# THE MECHANISM OF THE DECOMPOSITION OF CYANAMIDE IN THE SOIL.

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(With Five text-figures.)

A PREVIOUS investigation by the writer[1] showed that cyanamide readily breaks down, yielding ammonia in normal clay and sandy soils. The evidence, however, threw no light upon the cause or nature of this change. This question was accordingly reserved for a later investigation. The concensus of the available evidence indicated that the production of ammonia from cyanamide in the soil is due to direct bacterial action. This view was held by Immendorff[2] and Kappen[3], who concluded that in poor soils of low bacterial activities cyanamide is not converted into ammonia but is chemically transformed into dicyanodiamide. Löhnis[4] at first accounted in a similar way for the formation of ammonia from cyanamide in the soil. He[5] assumed later, however, that cyanamide is normally decomposed by soil colloids into urea or possibly some other substances, and the latter are then converted into ammonia by the soil organisms. He adduced no direct evidence of the production of urea from cyanamide in the soil.

Ulpiani[6] regarded the formation of ammonia as primarily due to a purely chemical, not a bacterial, change. He had formerly considered that cyanamide changed into dicyanodiamide in the soil and its value as a fertiliser depended on this change, an opinion also held by Perotti[7].

Ulpiani's later work, however, led him to the view that cyanamide breaks down by a purely chemical change to urea which then is converted into ammonia. The formation of urea was attributed to the soil colloids. This work was done in culture solutions of various concentrations and at various temperatures.

Our experiments were made in soil under natural conditions, using amounts of cyanamide comparable with those used in practise. Our results agree with those of Ulpiani.

Our experiments have consistently failed to show any appreciable amount of ammonia resulting from the decomposition of cyanamide in sterile soils (heated to 120° C. or 135° C.). The addition of the urease of soya-bean, however, produced considerable amounts of ammonia in these soils. This pointed to the presence of urea, which was later confirmed by the extraction of the soils by alcohol and identification of urea in the extract by the urea-nitrate test. Further experiments moreover demonstrated that urea actually remains stable in soils heated to 120° C. The addition of cyanamide to sterile soils thus leads to an accumulation of urea, which persists as such in consequence of the suppression of the necessary urea decomposing organisms.

In a similar way the addition of cyanamide to soils heated to 100° C. does not lead to an immediate production of ammonia. It forthwith yields urea, however, which then decomposes into ammonia after the recovery of the appropriate organisms.

The evidence shows, on the other hand, a rapid and progressive production of ammonia arising from the decomposition of cyanamide in unheated normal clay and sandy soils. Careful examination of these soils, however, revealed in the initial periods the presence of appreciable amounts of urea. This indicates that urea produced by the decomposition of cyanamide also accumulated to some extent under normal conditions.

The cumulative evidence thus leads to the conclusion that cyanamide in the soil is normally converted by a purely chemical process into urea and this change is not dependent on the activity of micro-organisms. The urea is then broken down to ammonia by a change which, as the curves indicate, is produced by soil organisms. Cyanamide appears to behave in this way in both clay and sandy soils, but the decomposition seems to be more rapid in the former than in the latter. The experiments have conclusively shown that cyanamide does not decompose into urea in ordinary impure quartz sand; whatever the decomposing agent may be it is not present in pure sand.

Cyanamide does not appear to break down in the manner above indicated in peat and fen soils; in these it gives rise to a relatively small production of urea under normal conditions.

The investigation has not revealed the exact nature of the decomposing agent in the soil. It is interesting to note, however, that a sample of Thanet sand taken from a boring through the London Clay near Chelmsford was found even after ignition to be active in decomposing cyanamide into urea. This particular sand [8] has been shown to contain a constituent resembling a zeolite in being reactive and possessing the

property of softening hard water by the substitution of sodium salts and possibly potassium for those of calcium and magnesium. In following up this clue it was found that the addition of a definite zeolite prehnite to ordinary inert sand produces a mixture capable of converting cyanamide into urea.

## EXPERIMENTAL.

The cyanamide was used in the form of fresh nitrolim in which the calcium cyanamide had undergone practically no change. The bulk of the soil to be used was first thoroughly mixed together and its moisture content raised, where necessary, to 12 to 15 per cent. according to its water capacity. The soil was next passed through a 3 mm. sieve, weighed out into lots of 200 grams which were then transferred to wide-mouthed bottles of 10 oz. capacity, a fresh bottle being taken for each determination at the end of the various periods. The application of the cyanamide to the soils was made in the following manner.

In the case of unheated soils the weighed quantity of cyanamide was sprinkled on to the 200 grams of soil previously spread out on a sheet of paper; the whole was then mixed and replaced in the bottles, which were plugged with cotton-wool.

In the case of the heated soils this procedure might have led to reinfection, and therefore the bottles containing soil, etc. were first plugged with cotton-wool and then heated for the proper period at the requisite temperature. After cooling, the cyanamide was carefully and rapidly introduced into the bottles from small glass tubes, in which the cyanamide itself had been heated for the same time at the same temperature in the air-oven. Tests showed that heating under these conditions produced no chemical change in the cyanamide. After careful replugging to avoid reinfection the bottles were vigorously shaken for some considerable time to ensure an adequate mixing of the cyanamide with the soils.

In no case did heating cause a loss of more than 2 per cent. moisture in the soils. All bottles were then kept in a dark cellar at the ordinary laboratory temperature.

In order to inhibit the nitrification of ammonia produced from cyanamide, a small amount of dicyanodiamide, equivalent to thirty parts N per million dry soil, was added with the cyanamide. A previous investigation showed conclusively that dicyanodiamide does not give rise to ammonia or affect appreciably the production of ammonia from cyanamide.

### THE STAGES IN THE PRODUCTION OF AMMONIA FROM CYANAMIDE IN SOILS.

Experiments were made to determine the relative rates of ammonia production from cyanamide in untreated soils and the same soils after heating for one half hour in the autoclave at 120° C. Both the heavy Rothamsted and the light Woburn soils were used. The results (plotted in Fig. 1 and given in Table I) show a rapid and progressive production of ammonia from cyanamide in the unsterilised soils, but they afford little evidence of ammonia resulting from the decomposition of cyanamide in the sterilised soils.
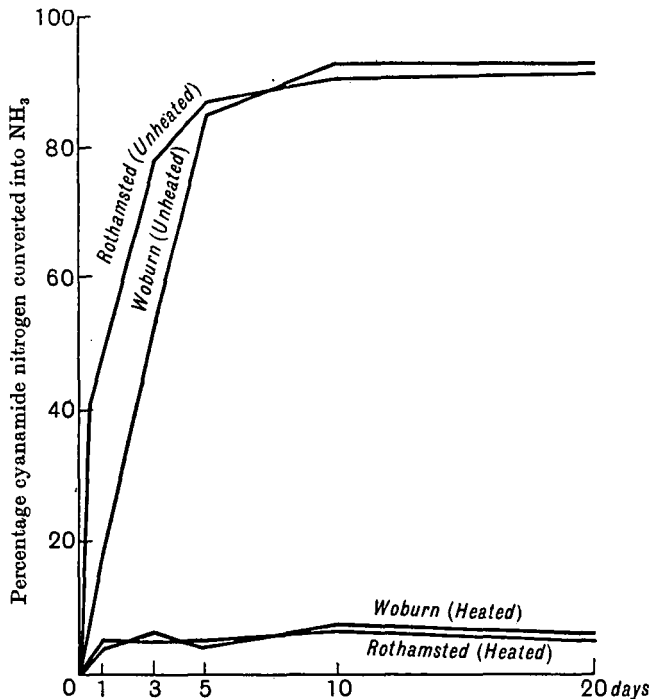


Fig. 1. Showing relative rates of ammonia production from cyanamide in unheated soils and soils heated to 120° C.

Although the sterilised soils showed little trace of ammonia production from cyanamide, there was the possibility that the decomposition had stopped at some intermediate stage. On chemical grounds the most probable stage seemed to be the formation of urea. Examination for this substance was therefore made in soil which had been stored for 20 days or more, the urease of the soya-bean being used as the testing

agent. Takeuchi(9) and also Armstrong and Horton(10) have shown this enzyme is quite specific in its action, decomposing urea only, and nothing else so far as is known. The soils were accordingly treated with

### Table I.

*Ammonia produced from Cyanamide in Soils.*

N present as $NH_3$ per million dry soil

| Treatment | Rothamsted Control at start 4·0 | | | | | Woburn Control at start 2·6 | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | After 1 day | After 3 days | After 5 days | After 10 days | After 20 days | After 1 day | After 3 days | After 5 days | After 10 days | After 20 days |
| Control | 4·0 | 3·4 | 4·0 | 6·7 | 6·7 | 2·6 | 1·9 | 2·6 | 5·2 | 5·2 |
| +Cyanamide | 44·5 | 82·3 | 90·3 | 97·1 | 97·1 | 22·0 | 55·5 | 87·8 | 98·2 | 100 |
| Heated soil (120° C.) | 9·3 | 19·9 | 23·9 | 25·2 | 25·2 | 7·7 | 7·7 | 19·1 | 17·9 | 21·7 |
| +Cyanamide | 14·6 | 14·6 | 29·2 | 31·9 | 30·5 | 12·8 | 14·0 | 23·0 | 25·5 | 26·8 |

Cyanamide N added = 100 parts per million dry soil.

well-powdered soya-bean and incubated at 35–40° C., the optimum temperature for the enzyme. The ammonia was then determined in the usual way. The results were as follows:

### Table II.

*Ammonia produced from Cyanamide in Sterilised Soils after addition of Soya-bean.*

| Treatment | N present as $NH_3$ per million dry soil | |
|---|---|---|
| | Rothamsted | Woburn |
| Heated soil ... ... ... ... | 23·6 | 21·7 |
| +soya-bean ... ... ... ... | 26·2 | 26·0 |
| +cyanamide +soya-bean* ... ... | 104·3 | 58·4 |
| Heated soil +cyanamide (no soya-bean) | 27·0 | 18·1 |

Cyanamide N added = 100 parts per million dry soil.

\* The mixture of heated soil and cyanamide had previously been stored for 20 days.

The addition of soya-bean thus resulted in a considerable production of ammonia in the sterilised soils treated with cyanamide. Further tests showed that the soya-bean does not produce any appreciable amount of ammonia direct from cyanamide.

The evidence thus indicated that the origin of the ammonia produced by soya-bean in the sterile soils was an accumulation of urea derived from cyanamide.

Table III.

*Non-formation of Ammonia by Action of Soya-bean on Cyanamide.*

| Treatment | N present as NH$_3$ per million dry soil Rothamsted |
|---|---|
| Sterilised soil + soya-bean ... ... ... ... ... | 26·2 |
| ditto     ditto    + cyanamide (freshly added) ... | 34·7 |

Cyanamide N added = 100 parts per million dry soil.

Tests were subsequently made to ascertain whether urea does actually remain stable in similarly heated soils. Another set of soils was therefore made up containing urea in place of cyanamide; the quantity of added nitrogen, however, being the same. In these soils urea and ammonia were both determined after an interval of 20 days; only a small decomposition of urea into ammonia took place under these conditions.
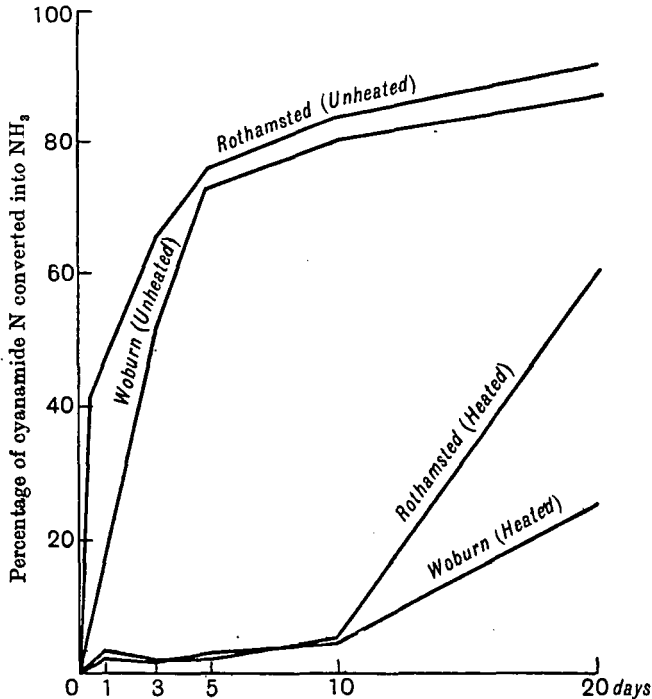


Fig. 2. Showing relative rates of ammonia production from cyanamide in unheated soils and soils heated to 100° C.

In order to confirm the production of urea from cyanamide in the sterilised soils, the latter were extracted with 96 per cent. alcohol and the solution evaporated to dryness *in vacuo*. The crystals thus obtained

were shown to include urea by means of the urea nitrate test. The application of the same test to a similar extract from the control soils gave negative results.

### Table IV.

*Stability of Urea in Sterilised Soils.*

N present as NH₃ per million dry soil after 20 days.

| Heated soil | ... | ... | ... | ... | ... | ... | ... | 14·2 |
| ditto | +urea | ... | ... | ... | ... | ... | ... | 20·7 |
| ditto | +soya-bean | ... | ... | ... | ... | ... | ... | 22·0 |
| ditto | ditto | +urea ... | ... | ... | ... | ... | ... | 115·0 |

Urea N added = 100 parts per million dry soil.

The above evidence indicates that cyanamide in the soil decomposes into urea which is then converted into ammonia by the soil organisms. The suppression of the organisms in sterilised soils, therefore, leads to a persisting accumulation of urea from cyanamide.

These conclusions were further confirmed by the behaviour of cyanamide in partially sterilised soils or those heated for half an hour at 100° C. In this case the production of ammonia was practically suspended for several days, after which it proceeded. The results are plotted in Fig. 2 and given in Table V.

### Table V.

*Ammonia produced from Cyanamide in partially Sterilised Soils.*

N present as NH₃ per million dry soil

| Treatment | Rothamsted at start 5·2 | | | | | Woburn at start 2·5 | | | | |
| | After 1 day | After 2 days | After 5 days | After 10 days | After 20 days | After 1 day | After 2 days | After 5 days | After 10 days | After 20 days |
|---|---|---|---|---|---|---|---|---|---|---|
| Control | 5·2 | 5·2 | 5·2 | 3·9 | 3·9 | 2·5 | 2·7 | 2·5 | 2·6 | 2·5 |
| +cyanamide | 47·0 | 69·9 | 80·5 | 87·1 | 95·6 | 20·2 | 52·2 | 75·7 | 78·1 | 88·3 |
| Heated soil (100° C.) | 9·0 | 9·0 | 9·0 | 9·0 | 35·6 | 6·0 | 6·0 | 6·4 | 6·4 | 12·3 |
| +cyanamide | 11·6 | 10·3 | 10·3 | 14·2 | 95·6 | 8·9 | 7·5 | 8·7 | 11·2 | 37·7 |

Cyanamide N added = 100 parts per million dry soil.

### Table VI.

*Formation of Urea from Cyanamide in partially Sterilised Soils.*

| Treatment | N present as NH₃ per million dry soil | |
| | Rothamsted | Woburn |
|---|---|---|
| Heated soil (100° C.) + cyanamide | 11·6 | 7·8 |
| Ditto* + soya-bean | 64·9 | 34·2 |

Cyanamide N added = 100 parts per million dry soil.

* By "ditto" is to be understood in each case the mixture used in the line above.
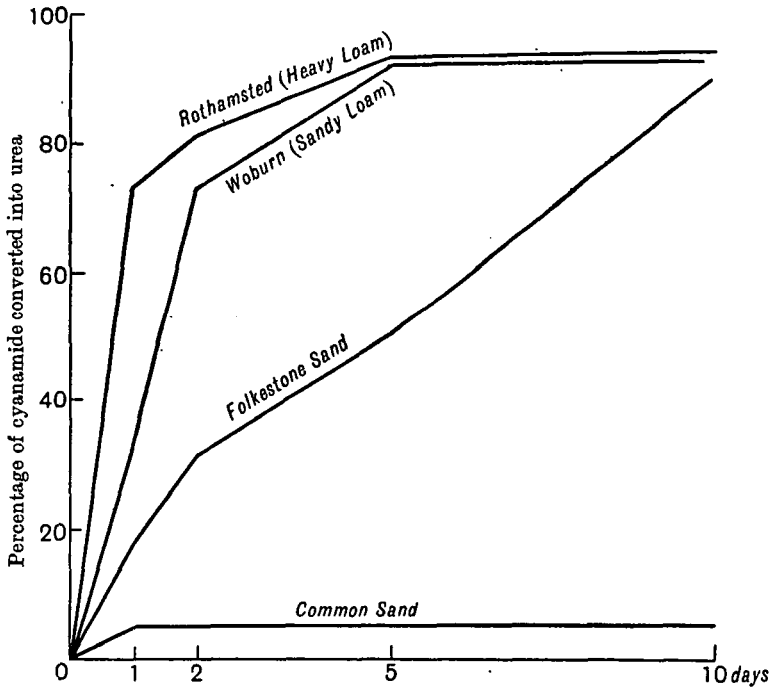
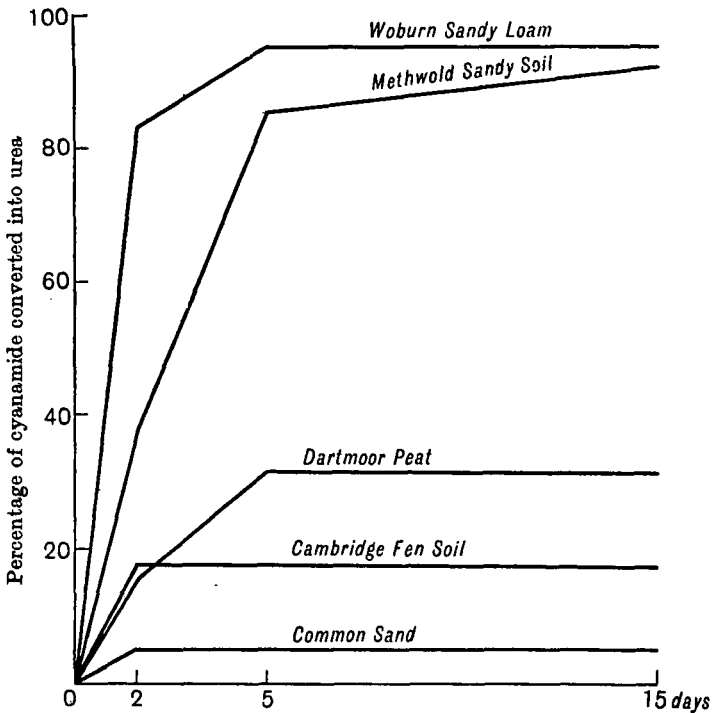Fig. 3. Showing rates of conversion of cyanamide into urea.



Fig. 4. Showing rates of conversion of cyanamide into urea.

In the first 10 days the addition of cyanamide resulted in but a small production of ammonia in the heated soils. Treatment of these soils with soya-bean, however, produced a considerable amount of ammonia at the end of the 4th day.

The above results thus show that cyanamide in the soil heated to 100° C. first decomposes to urea which subsequently changes into ammonia. The results further indicated that this subsequent change is biological because it is almost wholly suspended during the first 10 days, after which it becomes much more rapid.

Table VII.

*Production of Urea and Ammonia from Cyanamide in Unheated Soils.*

N present as NH$_3$ per million dry soil.

| Treatment | Rothamsted, at start 2·6 | | | | Woburn, at start 1·8 | | | | Folkestone, at start 13·6 | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | After 1 day | After 2 days | After 5 days | After 10 days | After 1 day | After 2 days | After 5 days | After 10 days | After 1 day | After 2 days | After 5 days | After 10 days |
| Control + soya-bean | 19·7 | 19·7 | 19·7 | 19·0 | 15·2 | 15·7 | 15·2 | 15·2 | 26·0 | 26·0 | 26·5 | 32·2 |
| Ditto + cyanamide | 93·2 | 100·4 | 113·0 | 112·8 | 46·9 | 89·0 | 108·0 | 108·0 | 43·4 | 57·0 | 74·4 | 122·6 |
| Difference = rates of urea production | 73·5 | 80·7 | 93·3 | 93·8 | 31·7 | 73·3 | 92·8 | 92·8 | 17·4 | 31·0 | 47·9 | 90·4 |
| Control | 3·9 | 3·9 | 3·0 | 2·6 | 1·8 | 1·8 | 2·0 | 1·8 | 13·6 | 13·6 | 13·6 | 18·6 |
| + cyanamide | 57·8 | 76·2 | 97·2 | 97·2 | 29·2 | 58·4 | 83·8 | 90·2 | 14·8 | 17·3 | 52·1 | 106·6 |
| Difference = rates of ammonia production | 53·9 | 72·3 | 94·2 | 94·6 | 27·4 | 56·6 | 81·8 | 88·4 | 1·2 | 3·7 | 38·5 | 88·0 |

| Treatment | Woburn Sandy loam Control at start 1·9 | | | Methwold Sandy soil Control at start 12·4 | | | Leighton Buzzard Ordinary sand Control at start 1·3 | | |
|---|---|---|---|---|---|---|---|---|---|
| | After 2 days | After 5 days | After 15 days | After 2 days | After 5 days | After 15 days | After 2 days | After 5 days | After 15 days |
| Control + soya-bean | 12·7 | 12·7 | 15·2 | 26·0 | 26·0 | 26·0 | 14·1 | 14·1 | 14·5 |
| Ditto + cyanamide | 95·3 | 107·9 | 108·0 | 63·2 | 110·3 | 117·8 | 19·2 | 19·2 | 19·7 |
| Control ... ... | 1·9 | 1·7 | 1·8 | 12·4 | 12·4 | 12·9 | 1·3 | 1·3 | 1·3 |
| + cyanamide ... | 57·5 | 83·8 | 90·2 | 50·7 | 95·4 | 107·9 | 5·0 | 5·1 | 5·1 |

Cyanamide N added = 100 parts per million dry matter.

The final step was to ascertain whether the change proceeded in the same manner in ordinary unheated soils. The figures in Table VII show that this is the case. The soils used were from Rothamsted (Clay-with-flints), Woburn (Lower greensand), a poor heath sand uncultivated, from Blackheath, Surrey (Folkestone beds of the Lower greensand

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formation), the very light sandy soil from Methwold, Norfolk; and sand from a sand pit at Leighton Buzzard, Beds (Lower greensand). The results are given in Table VII and plotted in Figs. 3 and 4.

The cumulative evidence thus leads to the conclusion that cyanamide in these soils is decomposed by a purely chemical process into urea, and the latter is subsequently converted into ammonia by the soil organisms. There is at first an accumulation of urea even in the unheated soils and a very marked accumulation in the heated soils. Cyanamide appears to undergo the same change in both clay and sandy soils, even in a soil containing no more than 0·9 per cent. clay, as in the Folkestone sand soils. In the sand from the sand pit, however, there is no formation of either urea or ammonia from cyanamide. The decomposition is the more rapid in the soils containing the larger proportion of clay. The greater persistence of the urea in the poor uncultivated Folkestone sand, as compared with the cultivated Rothamsted and Woburn soils, is both interesting and significant.

### Soils in which the Formation of Urea from Cyanamide does not take place.

In Table VII it is shown that the formation of urea from cyanamide does not take place in sand from a sand pit. Table VIII shows that the action likewise does not occur in peat and fen soils.

### Table VIII.

*Non-formation of Urea from Cyanamide in Peat and Fen Soils.*

N present as $NH_3$ per million dry soil.

| Treatment | Dartmoor Peat Control at start 15·6 | | | Cambridge Fen Control at start 4·7 | | |
|---|---|---|---|---|---|---|
| | After 2 days | After 5 days | After 15 days | After 2 days | After 5 days | After 15 days |
| Control + soya-bean | 21·8 | 21·8 | 46·9 | 9·5 | 9·5 | 9·0 |
| Ditto + cyanamide | 57·5 | 53·2 | 72·0 | 27·1 | 27·0 | 27·5 |

Cyanamide N added = 100 parts per million dry soil.

### The Mechanism of the Production of Urea from Cyanamide in Soil.

Further investigation showed that the decomposition of cyanamide into urea still proceeded in a soil previously heated as high as 135° C. for half an hour in the autoclave. It is evident therefore that the action is not brought about by living organisms or by an enzyme decomposable —as most enzymes are—at 135° C.

## Table IX.

*Production of Urea from Cyanamide in soil heated to 135° C.*

N present as $NH_3$ per million dry soil. After 10 days.

| | |
|---|---|
| Heated soil (135° C.) ... ... ... | 13·8 |
| + soya-bean ... ... ... ... | 27·0 |
| Ditto + cyanamide ... ... ... | 101·3 |
| Heated soil + cyanamide ... ... | 18·8 |

Cyanamide N added = 100 parts per million dry soil.

The change was found to occur in a sample of Thanet sand taken from a boring through the London Clay at Broomfield in Essex.

## Table X.

*Production of Urea from Cyanamide in Thanet Sand.*

### (a) Unignited material.

N present as $NH_3$ per million dry sand. Control at start 19·8.

| Treatment | After 1 day | After 2 days | After 5 days |
|---|---|---|---|
| Control + soya-bean | 31·0 | 30·1 | 31·0 |
| Ditto + cyanamide | 97·9 | 106·6 | 133·8 |
| Control ... ... | 19·5 | 19·8 | 19·9 |
| + cyanamide ... | 19·8 | 19·8 | 37·2 |

Cyanamide N added = 100 parts per million dry sand.

### (b) Ignited Thanet Sand.

N present as $NH_3$ per million dry sand.

| Treatment | After 1 day | After 3 days |
|---|---|---|
| Ignited Thanet sand + soya-bean ... | 45·8 | 45·0 |
| Ditto + cyanamide ... ... ... | 84·5 | 108·1 |

Cyanamide N added = 100 parts per million dry sand.

The Thanet sand was then ignited at a dull red heat but it still remained active in decomposing cyanamide into urea (Table X and Fig. 5). It appears therefore that the decomposing agent, in this case at any rate, is inorganic in its nature. This Thanet sand possessed the property of softening hard water by the substitution of sodium and possibly potassium salts for calcium. Presumably therefore it contained a constituent resembling a zeolite in being reactive. A definite zeolite, viz. prehnite, was therefore tested and when added to inert quartz sand it gave a mixture capable of converting cyanamide into urea. Table XI

shows the extent to which action had proceeded after 5 days. Further investigation promises very interesting results; this is being carried out in the Rothamsted laboratories by Mr A. G. Pollard.
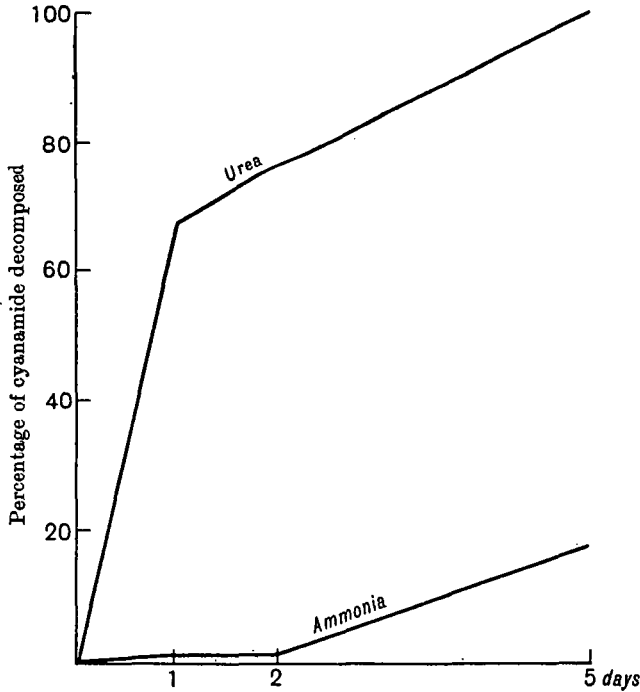


Fig. 5. Showing relative rates of urea and ammonia formation
from cyanamide in Thanet sand.

## Table XI.

*Production of Urea from Cyanamide in a Mixture of Inert Sand
and a Zeolite (Prehnite).*

| Treatment | N present as $NH_3$ per million dry matter After 5 days |
|---|---|
| Inert sand + prehnite ... ... ... ... ... | 1·2 |
| Inert sand + prehnite + soya-bean ... ... ... | 11·1 |
| Inert sand + prehnite + cyanamide + soya-bean ... | 39·6 |
| Inert sand + prehnite + cyanamide ... ... ... | 4·9 |

    Finely ground prehnite added = 9 per cent. of mixture.

    Cyanamide nitrogen added = 100 parts per million dry matter.

## THE DETERMINATION OF AMMONIA IN THE SOIL.

The ammonia in the soil was determined by the aeration method used in the previous investigation and shown to produce no appreciable formation of ammonia by hydrolysis from cyanamide. A stronger current of air, however, was used whereby it was found possible to recover, with satisfactorily concordant duplicates, approximately 95 per cent. of any added ammonia in the soil. Average results of repeated tests were:

N added = 100 parts per million dry soil.

| | N recovered per million dry soil | |
| --- | --- | --- |
| | Rothamsted | Woburn |
| Soil alone ... ... ... ... | 3·2 | 1·3 |
| Soil and ammonium sulphate ... ... | 98·1 | 99·1 |

## THE DETERMINATION OF UREA IN THE SOIL.

The amounts of urea in the soil were determined by the use of the urease of the soya-bean. Prior to estimating the ammonia by the aeration method the soil was treated with well-powdered soya-bean, using 1 gram of the latter to 25 grams of the fresh soil. After thorough mixing the whole was incubated at 35° C.–40° C. for one hour, and then aerated after cooling for the determination of the ammonia. Repeated tests showed that approximately 90 per cent. of the urea added to a sterile soil or sand could be recovered as ammonia. Aeration caused no appreciable decomposition of the urea. As a small amount of ammonia was evolved by the soya-bean, a control experiment with soya-bean and soil was carried out in each case. Typical results were:

| Treatment | N recovered as $NH_3$ per million dry soil | |
| --- | --- | --- |
| | Sterile soil | Sand |
| Control ... ... ... ... ... ... | 14·2 | 1·3 |
| Ditto  + soya-bean ... ... ... ... ... | 22·0 | 14·1 |
| Ditto   ditto   + cyanamide (100 parts nitrogen) | 115·5 | 102·0 |
| Control + urea (100 parts nitrogen) (no soya-bean) ... | 20·7 | 3·8 |

The added urea contained 100 parts of nitrogen as also did the added cyanamide.

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