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Effect of Nitrogenous Forms on Immobilization and Release of Nitrogen in Soil

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The effect of nitrogenous forms on immobilization and release of nitrogen following addition of glucose and mineral nitrogen to soils was studied. The amount of nitrogen immobilized increased with increasing C/N ratio independent of the forms of nitrogen added, but the nitrogen immobilized to a maximum at a given C/N ratio was a little larger in the form of ammonia than in the form of nitrate. The amount of carbon which must be added to the soil in order to immobilize one unit of nitrogen into organic form was larger in the latter than in the former. The percentage of released nitrogen to the maximum-immobilized nitrogen at a given C/N ratio was higher in the latter than in the former.

Several investigators (Allison 1927, Jansson 1955, Winsor et *al.* 1956, Allison *et al.* 1962, Harada *et al.* 1968, Kai *et al.* 1969, Ahmad *et al.* 1969) have reported that incorporation of energy rich materials together with mineral nitrogen to soil causes immediate nitrogen immobilization and after some time turns into an increased mineralization. It has been observed that the immobilization and mineralization processes in soil are governed by many factors among which the kind of carbonaceous materials incorporated, their C/N ratio, form of nitrogen added, soil reaction, soil temperature, soil moisture, etc. play an important role.

According to some authors (Richards *et al.* 1935, Jansson 1955, Winsor *et al.* 1956, Jansson 1958, Allison *et al.* 1962, Broadbent et *al.* 1962), the microorganisms concerned in the immobilization of nitrogen showed a marked preference for the form of ammonia rather to the form of nitrate, but this view was contradicted by others (Walunjkar, Bartholomew and Woltz, 1959), and Norman (1931) pointed out that ammonium and nitrate nitrogen were equally available to the microorganisms as a source of nitrogen. The variation in the findings of workers may possibly be due to their experimental plans and procedures and often give different nitrogen immobilization values.

With a view to add more exact informations and to contribute to a better understanding of the processes of immobilization and release of nitrogen in soil, the present experiment was designed to observe the role played by the different nitrogenous forms in the processes following addition of glucose and mineral nitrogen to soil.

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MATERIALS AND METHODS

Soil samples

Soil samples used in this study were collected from the ploughed layers of four soils, two of which have been receiving the three elements of fertilizer and compost and the other two receiving no fertilizer and compost since about last forty years. They are located at the lowland and highland fields of the Kyushu University farm. The moist soils were sieved through a 2 mm sieve. The denotation and some of the chemical and physical properties of the soil samples are given in Table 1.

Table 1.	Description	of soils	used.
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Soils	Denotation	Texture	Total C (%)	Total N (%)	pH (KCl)
Lowland, fertilized	LF	LiC	$2.65 \\ 1.86 \\ 1.38 \\ 0.89$	0.23	4.70
not fertilized	LnoF	LiC		0.15	4.50
Highland, fertilized	HF	CL		; 0.11	5.15
not fertilized	HnoF	CL		; 0.05	4.19

Incubation and determination

Moist soil samples corresponding to 20 g dry soil were weighed into 50 ml Erlenmeyer flasks, and treated with 4 mg N as $(NH_4)_2SO_4$ or $NaNO_3$ and various levels of C as glucose. The level of mineral nitrogen added was constant throughout the experiments, only carbon level was varied to give different C/N ratios. Carbon added to 20 g dry soil as glucose was 0, 8, 16, 32 and 64 mg giving C/N ratio 0, 2, 4, 8 and 16 in all the soils. An adequate amount of $CaCO_3$ was added to bring the pH of the soils to 6.2, and then the soil water content was adjusted to 60 per cent of maximum water holding capacity with additional water. The basal dressing with 2 mg P_2O_5 , 2 mg K_2O and 0.5 mg MgO per 20 g dry soil was also made. All the flasks were covered with polyethylene film and incubated at 30°C for the periods of 3 days and 1, 2, 4, 8 and 12 weeks, respectively, water being added at intervals to compensate for water loss during incubation as determined by weighing the flasks. Mineral nitrogen produced during various time intervals of incubation was extracted with N KC1 (pH 7.0) and determined by Conway's microdiffusion method (Bremner et al. 1955, Kai 1961). Determination of mineral nitrogen was made on duplicate flasks of soil. The amount of nitrogen immobilized at a given C/N ratio has been calculated as the difference in content of mineral nitrogen between the soil treated with mineral nitrogen and the soil treated with mineral nitrogen and glucose. The amount of nitrogen released has been calculated as the difference in content of mineral nitrogen between the soil at a time of maximum immobilization of nitrogen and the soil at a given time interval. The term "check soil" refers to soil receiving no treatment other than the maintenance of moisture content.

RESULTS AND DISCUSSION

The results obtained by the experiments on the transformation of nitrogen

								In	cubati	on per	iod (d	ays or	week	s)							
C/N		0			3*	[1			2		_	4	- 7		8			12	
	NH ₄ - N	NO ₃ - N	Sum	NH4- N	NO ₃ - N	Sum	NH4- N	NO ₃ - N	Sum	NH4- N	NO ₃ - N	Sum	$_{\rm NH_{4}^{-}}$	NO ₃ - N	Sum	NH4- N	NO ₃ - N	Sum	NH4- N	NO ₃ - N	Sum
										(N	$(H_4)_2S$	04									
0	18.94,	0.50	19.44;	19.81	0.93	20.74	20.23	1.18	21,41	21.59	0.57	22.16	24.39	2.57	26 . 96	1.99	26.65	2 8. 64	0.39	30.78	31.17
2	18.94	0. 50'	19. 441	15. 421	2.00	17.42	17.46	1.97	19.43	19.23	1.16	20.39	22.29	3. 57	25.86	3.19	2 3. 01	26.20	0.39	29.26	29.65
4	18. 94	0.50	19.44	13.70	0.89	14.59	15.19	1.12	16,31	1 7. 51	0.83	18.34	19.74	3.20	22.94	0.39	24.69	25.08	0.36	27.55	27.91
8	18.94	0.50	19. 44	8. 78	0.04	8.82	11.39	0.53	11.92	12.77	0.24	13.01	17.13	1.60	1 8. 73	0.94	20.58	21. 30	0.69	23.23	23.92
16	18. 94	0. 50	19. 44	3.10	0.07	3.17	6.06	0.00	6.06	6.64	0.18	6.82	11.07	0.92	11.99	0.18	16.15	16.33	0.44	21.52	21.96
Check	0.83	0.50	1.33	0.25	3.52	3.77	4.20	1.82	6.02	5.15	1.63	6.78	8. 20	1.82	1 0. 02	0.76	13.03	13.79	0.79	14.81	15.60
								i			NaNO	·									
0	0.83	18.49	19.32	2.38	17.91	20.29	3.77	17.64	21.41	5.23	19.45	24.68	7.69	20.87	2 8. 56	0.25	31.10	31.35	0.22	32.19	32.41
2	0.83	18.49	19.32	1.89	15.09	16.98	3.90	13.56	17.46	5.28	17.73	23.01	8. 04	19. 40	27.44	0.51	29.25	29.76	0.76	30.77	31.53
4	0.83	1 8. 49	19.32	2.34	13.32	15.66	4.41	12.76	17.17	6.03	14.07	2 0. 10	11.21	14.79	26.00	0.29	2 8. 53	28.82	0.94	29.83	30.77
8	0.83	18.49	19.32	3. 03	8.08	11.11	5.84	8.42	14.26	7.84	8. 38	16.22	10.16	10.29	20.45	3.19	22.36	25.55	0.73	26.71	27.44
16	0.83	18.49	19.32	3.79	0.58	4.37	7.08	0.00	7.08	9.29	1.14	10.43	11.19	3.6 5	14.84	4.32	16.47	20.79	0.83	24.03	24.86
Check	0.83	0.50	1.33	0.25	3. 52	3.77	4.20	1.82	6.02	5.15	1.63	6.78	8. 20	1.82	10.02	0.76	1 3. 03	13.79	0.79	14.81	15.60

 $\label{eq:table 2. Transformation of mineral nitrogen at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and $NaNO_3$ to LF soil.}$

(mg N/100 g dry soil)

* Days.

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following addition of glucose and mineral nitrogen as $(NH_4)_2SO_4$ or NaNO₃ to LF-, LnoF-, HF- and HnoF-soils are shown in Tables 2, 6, 10 and 14, respectively.



Fig. 1. Amounts of mineral nitrogen at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and $NaNO_3$ to LF soil.

In LF soil, as shown in Table 2 and Fig. 1, it was clear that immobilization proceeded rapidly, reached its maximum on 3-day incubation and was followed by a release of nitrogen irrespective of the C/N ratios and the forms of nitrogen. These trends were similar to the results obtained by Allison et *al.* (1960) and Harada et *al.* (1968). The amounts of immobilized nitrogen, as shown in Table 3, varied with the varing C/N ratio. The wider the C/N ratio, the larger the amount of immobilized nitrogen in both the forms of nitrogen, and the estimated amount of immobilized nitrogen was found larger in the form of ammonia given as (NH₄)₂SO₄ than in the form of nitrate given as NaNO₃, which agreed with

Table 3. Estimated amounts of immobilized nitrogen at various time intervals, following addition of either glucose and $(\rm NH_4)_2\rm SO_4$ or glucose and $\rm NaNO_3$ to LF soil.

(mg N/100 g dry soil)

C () I		Inc	ubation period	(days or v	veeks)		
C/N	3*	1	2	4		8		12
			(NH4) ₂ SO ₄				
2 4	3.32 6.15	1.98 5.10	1.77 3.82	4.02 1.10		2.44 3.56	ſ	1.52 3.26
8 16	11.92 17.57	15.35 9.49	15.34 9.15	14.97 8.23		12.31 7.34	1	7.25 9.21
	1	1	Na	NO ₃				
2 4 8 16	3.31 4.63 9.18 15.92	3.95 4.24 7.15 14.33	$ \begin{array}{r} 1.67 \\ 4.58 \\ 8.46 \\ 14.25 \end{array} $	$1.12 \\ 2.56 \\ 8.11 \\ 13.72$		1.59 2.53 5.80 10.56	I	0.88 1.64 4.97 7.55

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* Days.

the findings of Richards and Shrikhande (1935), Winsor and Pollard (1956), Jansson (1958) and Broadbent and Tyler (1962), but was contrary to the finding of Walunjkar et **al**. (1959). The rate of release of nitrogen was found faster at an early period than at a late period in both the forms of nitrogen as shown in Tables 4 and 5.

 $\label{eq:table_$

(mg N/100 g dry soil)

C ()]			Incu	ubation period (weeks)		
C/N		1	1	4	8		12
	1	1. 34 1. 03 2. 43 2. 22	1. 55 2. 33 2.77 2.23	(NH ₄) ₂ SO ₄ 2.22 2.13 3.69 2.60	0.88 2.59 4.58 5.26	1	1.80 2.89 4.67 8.36
				NaNO ₃		1	
		0,64 2.03 1.59	0.051.64 0.72 1.67	2.07 2.19 1.07 2.20	210 1.72 3.38 5.36		243 299 4.21 8.37

Table 5. Estimated rate of release of immobilized nitrogen per half a week at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and $NaNO_3$ to LF soil.

(mg N/100 g dry soil/half week)

C (N		Incu	ubation period (w	eeks)	
C/N		2	'1	8	12
-			(NH ₄) ₂ SO ₄		
4 8 16	$1.17 \\ 0.92 \\ 2.13 \\ 1.94$	0. 11 0. 64 0.17 0.01	0.17 0.05 0.23 0.09	-0.17 0.06 0.11 0.33	0.12 0.04 0.01 0.39
			NaNO ₃		*
	-0.56 0.34 1.78 1.39	$ \begin{array}{r} 1.14 \\ -0.17 \\ -0.66 \\ 0.04 \end{array} $	0. 14 0. 50 0. 09 0. 13	- 0.06 0.00 0.29 0.40	0.09 0.26 0.10 0.38

In LnoF soil, as shown in Table 6 and Fig. 2, the trend of immobilization and release of nitrogen which was found in LnoF soil was the same in nature that was found in LF soil. Here also we found that immobilization reached its maximum on 3-day incubation and was followed by a release of nitrogen. And as shown in Table 7, the wider the C/N ratio, the larger the amount of immobilized nitrogen. Here also, the superiority of $(NH_4)_2SO_4$ to $NaNO_3$ on the immobilization of nitrogen was maintained. The rate of release of nitrogen was

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		Table and	e 6. T (NH ₄) ₃	Cransfe SO4 or	ormatic	on of 1 se and	ninera NaNC	l nitre	gen at noF se	t vario oil.	us tin	inte	rvals,	follow	ing ad	dition	of eith	er gluc	cose		
					I											(mg N	1 /100 g	dry s	oil)		
			.					In.	cubatic	n peri	iod (di	ays or	weeks								
C/N		0	 		3*		-	1			2			4			8			12	
	$^{+4-}$ N	$NO_{3^{-}}$	Sum	NH4- N	N03- N	Sum	NH4- N	NO3- NO3-	Sum	NH4- N	N03- N	Sum	NH₄- N	N03- N	Sum	-'HN N	NO3- N	Sum	NH₄- N	NO ₃ - N	Sum
									-	N.	H4) ₂ S(-				-	-	
0	18.22	0.04	18.26	18.80	1.08	19.88	18. 43	1.78	20.21	20.30	0.86	21.16	23.21	1.96	25.17	8.61	17.18	25.79	0.54	26.99	27.53
2	18.22	0.04	18.26	14.21	0.00	14.21	15.52	0.52	16.04	17.49	0.90	18.39	20.82	3.66	24.48	7.13	16.60	23.73	0.65	25.32	25.97
4	18.22	0.04	18.26	12.44	0.00	12.44	12.96	1.22	14.18	15.52	0.40	15.92	18.11	1.53	19.64	5.68	15.99	21.67	0.96	22.68	23.64
8	18.22	0.04	18.26	7.80	0.00	7.80	9.09	0.46	9.55	10.37	0.75	11.12	15.12	0.51	15.63	2.32	15.19	17.51	0.47	18.77	19.24
16	18.22	0.04	18.26	2.31	0.00	2.31	3.02	0.52	3.54	4.81	0.00	4.81	9.59	0.00	9.59	0.51	11.21	11.72	0.39	13.43	13, 82
Check	0.09	0.04	0.13	0.09	2.26	2.35	0.08	3.46	3.54	3.98	0.14	4.12	5.46	1.09	6.55	0.51	8.89	9.40	0.25	10.42	10.67
				/		-	-			4	VaNO ₃		j		1		-	<u>.</u>	-		
0	60.0	18,14	18.23	0.12	19.85	19.97	1.92	18.90	20.82	3.11	18.92	22.03	6.00	19.54	25.54	0.11	26.95	27.06	0.54	28.07	28.61
Ċ1	0.09	18.14	18.23	0.10	17.48	17.58	1.83	16.62	18.47	3.26	16.91	20.17	6.75	17.48	24.23	0.04	26.00	26.04	0.33	27.23	27.56
4	0.09	18.14	18.23	0.11	15.05	15.16	2.33	13.48	15.81	3.67	14.37	18.04	8.03	13.61	21.64	0.11	24.08	24.19	0.36	25.36	25.72
ò	0.09	18.14	18.23	1.90	6.96	8.86	6.73	4.23	10.96	5. 77	8.19	13.96	9.39	9.09	18.48	9.44	11.25	20.69	1.99	20.25	22.24
16	0.09	18.14	18.23	2.46	0.14	2.60	5.01	0.60	5.61	7.32	0.25	7.57	12.37	0.80	13.17	11.72	5.17	16.89	6.66	12.11	18.77
Check	0.09	0.04	0.13	0.09	2.26	2.35	0.08	3.46	3.54	3.98	0.14	4.12	5.46	1.09	6.55	0.51	8.89	9.40	0.25	10.42	10.67

* Days:

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Fig. 2. Amounts of mineral nitrogen at various time intervals, following addition of either glucose and $(\rm NH_4)_2SO_4$ or glucose and $\rm NaNO_3$ to LnoF soil.

Table 7. Estimated amounts of immobilized nitrogen at various time intervals, following addition of either glucose and $(\rm NH_4)_2SO_4$ or glucose and $\rm NaNO_3$ to LnoF soil. (mg N/100 g dry soil)

				Incuba	tion perio	od (days	or	weeks)		
C/N	3*		1		2	4		8	Į	12
					(NI	$(I_4)_2 SO_4$				
$\begin{array}{c}2\\4\\8\\16\end{array}$	4.67 7.44 12.08 17.57	ł	$\begin{array}{r} 4.70 \\ 6.56 \\ 10.76 \\ 16.68 \end{array}$	 	2.51 5.83 9.83 16.21	2 5 9 14	. 48 . 74 . 13 .04		$2.06 \\ 4.82 \\ 8.28 \\ 11.87$	$1.74 \\ 4.49 \\ 8.05 \\ 10.54$
					N	aNO3				
$\frac{2}{4}$	2.39	I	2.35	ļ	1.86	1	. 31		1.02	1.05
$\frac{4}{8}$ 16	11.11 4.81 17.37		9.86 5.01 15.21		3.99 8.07 14.46	3 <u>9</u> 12	0 7.06 .37		2.87 6.37 10.10	2.89 6.37 9.84

* Days.

Table 8. Estimated amounts of released nitrogen at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and $NaNO_3$ to LnoF soil. (mg N/100 g dry soil)

				(ee g urj son)
		Incu	bation- period (v	veeks)	
C/N	3.	2	4	8	12
			$(\mathrm{NH}_4)_2\mathrm{SO}_4$		
2	- 0.03	2.16	2.19	2.61	2.93
	0.88 1:32 b.89	2.25 1.61 1.36	2.95 1.70 3.53	2.62 3.80 5.70	2.95 4.03 7.03
	1	· · · · · · · · · · · · · · · · · · ·	NaNO3		+
$\frac{4}{8}$	0.04 -0.20 1.25	0.53 0.82 3.04	1.08 0.91 4.05	1.37 4.74 1.94	1.34 4.74 1.92
16	2.16	2:91	5:00	7.20	7.53

Table 9. Estimated rate of release of immobilized nitrogen per half a week at various time intervals, following addition of either glucose and $(\rm NH_4)_2SO_4$ or glucose and NaNO3 to LnoF soil.

C / N			In	cubati	ion period	(week	s)		
C / N	1		2	ĺ	4		8		12
					$(\mathrm{NH}_4)_2\mathrm{SO}_4$	Ļ			
2 4	-0.03 0.77		1.09 0.37	i	0.0.0201		0.05 0.12		0.04 0.04
0 16	0.78 1.16		0.47 0.24	I	0.18 0.54		0.11 0.27		0.03 0.17
					$NaNO_3$			<u>-</u>	
$\frac{2}{4}$	-0.18 0.04	}	0.25 0.51		0.14 0.02		0.04 0.13	I	0.00 0.00
16	1.09 1.89	ł	0.89 0.43		0. 0.52 25	ĺ	0.09 0.28		0.00 0.04

(mg N/100 g dry soil/half week)

also faster at an early period than at a late period in both the forms of nitrogen, as given in Tables 8 and 9.

In HF soil, as shown in Table 10 and Fig. 3, it was also observed that immobilization occurred just after addition of mineral nitrogen together with glucose and reached its maximum on 3-day incubation irrespective of the C/N ratio and the forms of nitrogen added. And as shown in Table 11, the wider the C/N ratio, the larger the amount of immobilized nitrogen. The nature of immobilization and release of nitrogen in HF soil was also the same that in LF and LnoF soils. Only the amounts of immobilized and released nitrogen in HF soil were found smaller than those in LF and LnoF soils. Here also, $(NH_4)_2SO_4$ and NaNO₃ maintained their respective position with regard to immobilization and release of nitrogen in the soil. And the rate of release of nitrogen in both the forms of nitrogen was also the same nature as found in LF and LnoF soils, as shown in Tables 12 and 13.

From Table 14 and Fig. 4 in which the results obtained from HnoF soil are shown, it was seen that immobilization and release of nitrogen in HnoF soil which was smallest in total nitrogen among the four soils used, were also the same manner as found in LF, LnoF and HF soils. Here also, maximum immobilization occurred on 3-day incubation, and was followed by a release of nitrogen. Here also, as shown in Table 15, the wider the C/N ratio, the larger the amount of immobilized nitrogen, and $(NH_4)_2SO_4$ was found superior on immobilization to NaNO₃. The rate of release of nitrogen was also faster at an early period of incubation than at a late period in both the forms of nitrogen, as shown in Tables 16 and 17.

In view of the results obtained on immobilization and release of nitrogen following addition of glucose and mineral nitrogen as $(NH_4)_2SO_4$ or $NaNO_3$ to soil, it can be clearly said that the nature of immobilization and release of nitrogen was the same in all the soils used.

Experimental results obtained under the experimental conditions employed

Table 10. Transformation of mineral nitrogen at various time intervals, following addition of either glucose and $(\rm NH_4)_2SO_4$ or glucose and $\rm NaNO_3$ to HF soil.

(mg N/100) g	dry	soil)
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	-							In	cubati	on per	riod (d	ays or	week	.s)				_			
C/N		0	1		3"			1	1		2			4				8		12	
	$\frac{NH_4}{N}$	NO ₃ - N	Sum	NH4- N	$\underset{N}{\overset{NO_{3}-}{N}}$	Sum	NH4- N	NO ₃ - N	Sum	NH ₄ - N	NO ₃ - N	Sum	NH4- N	NO ₃ - N	Sum	NH4- N	NO ₃ - N	Sum	NH₄- N	NO ₃ - N	Sum
										(N	IH₄)₂S	O4									
0	19.84~	0.00	19.84~	15.68~	3.07,	18.75'	11.92	6.84,	18.76	4.63	14.09	18.72	0.54	19.21	19.75	0.00	23.08	, 23.08	0.00'	22.75	22.75
2	19.84~	0.00;	19.84~	12.49	3.28'	15.77	9.32	6.77	36.09,	2.69	13. 46	16.15	0.43	17.37	17.80	0.00	20.88	20.88	0.00	21.49	21.49
4	19.84	0.00	19.84	10.44	2.47	12.91	8.10	5.01	13.11	2.50	11.39	13.89	0.34	15.52	15.86	0.00	18.83,	18.831	0.00	19.26'	19.26
8	19.84	0.00	19.84~	6.79~	1.33	8.12	4.97'	3.69	8.66~	1.16	8.89	10.05	0.16	11.9 0	12.06'	0.00	15.48	' <u>15. 48</u>	0.00	15.75	15.75
16	19.84~	0.00	19.84	0.72	3.59	4.31	3.41	1.94	5.35	0.94	6.82	7.76	0.54	8. 93	9.47	0.00	14.36	14.36	0.00	14.81	14.81
Check	0. 01	0.00	0.01	0.29	0.82	1.11	0.11	1.22	1.33	0.76	0.00	0.76	0.00	3.38	3.38	0.00	5.69	5.69	0.00	6.59	6.59
								_		i	NaNO							<u> </u>			
0	0.01	18.71	18.72	0.00	19.85	19.85	0.90	19.12	20.02	0.00	20.87	20.87	0.04	21.08	21.1 2	0.00	23.36	23.36	0.00	22.89	22.89
2	0.01	18.71	18.72	0.00	17. 1 2	17.12	0.61	17.50	18.11	0.00	18.61	18.61	0.00	19. 04	19.04	0.00	20.99	20.99	0.00	21.92	21.92
4	0.01	18.71	18.72	0.25	14.87	15.12	1.26	14.94	16.20	0.00	17.71	17.71	0.09	18.07	18.16	0.00	20.19	20.19	0.00	20.48	20.48
8	0.01	18.71	18.7 2	3.94	9.56	13. 50	3.58	10.89	14.47	0.00	14.15	14.15	0.14	17. 55	17.69	0.00	20.12	20.12	0.00	19.83	19.83
16	0.01	18.71	18 . 72	5.28	1.71	6.99	5.61	3.07	8.68	0.39	9.87	10.26	0.15	13.55	13.70	0.00	17.11	17.11	0.00	16.90	16.90
Chesk	0.01	0.00	0.01	0.29	0.82	1.11	0.11	1.22	1.33	0.76	0.00	0.76	0.00	3.38	3.38	0.00	5.69	5.69	0.00	6.59	6,59

* Days.



Fig. 3. Amounts of mineral nitrogen at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and $NaNO_3$ to HF soil.

Table 11. Estimated amounts of immobilized nitrogen at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and $NaNO_3$ to HF soil. (mg N/100 g dry soil)

	_]	Incuba	tion peri	iod (days or v	veeks)		
C / N	I3*:		1		2	1	4	l	8		12
					(N	$H_4)_2S$	O4				
$ \frac{2}{4} 8 16 $	2.98 5.84 10.6 3	 	2.67 5 65 13.41		2.57 4.83 8.67 10.96		$ \begin{array}{r} 1.95 \\ 3.89 \\ 7.69 \\ 10.28 \end{array} $	-	2.20 4.25 7.60 8.72	1	1.26 3.49 7.00 7.94
[N	laNO ₃	3				
	27:: 4.73 6. 35 12.86		3.82 1.91 5.55 1.1.34	ł	3.16 2.26 5.72 10.61		2.96 2.08 3.43 7.42	i J	3.17 2.37 3.24 6.25		0.97 2.41 3.06 5.99

* Days.

Table 12. Estimated amounts of released nitrogen at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and $NaNO_3$ to HF soil.

THE INTOOR OF SOLD	(mg	N/100	g drv	soil)
--------------------	-----	-------	-------	-------

				In	cubatio	on period	(week	s)		
C/N	1	1		2	4	i I	8			12
					((NH ₄) ₂ SO	4			
2		0.31.		0.11	T	1.03		0.78]	1.72
$^{4}_{16}$;	0.190.5:: 1.03	1	1.01 1.96 3.48		2.94 1.93 4.16		3.1.5903 5.72		2.35 3.63 6.50
						NaNO3				
$\frac{4}{8}$	ĺ	0.82 0.91 0.80		0.47 0.63 1.57		0.65 2: 1.77 92		0.36 3.11 1.56		1.78 2.32 3.29
16		1.52		2.25	!	5.44	I	6.01		6.87

Table 13. Estimated rate of release of immobilized *nitrogen per* half a week at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and NaNO₃ to HF soil.

				(mg N/100 g ary	son/n	iali week)
		In	cubati	on period	(weeks)		
C/N	1	2		4	8		12
2				$(NH_4)_2SO_4$			
$\overline{4}$	0.27 0.14	I 0.05 0.41		0.0.2413	- 0. 01	I	0.09
8 16	0.46 0.90	0.72 1. 23	I	0.0.1725	$ -0.06 \\ 0.02$	I	0.08 0.06
		I	•	-	0.26		0.08
		1		$NaNO_3$			
$\frac{2}{4}$	0.72	-0.18		0. on	-0.05 -0.04		0.14
8	0.17 0.10	-0.59		0.02100	0.03		0.02
16	1.33	0.37		0.79	0.19		0.03



8



Incubation period (weeks)

Check

12

Fig. 4. Amounts of mineral nitrogen at various time intervals, following addition of either glucose and $\rm (NII_4)_2SO_4$ or glucose and NaNO_3 to HnoF soil.

Table	15.	Estin	nated	d amou	ints o	f	immo	bilized	nitro	ogei	ı at	vario	ous	time	interval	s, fol-
lowing	add	ition	of	either	gluco	se	and	(NH_4)	₂SO₄	or	gluco	ose a	nd	NaNO	3 to Hno	F soil.
													(n	ng N/3	100 g dr	y soil)

			I	ncub	ation period	(days or	weeks)	
C/N	3*		1		2	4		8	12
1					(NH	1)2SO4			
$24 \\ 8 \\ 16$	3.40 6.05 9.36 14.19		$2.81 \\ 5.28 \\ 8.88 \\ 13.24$	ļ	2.65 4.87 8.41 31.17	1.61 3.54 6.92 9.16		1.97 3.93 6.44 7.84	1.G8 3.76 G.80 7.63
1					Nal	NO ₃			
$\frac{2}{4}$	$\begin{array}{c} 2.39\\ 4.75\end{array}$		$\begin{array}{c} 2.34\\ 4.12\end{array}$	ł	1.98 4.69	2. 1 0 3.92		1.76 2.90	1.68 3.26
8 16	12.59 8.38	1 [11.44 7.90		10.53 6.59	6.22 9.92		5.80 8:41	5.52 8.20

* Days.

mg Mineral $N/100~{\rm g}$ dry soil

0

12

Table	e 14.	Transformation	of n	nineral	nitrogen	at	various	time	intervals,	following	addition	of	either	glucose
and	(NH_4)	₂ SO ₄ or glucose	and	NaNO	₃ to HnoF	` so	oil.			_				-

(mg N/100 g dry soil)

								In	cubati	on per	iod (da	ays or	week	s)							
C/N		0			3*			1	4.00.000		2			4			8			12	
	NH ₄ -	NO ₃ - N	Sum	NH ₄ - N	NO ₃ - N	Sum	NH4- N	NO ₃ - N	Sum	NH ₄ - N	NO ₃ - N	Sum	$\frac{\mathrm{NH_{4}}}{\mathrm{N}}$	NO ₃ - N	Sum	NH ₄ - N	NO ₃ - N	Sum	NH ₄ - N	NO ₃ - N	Sum
,										(N	$(\mathrm{H}_4)_2\mathrm{S}$	J₄									
0	18.54	0.00	18. 54	1 8. 01	0.77	18.78	17.15	1.36	18.51	13.23	4.67	17.90	3.13	14.90	18.03	0.00	19.15	19.15	0.00	18.69	18.69
2	18.54	0.00	1 8. 54	15.25	0.13	15.38	15.18	0.52	15.70	13.71	1.54	15.25	10.28	6.14	16.42	0.00	17.18	17.18	0.00	17.01	17.01
4	18. 54	0.00	1 8. 54	12.73	0.00	12 . 73	13.07	0.16	13,23	11.03	2.00	13.03	5.09	9. 40	14.49	0.00	15.22	15.22	0.00	14.93	14.93
8	18. 54	0.00	1 8. 54	8.77	0.65	9.42	8,86	0.77	9. 63	8.08	1.41	9.49	5.30	5.81	11.11	0.00	12.71	12.71	0.00	11.89	11.89
16	18. 54	0.00	1 8. 54	4.17	0.42	4.59	4.94	0.33	5.27	5.96	0.77	6.73	2.29	6.58	8.87	0.00	11.31	11.31	0.00	11.06	11.06
Check	0.02	0.00	0.02	0.00	0.47	0.47	0.00	0.32	0.32	0.00	0.72	0.72	0.07	1.72	1.79	0.00	2.51	2.51	0.00	2.15	2.15
]	NaNO ₃			<u></u>							
0	0.02	18.43	1 8. 45	0.36	18.92	19.28	0.00	19.37	19.37	0.00	18,51	18, 51	0.07	19. 64	19.71	0.00	20.05	20.05	0.00	19.87	19.87
2	0.02	18. 43	18.45	0.32	16.57	16.89	0.93	16. 10	17.03	0.25	16.28	16.53	0.14	17.47	17.61	0.00	18.29	18.29	0.00	1 8. 19	1 8.1 9
4	0.02	18. 43	18,45	0.35	14.17	14.52	1.59	13.66	15.25	0.00	13.82	13.82	0.21	15. 58	15.79	0.00	17.15	17.15	0.00	16.61	16.61
8	0.02	1 8. 43	18.45	1.68	9.22	10.90	2.92	8.55	11.47	1.55	10.37	11.92	0.43	13.06	13.49	0.00	14.25	14.25	0.00	14.35	14.35
16	0.02	18.43	18.45	4.01	2.68	6.69	4.29	3.64	7.93	2.83	5.15	7.98	0.35	9.44	9.79	0.00	11.64	11.64	0.00	11.67	11.67
Check	0.02	0.00	0.02	0.00	0.47	0.47	0.00	0.32	0.32	0.00	0.72	0.72	0.07	1.72	1.79	0.00	2.51	2.51	0.00	2.15	2.15
		I	1	1		1			1	r			1			1	1				1

* Days.

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Tabl	le 16.	Est	imate	ed a	amounts	of	released	nitr	ogei	n at	var	ious	time	interval	ls,	folle	-wc
ing	addit	ion c	of eit	her	glucose	an	d (NH ₄) ₂	SO_4	or	gluco	ose	and	NaNO	3 to Hno	F	soil.	
													(mg	N/100	g	dry s	oil)

0.01		Incul				
C/N	1	2	4	8		12
			$(\mathrm{NH}_4)_2\mathrm{SO}_4$			
$\frac{2}{8}$ 16	0.59 0.77 0.48 0.95	0.75 I.18 0.95 3.02	1.79 2.51 2.44 5.0;3	1 43 2:12 2.92 6.35	i i i	1.72 2.29 2.56 6.56
		L .	NaNO3			
2 4 8 16	$0.05 \\ 0.63 \\ 0.48 \\ 1.15$	$0.41 \\ 0.06 \\ 1.79 \\ 2.06$	0.29 0.83 2.16 2.67	0.63 1.85 2.58 4.18		0.71 1.49 2.86 4.39
			l.		1	

Table 17. Estimated rate of release of immobilized nitrogen per half a week at various time intervals, following addition of either glucose and $(NH_4)_2SO_4$ or glucose and NaNO₃ to HnoF soil.

(mg N	/100	g	dry	soil/half	week)
· ·	,				

C / N	Incubation period (weeks)											
C / N	1	2	4	8	12							
			$(NH_4)_2SO_4$									
2	0.52	0.08	0.26	-0.06	0.03							
4 8	0.67	0.24	Q. \$7	- 00 08	-0.02							
16	0.83	1.04	0.50	0.22	0.02							
			NaNO3									
2	0.04	0.18	-0.03	0.06	0.01							
4	0.55	-0.29	0.19	0.17	-0.04							
8	0.42	0.66	0.09	0.07	0.03							
16	1.01	0.46	0.15	0.25	0.02							

in this work will be concluded as follows:

(a) Immobilization of nitrogen in the soils receiving glucose and mineral nitrogen as either ammonium sulfate or sodium nitrate at different levels of C/N ratio proceeded rapidly and reached its maximum on a 3-day incubation irrespective of the forms of nitrogen added and the levels of C/N ratio treated. The amount of the nitrogen immobilized increased with increasing C/N ratio independent of the forms of nitrogen added. These tendencies support the findings of Winsor et *al.* (1956), Allison *et al.* (1960) and Harada *et al.* (1968). Figures for the quantity of a maximum-immobilized nitrogen at a given C/N ratio in each soil was a little larger in the form of ammonia than in the form of nitrate. From this result, it is likely that the soil microorganisms concerned in immobilization of nitrogen prefer ammonia rather to nitrate, as pointed out by several

investigators (Richards *et al.* 1935, Winsor 1956, Jansson 1958, Broadbent *et al.* 1962). Comparison between the maximum immobilization of nitrogen at the C/N ratios of 4, 8 and 16 of the lowland soils and that at the corresponding C/N ratios of the highland soils led to a result that lowland soils might be better in respect of immobilization of nitrogen in soils receiving glucose and mineral nitrogen than highland soils.

(b) Next, immediately after reaching a maximum immobilization of nitrogen in soil release of nitrogen occurred and increased with the period of incubation, the increase being probably due to mineralization of a part of the nitrogen originally immobilized in soil. The amounts of released nitrogen increased with increasing amounts of immobilized nitrogen, and the rate of release of immobilized nitrogen per half a week in each soil was largest immediately after reaching a maximum immobilization of nitrogen in all the C/N ratios and the forms The rate was faster at an early period of incubation than of nitrogen added. at a late period of incubation. Table 18 gives the ratios of the nitrogen released for 12-week incubation to the nitrogen immobilized for 3-day incubation on which immobilization of nitrogen in each soil reached its maximum. The data showed that when the amount of nitrogen released at a given C/N ratio was expressed as percentage of the maximum-immobilized nitrogen at a corresponding C/N ratio, the percentage was higher in the form of nitrate than in the form of ammonia. This experimental result will be caused by the changing of pH in the soil after reaching a maximum immobilization of nitrogen added in the forms of either ammonia or nitrate, i. e., the increase in pH accompanying the immobilization of sodium nitrate-N and the fall in pH accompanying the immobilization of ammo-

C/N	ratios	mobilized N for Released N for 12- -day incubation (a) week incubation (b)				(b)/(a)×100	
5011 tre	ated	(NH ₄) ₂ SC added	D ₄ Na ad	$\begin{array}{c c c c c c c c c c c c c c c c c c c $	SO₄ NaNO₃ d added	$(NH_4)_2SO_4$ added	NaNO3 added
$\longleftarrow mg N per 100 g dry soil \longrightarrow \qquad \longleftarrow \ \% \longrightarrow$							
LF	4 8 16	6.35 1 1 . 9 2 17.57	4.63 9.18 15.92	2.89 4.67 8.36	2.99 4.21 8.37	47.0 38.2 47.6	66.0 46.0 52.5
Averag	Average					44.2	54.8
LnoF	4 8 16 i	7.44 12.08 17.57	4.81 11.11 17.37	2.95 4.03 7.03	1.92 4.74 7.53	39.6 33.3 40.2	40.0 43.0 43.0
Average						37.7	42.0
HF	4 8 16	5.84 10.63 14.44	4.73 6.35 12.86	2.35 3.63 6.50	2.32 3.29 6.87	23.5 34.2 45.0	49.0 51.8 53.0
Averag	ge					34.2	51.3

Table 18. Ratio of the nitrogen released for 12-week incubation to the maximum-immobilized N.

0.01	Incubation period (days or weeks)							
C/N	3**	1	2	- 1		8	12	
0	(NH ₄) ₂ SO ₄							
16	6.25.6	G. 5.61	1				4.8	
Check	6.2	6.2	5.5 6.1	$5.2 \\ 6.0$	I	4.9 5.9	4.9 5.9	
0 1		_ '-	NaNO	D ₃				
16	6.2 6.6	6.3	6.3	6.3	I	6.1	6.0	
Check	6.2	$\begin{array}{c} 6.6\\ 6.2 \end{array}$	6.7 6.1	$6.5 \\ 6.0$	i 	6.3 5.9	6.2 5.9	

Table 19. Changes of soil pH* during the each interval of incubation period following addition of mineral nitrogen as $(NH_4)_2SO_4$ or $NaNO_3$ and glucose to HF soil.

* Initial pH: 6.2.

** Days.

nium sulfate-N (see Table 19).

(c) Finally, the numerical relationship between carbon initially added to soil and nitrogen immobilized which was proposed by Winsor (1958) is of particularly interest when immobilization of nitrogen reaches its maximum, this being attained on incubation of approximately three days under the experimental condition used in this experiment. Therefore, ratio of carbon added initially to nitrogen immobilized for 3-day incubation was given in Table 20. The data led

Table 20. Ratio of carbon added initially to nitrogen immobilized on incubation for three days.

Soil	Carbon added	Nitrogen immol incubat	ilized for S-day ion (b)	(a)/(b) ratios		
	(a)	$(\mathrm{NH}_4)_2\mathrm{SO}_4$ added	NaNO₃ added	$(NH_4)_2SO_4$ added	NaNO₃ added	
	mg C per 100 g dry soil	mg N 100 g c	per Iry soil		1	
LF	40 80 160 320	3.32 6.15 11.92 17.57	3.31 4.63 9.18 15.92	13.6 14.6 13.4 38.2	12.1 17.2 17.4 "0.1	
LnoF	40 80 160 1 320	5.67 7.44 12.08 17.57	2.39 4.81 11.11 17.37	11.7 10.8 13.3 18.1	16.7 1.6.G 14.4 18.4	
HF	40 80 160 320	$2.98 \\ 5.84 \\ 10.63 \\ 14.44$	2.73 4.73 6.35 12.86	13.4 13.7 15.1 22.0	14.7 16.9 25.2 24.8	
HnoF	80 160 320	3.40 6.05 9.36 14.19	2.34 4.12 7.90 11.44	11.6 13.4 17.1 22.7	16.7 16.8 19.1 25.4	

to results that the amount of carbon which must be added to the soil in order to immobilize one unit of nitrogen into organic form ranged from 11 to 25 units, increasing with the increasing carbon added initially, and it was larger in the form of nitrate than in the form of ammonia at a given C/N ratio of all the soils used. These results were different from the findings of Winsor *et al.* by which there was little difference between in the form of nitrate and in that of ammonia.

CONCLUSIONS

The effect of nitrogenous forms on immobilization and release of nitrogen following addition of glucose and mineral nitrogen to soils was studied. Principal experimental results obtained revealed the following facts :

1) Immobilization of nitrogen in soils receiving glucose and mineral nitrogen as ammonium sulfate or sodium nitrate at different levels of C/N ratio proceeded rapidly and reached its maximum on incubation for 3 days irrespective of the forms of nitrogen added and the levels of C/N ratio treated. The amount of nitrogen immobilized increased with increasing C/N ratio independent of the forms of nitrogen added. The nitrogen immobilized to a maximum at a given C/N ratio was a little larger in the form of ammonia than in the form of nitrate.

2) Immediately after reaching maximum immobilization of nitrogen release of nitrogen occurred and increased with the period of incubation, the increase being probably due to mineralization of part of the nitrogen originally immobilized. The rate of release of nitrogen was faster at the early period of incubation than at the late period of incubation. When the amount of nitrogen released at a given C/N ratio was expressed as percentage of the maximum-immobilized nitrogen at a coresponding C/N ratios, the percentage was higher in the form of nitrate than in the form of ammonia. This result might be caused by the changing of pH in the soil after reaching a maximum immobilization of nitrogen added in the forms of either ammonia-N or nitrate-N.

3) The amount of carbon which must be added to the soil in order to immobilize one unit of nitrogen into organic form increased with the increasing carbon added initially and it was larger in the form of nitrate than in the form of ammonia at a given C/N ratio of all the soils used.

Note: This paper is a part of the work entitled "Factors affecting immobilization and release of nitrogen in soil and chemical characteristics of the nitrogen newly immobilized".

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