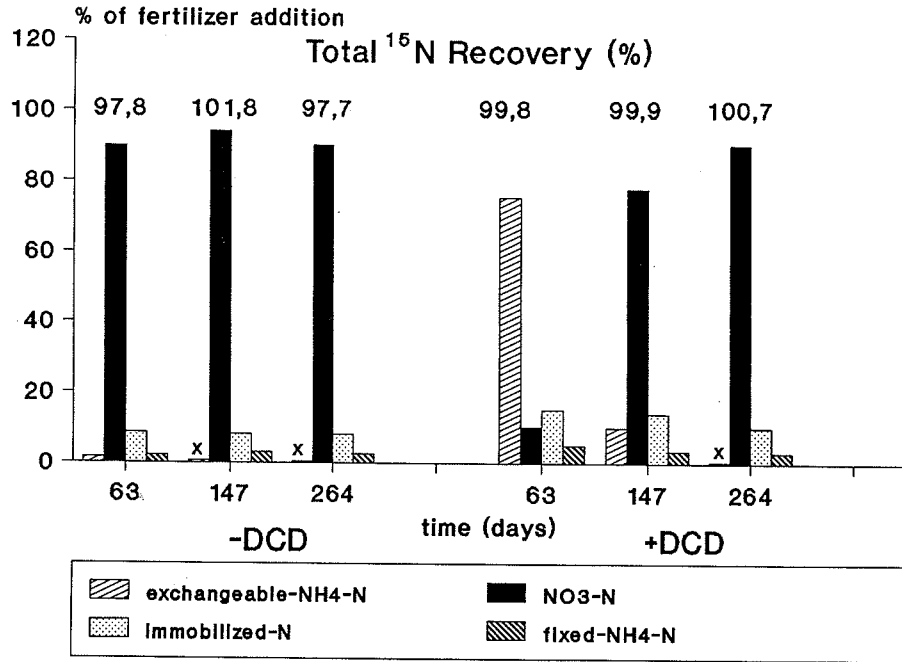
#### Results

Solvent data on the aerobic incubation are summarized in Figures 1 and 2. Within 28 days, ammonium sulfate without DCD was nitrified

completely in soil Weihenstephan I (W I), but only by 30% in soil Weihenstephan II (W II); nitrification in the latter case was completed after 63 days. Addition of 20 ppm DCD-N (concentration in soil) clearly inhibited nitrification in soil (W I) for 63 days, in soil (W II) even for more than 147 days. In all samples 8–15% (W I) or 25–45% (W II) of the supplied nitrogen could not be extracted by 0.05 M K<sub>2</sub>SO<sub>4</sub> nor by 2 M KCl (= immobilized-N). The values of this immobilized N-fraction (determined by Kjeldahl-digestion of the soil) were up to 10% higher after 28 and 63 days than at the end; in other words, initial immobilization was followed by a release of N after 147, and even more after 264 days. Part of this bound N was fixed ammonium, in soil (W I) 2–5%, in soil (W II) 20–36% because of its high NH<sub>4</sub> fixing capacity. Immobilized N as well as fixed ammonium were higher in the presence of dicyandiamide.

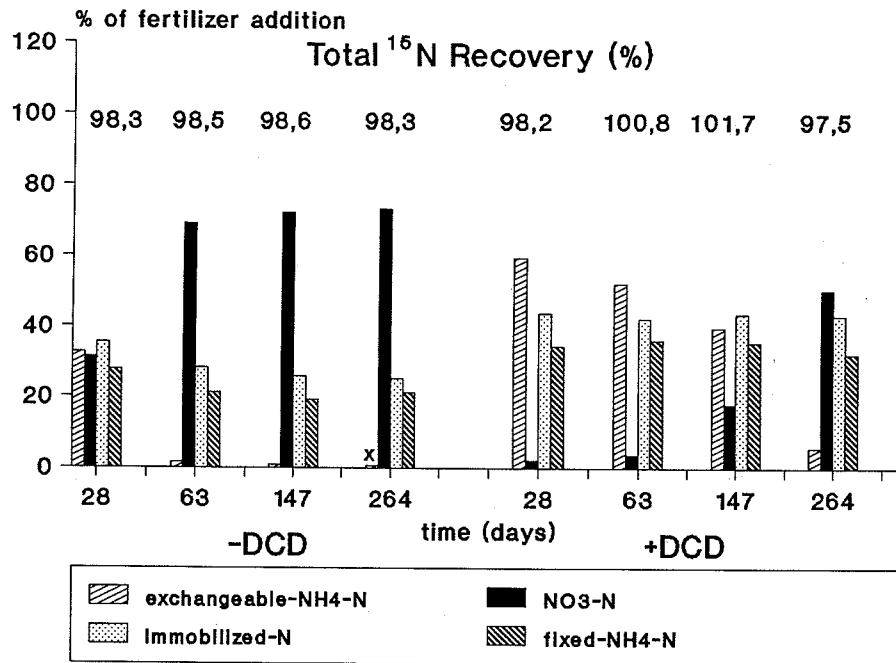
These experiments on aerobic turnover of ammonium sulfate without and with DCD had been designed in a way that losses of N could not occur. Recovery rates were in the range of  $100 \pm 2.4\%$ , with the deviation fully attributable to the analytical error.

Anaerobic incubation data are given in Figure 3 and Figure 4. In these experiments the efficiency of DCD to lower gaseous losses of N by reducing nitrification was investigated. For this purpose, the fertilized soil samples were incubated for 28 or 42 days aerobically and then water-logged. Without DCD, ammonium sulfate was rapidly nitrified (Fig. 3). One week after water-logging no N-losses could be measured by the <sup>15</sup>N balance. After 5 weeks of water-logged conditions (63 days), denitrification losses were low, amounting to 9% of the added nitrogen. By adding DCD, inhibition of nitrification occurred as described above. The oxygen concentrations in these treatments might have been too high and the supply of decomposable carbohydrates



x = traces

Fig. 1. Effect of dicyandiamide on the fate of <sup>15</sup>N ammonium sulfate (soil W I).



x = traces

Fig. 2. Effect of dicyandiamide on the fate of <sup>15</sup>N ammonium sulfate (soil W II).

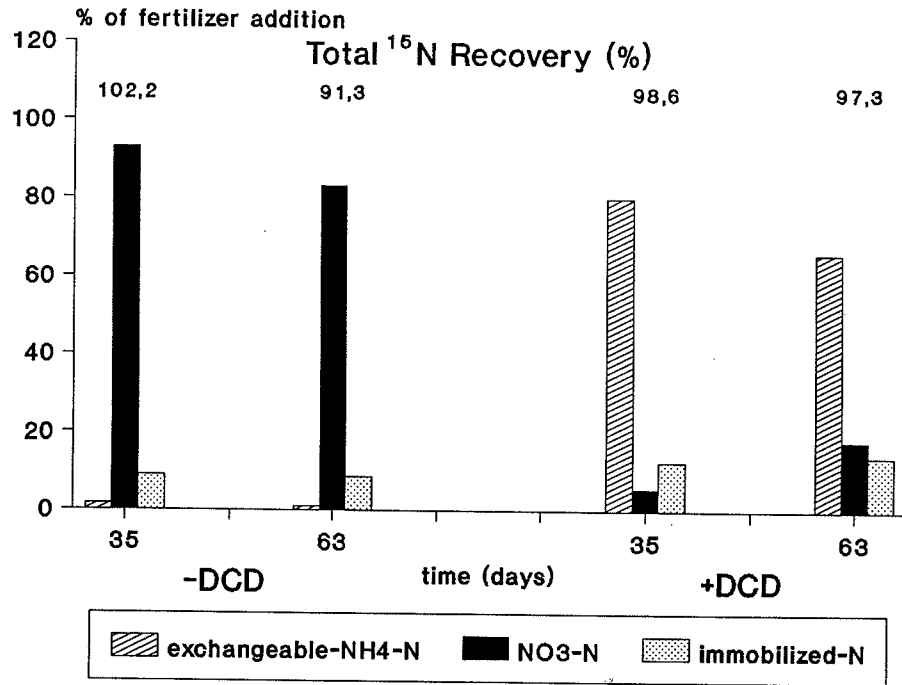
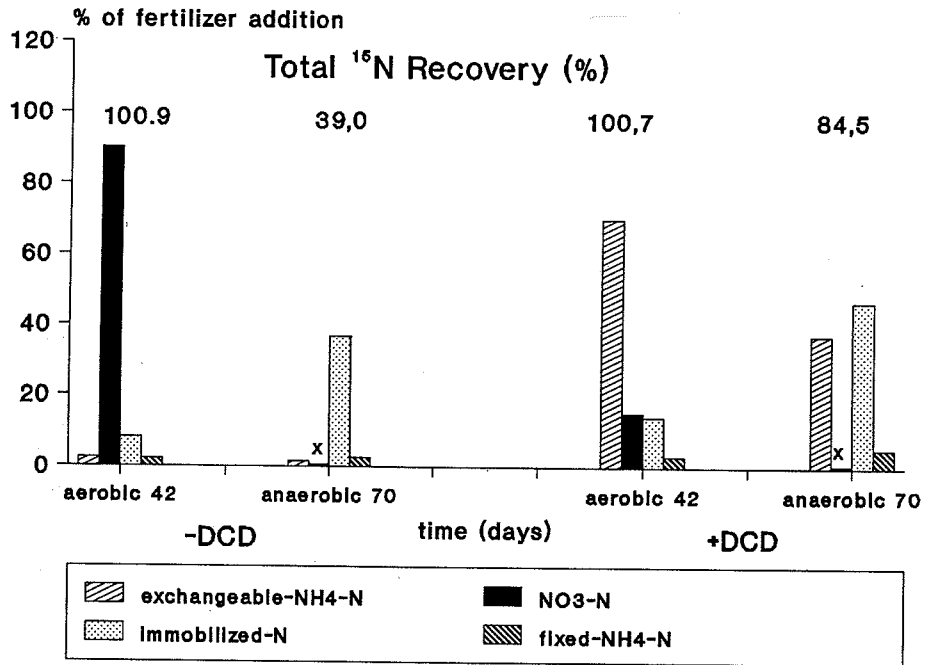


Fig. 3. Effect of dicyandiamide on the fate of <sup>15</sup>N ammonium sulfate under anaerobic conditions (soil W I).



x traces

Fig. 4. Effect of dicyandiamide on the fate of <sup>15</sup>N ammonium sulfate under anaerobic conditions (soil W II).

too low to cause significant denitrification. To provoke such losses, the experiment was repeated, but with the addition of a solution of glucose (200 mg C/pot) upon water-logging. During aerobic preincubation without DCD, no differences occurred compared to the preliminary experiment. The subsequent water-logging with a solution of glucose resulted in high denitrification losses after 28 days: only 39% of the supplied AS-N was recovered, found almost completely in the immobilized N fraction (Fig. 4). By adding DCD, after 42 days as much as 71% of the supplied N were in the form of  $\text{NH}_4\text{-N}$  as a result of inhibited nitrification, and thus could not be denitrified. At the end of the anaerobic phase with DCD a total of 85% was recovered, 47% as non-extractable N.

### Discussion

Probably due to a low activity of nitrifying bacteria, formation of  $\text{NO}_3$  in soil (W II) was markedly slower than in soil (W I). Dicyandiamide addition inhibited nitrification as has been observed previously [1, 2, 10, 11, 13, 15].

Under aerobic conditions ( $\pm$ DCD) after 147 or 264 days only 56% up to 91% of the supplied N was extractable as  $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$ . The slower the nitrification of  $\text{NH}_4\text{-N}$  and the longer N was maintained as  $\text{NH}_4$  in this system, the more fertilizer nitrogen was transferred to a form neither extractable with 0.05 M  $\text{K}_2\text{SO}_4$  nor with 2 M KCl (8–45% of N supply). This is due to fixed ammonium (max. 36% of the  $^{15}\text{N}$ ) and/or a reaction with the soil organic matter. At the end of the incubation period, however, a certain release of nitrogen from these fractions occurred.

The initial experiments on reducing losses by denitrification showed only minor effects, even without DCD; obviously an easily decomposable carbon source was lacking for providing energy to the bacteria and for further oxygen consumption. Adding glucose and water-logging after aerobic preincubation created favourable conditions for denitrification. This resulted in high losses, and considerable incorporation of  $\text{NO}_3\text{-N}$  in the immobilized N fraction, too. The latter is

likely to be due to the glucose addition as well [5].

After addition of the nitrification inhibitor DCD oxidation of ammonium was markedly slower; this meant higher amounts of  $\text{NH}_4$  at the beginning of water-logging which, in turn, allowed more of the N to be immobilized into organic N.

Therefore, considerably lower N losses (15%), and on the other hand an increased incorporation of nitrogen (appr. 50% of added amount) into 'non-extractable' forms were the consequence.

In practical agriculture the addition of DCD to  $\text{NH}_4$ -releasing fertilizers (e.g. ammonia sulfate, urea) may reduce denitrification losses especially in poorly drained or flooded (rice) soils. The danger of losses by denitrification is especially high under conditions, where high amounts of easily decomposable organic matter are present, which in part, may be overcome by adding nitrification inhibitors such as DCD.

### Acknowledgements

We thank Dr. C. Amann for the English translation.

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