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Gaseous Losses of Nitrogen from the Soil of Semi-Arid Regions

T. C. Tucker

R. L. Westerman

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GASEOUS LOSSES OF NITROGEN FROM THE SOIL OF SEMI-ARID REGIONS

T. C. Tucker, Project Leader

and

R. L. Westerman

University of Arizona

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ABSTRACT

Soil profile samples were selected for laboratory studies of gaseous nitrogen (N) losses from two soil types at each Sonoran Desert Validation Site near Tucson. Oxygen consumption, gaseous evolution during anerobic incubation employing $^{15}NO_3$, changes in the N fractions, and most probable number (MPN) of denitrifying organisms were examined to determine N loss potential. The potential for gaseous loss of N was observed in all soils at all depths and resulted in $^{15}N_2$ and $^{15}N_2O$ evolution. The calculated loss in 18 days of added $^{15}NO_3$ ranged from approximately one-tenth to more than two-thirds of the total amendment. An organic carbon energy source was included in portions of these studies. Glucose additions increased N in the gaseous phase and increased the calculated loss based on ^{15}N remaining in the soil after incubation. Oxygen consumption increased, indicating biological and/or chemical activity, when the soils were moistened. Although oxygen was consumed in soils from all profile depths within 24 hr, the rate of consumption was higher in the surface soils of all profiles except the one having the buried A horizon. The number of denitrifying organisms under saturated moisture increased with soil depth, incubation time, and additions of NO_3-N and organic carbon.

INTRODUCTION

This study was conducted to determine the potential for gaseous losses of nitrogen (N) from native desert soils and to quantify these losses in relation to the various parameters of the system. The previous work was concerned with the potential for gaseous loss and the identity of the gaseous products evolved, as well as techniques for the study. Some of this work was continued during 1972. The biological and non-biological contributions to gaseous N losses were studied and satisfactory techniques have been developed to differentiate between these two processes. Initial studies on rates of gaseous N loss as a function of temperature, moisture, nitrate concentrations, and organic carbon have been initiated and will be continued.

OBJECTIVES

The objectives for 1972 were:

1. To complete the study of denitrification potential on soil samples from various profile depths of two soils from each Sonoran Validation Site -- Santa Rita and Silverbell.
2. To determine the relative contributions of non-biological and biological gaseous losses.
3. To initiate rate studies of the gaseous loss process in relation to moisture, temperature, nitrate concentration and organic carbon source. This phase of the study will be continued and completed in 1973.

METHODS

Soil profile samples (DSCODE A3UTH-) were taken from two locations on the Santa Rita site (Sonoita sandy loam - SR-1, Anthony sandy loam - SR-2), and two from the Silverbell site (Rillito loam - SB-1, Rillito loam, eroded - SB-2). Sampling depths were 0-5, 15-20, 30-35, 60-65, and 90-95 cm, with exception of the 90-95 cm depth in SB-2 because of bedrock. These soils were air-dried, crushed, passed through a 2 mm sieve, and stored in plastic containers with sealed air-tight lids.

Ten-g air-dry soil samples from desert profiles were placed individually into 125 ml Warburg respirometer flasks and moistened with 2 ml of H_2O. One ml of 20% KOH

was placed in the center well and oxygen and gas exchange were measured at 37 C using standard Warburg respirometer techniques (Umbreit, Burris and Stauffer, 1964).

Fifty-g samples of air dry soil were placed in calibrated 125 ml Erlenmeyer flasks containing 2 ml of saturated KOH in wells. The samples were moistened with 10 ml solutions containing 4.9 g of NO_3-N in water or in 0.25 M glucose solution. The NO_3-N added was enriched with 33 atom percent ^{15}N. The flasks were closed with rubber septum caps, flushed for 10 min with argon gas, then incubated for 18 days at 36 C. Gas samples were taken from the air in each flask with a gas tight syringe at varying time intervals and analyzed by mass spectrometry. Calculations were made for gaseous composition, ^{15}N in the N_2 and N_2O components, and N_2 to N_2O ratios. Samples of the initial soil before treatment and after incubation were analyzed for NH_4^+-N, NO_2^- + NO_3^- - N, and organic N fraction by micro-Kjeldahl procedure (Bremner, 1965). Atom percent ^{15}N in each fraction was determined by mass spectrometry and percent loss of added NO_3-N was calculated.

Detection of the presence of denitrifying organisms in soil profiles was accomplished using standard techniques (Alexander, 1965a). One hundred ml of sterile water was added to 10-g soil samples in 150 ml bottles which were shaken horizontally in a mechanical shaker for 10 min. Samples were removed from the shaker and were shaken again by hand just prior to removing a 10 ml aliquot with a sterile pipette from the center of the suspension. The aliquot was added to 90 ml of sterile water. One ml aliquots from the above dilution were added to each of five sterile tubes containing the sterile liquid denitrifican medium. The tubes were plugged and incubated for 5 days at 37 C. Presence of denitrifiers was determined in this specific medium by a pH indicator color change from green to blue plus the presence of gas (Alexander, 1965a). It was difficult to identify denitrifiers by this procedure and a more sensitive procedure was adopted. A nitrate nutrient agar was inoculated and sulfanilic acid and α-napthylamine was used to detect the presence of nitrite (Difco, 1953). Denitrifiers were detected by a pink color, specific for nitrite, and presence of gas bubbles in the agar.

Most probable numbers of denitrifying organisms were measured at 37 C using procedures outlined by Alexander (1965b) in a specific liquid denitrifican broth. The liquid broth contained succinic acid, K_2HPO_4, $MgSO_3 \cdot 7H_2O$, KNO_3, $CaCO_3$, yeast extract and tap water, respectively, in the following g/l: 2.0, 1.0, 0.5, 10.0, 2.0, 1.0, with tap water used to make to final volume.

Soils were sterilized using standard steam sterilization procedures (Alexander, 1965a) and a methyl bromide gas treatment developed in this laboratory. Ten-g soil samples were spread thinly over a 9 cm petri dish and steam sterilized in an autoclave

for 30 min at 121 C and 15 psi. For methyl bromide sterilization, 10-g samples of soil were placed in tubular glass and separated by cotton. The gas was diffused through the soil slowly for 24 hr using an alcohol absorbent trap to catch the CH_3Br gas passing through the column. Following the methyl bromide gassing the soils were scrubbed for 8 hr with compressed air that had been passed through dilute sulfuric acid and deionized water baths. This scrubbing procedure was adopted to remove CH_3Br adsorbed to clay surface areas.

After sterilization by steam or by methyl bromide, the soils were placed aseptically in sterile glass petri dishes and allowed to de-gas for 24 hr. Then soils were placed into acetone-heat-treated Warburg flasks for oxygen consumption studies. Other soil samples were plated on denitrifican agar to check sterility.

RESULTS

Wetting dry desert soils increased the oxygen consumption, which showed in the initiation of biological and/or chemical activity (Table 1, DSCODE A3UTH04). No consistent pattern was evident for differences between soils or in profile depths. However, oxygen consumption tended to increase with increasing organic matter in the profile. For example, the Anthony sandy loam (SR-2) has a buried A horizon at approximately 60 cm and is higher in organic matter (0.39%) and consumed more oxygen (0.15 µl/g/hr) when moistened than the overlying sandy material which was lower in organic matter (0.17%) and consumed less oxygen (0.03 µl/g/hr).

Table 1. Organic matter and oxygen consumption in Sonoran Desert profile soil samples after wetting DSCODE——A3UTH04

Soil	SR-1		SR-2		SB-1		SB-2	
	µl O_2/g/hr	%O.M.	µl O_2/g/hr	%O.M.	µl O_2/g/hr	%O.M.	µl O_2/g/hr	%O.M.
Depth, cm								
0-5	0.16	0.46	0.09	0.27	0.14	0.32	0.17	0.54
15-20	0.17	0.37	0.04	0.16	0.09	0.28	0.13	0.53
30-35	0.11	0.26	0.03	0.17	0.10	0.34	0.12	0.39
60-65	0.05	0.23	0.15	0.39	0.12	0.29	0.18	0.35
90-95	0.11	0.18	0.15	0.35	0.05	0.24	- bedrock -	

Percentages of N_2, O_2, N_2O and the N_2/N_2O ratio in the gas phase above soils incubated anaerobically are shown in Table 2. In soils amended with nitrate, the percentage of N_2 and O_2 increased with time in practically all depths of the three soil profiles. There were only slight changes in the percentage of N_2O, regardless of time, depth or soil profile. The N_2/N_2O ratios ranged from 138 to 6773 in the soils that were amended with nitrogen. In soils amended with nitrate plus glucose, the percentages of N_2 in the atmosphere above incubating soils generally were higher than observed in soils amended with only nitrate, and tended to increase with time. Percent oxygen tended to be lower at the 355 hr time of sampling than was observed at previous sampling times. The N_2/N_2O ratios were much smaller than in the soil amended with only nitrate, indicating a larger percentage of the gas was in the form of N_2O. The presence of the organic energy source apparently stimulated loss of nitrogen in the form of N_2O and increased the amount of N_2 gas in the atmosphere above the incubating soils.

Table 2. Composition of gas above Sonoran Desert soil profile samples incubated under argon atmosphere and amended with nitrate and nitrate plus glucose
DSCODE---A3UTH01

Depth (cm)	Time (hr)	N_2	O_2	N_2O	N_2/N_2O
			%		
		Anthony sandy loam (SR-2) - Nitrate			
0- 5	18	7.37	1.131	0.012	640
	91	14.86	2.460	0.039	383
	187	14.38	2.381	0.024	609
	355	14.45	2.473	0.015	971
15-20	18	0.94	0.198	0.001	1238
	91	5.49	1.066	0.006	959
	187	8.06	1.588	0.004	1873
	355	15.87	3.014	0.005	3051
30-35	18	2.90	0.481	0.003	899
	91	3.07	0.609	0.017	180
	187	5.25	1.136	0.013	400
	355	7.01	1.739	0.010	680
60-65	18	8.03	1.155	0.015	535
	91	13.42	1.995	0.038	355
	187	9.92	1.187	0.005	2031
	355	12.99	1.250	0.012	1086
90-95	18	11.05	1.697	0.024	456
	91	15.86	2.460	0.115	138
	187	16.60	2.481	0.037	500
	355	12.02	1.387	0.016	758

Continued

Table 2. Continued

Depth (cm)	Time (hr)	N_2	O_2 %	N_2O	N_2/N_2O
		Anthony sandy loam (SR-2) - nitrate + glucose			
0- 5	20	2.71	0.288	0.051	53
	91	21.05	2.193	0.277	76
	188	3.38	0.314	0.039	86
	355	29.48	0.372	0.564	52
15-20	20	1.06	0.055	0.031	34
	91	13.77	1.886	0.098	140
	188	16.20	0.406	0.077	211
	355	26.30	0.419	0.126	210
30-35	20	1.49	0.170	0.071	21
	91	13.49	1.433	0.028	478
	188	16.69	1.570	1.848	9
	355	22.04	0.588	0.048	457
60-65	20	10.10	1.234	0.059	171
	91	25.24	2.508	0.041	612
	188	38.31	3.048	2.970	13
	355	39.61	0.611	8.733	5
90-95	20	10.64	0.919	0.021	508
	91	32.58	3.141	0.094	348
	188	41.92	3.089	1.895	22
	355	47.54	1.581	8.520	6
		Rillito loam (SB-1) - nitrate			
0- 5	18	10.47	1.574	0.034	312
	91	20.60	2.989	0.067	309
	187	25.19	3.232	0.013	1894
	355	25.05	3.062	0.006	4464
15-20	18	2.79	0.405	0.004	676
	91	15.75	2.584	0.055	286
	187	17.80	2.838	0.021	842
	355	14.10	2.012	0.005	2801
30-35	18	2.51	0.304	0.006	388
	91	13.40	2.011	0.036	375
	187	24.40	3.679	0.026	932
	355	24.42	3.430	0.010	2489
60-65	18	9.66	1.405	0.016	596
	91	25.17	3.756	0.024	1030
	187	35.24	5.068	0.013	2743
	355	44.16	6.326	0.007	6773
90-95	18	1.11	0.116	0.003	396
	91	10.18	1.580	0.042	241
	187	6.96	1.026	0.004	1691
	355	18.75	2.911	0.006	3023

Continued

Table 2. Continued

Depth (cm)	Time (hr)	N_2	O_2 %	N_2O	N_2/N_2O
			Rillito loam (SB-1) - nitrate + glucose		
0- 5	20	7.12	0.890	0.098	72
	91	17.11	6.640	0.079	217
	188	32.65	3.370	0.458	67
	355	31.04	1.099	4.813	6
15-20	20	7.10	0.702	0.123	58
	91	17.97	1.067	0.013	1370
	188	32.55	2.273	0.149	219
	355	42.21	0.808	5.276	8
30-35	20	4.29	0.289	0.210	20
	91	27.40	2.858	0.035	794
	188	31.76	1.827	0.027	1194
	355	22.02	0.229	0.012	1775
60-65	20	7.84	0.952	0.283	28
	91	31.59	3.658	0.085	371
	188	21.46	0.063	0.454	47
	355	36.50	0.801	4.587	8
90-95	20	36.70	0.150	0.166	22
	91	26.58	2.114	0.031	860
	188	31.15	0.091	0.003	10383
	355	51.50	0.641	0.536	96
			Rillito loam, eroded(SB-2) - nitrate		
0- 5	18	8.51	1.098	0.036	234
	91	20.58	2.586	0.044	465
	187	26.05	2.354	0.015	1728
	355	38.40	3.981	0.017	2305
15-20	18	1.32	0.119	0.006	209
	91	17.93	2.498	0.084	213
	187	20.12	2.041	0.010	1971
	355	29.50	2.414	0.008	3508
30-35	18	1.40	0.136	0.008	173
	91	7.30	0.983	0.018	404
	187	12.08	1.531	0.009	1324
	355	13.07	1.204	0.005	2558
60-65	18	0.82	0.051	0.004	188
	91	13.62	2.047	0.025	605
	187	16.82	2.378	0.013	1281
	355	12.03	1.229	0.003	3965

Continued

Table 2. Continued

Depth (cm)	Time (hr)	N_2	O_2 %	N_2O	N_2/N_2O
			Rillito loam, eroded (SB-2) - nitrate + glucose		
0- 5	20	7.08	0.668	0.147	48
	91	26.76	1.915	0.023	1154
	188	27.35	0.154	0.028	980
	355	43.88	0.555	3.109	14
15-20	20	6.82	0.507	0.018	377
	91	13.70	0.122	0.016	869
	188	30.20	0.328	0.085	356
	355	46.99	1.265	4.039	12
30-35	20	11.21	0.460	0.012	969
	91	25.56	0.342	0.007	3695
	188	34.98	0.243	0.036	976
	355				
60-65	20	2.61	0.032	0.007	351
	91	8.98	0.017	0.044	204
	188	22.47	0.124	0.771	29
	355	51.52	0.262	6.118	8

Data in Table 3 illustrate the amounts of nitrogen in different forms in initial profile samples and after 18 days of incubation following the addition of $^{15}NO_3$-N either with or without glucose. Initial inorganic N fractions generally were low in all samples except the surface of SB-2 and depth samples of SB-1. These samples were higher in NH_4^+-N than has been found for most desert soils. A large amount of NO_3-N remained in all samples following incubation when only nitrate was added. However, the addition of the organic carbon source, glucose, resulted in almost complete disappearance of nitrate in all samples but the Anthony (SR-2) sandy surface soil. The result of adding nitrate was increased organic N in many but not all samples. The NH_4^+-N fraction increased in the Rillito (SB-2) soil, and to a greater extent with glucose.

The calculated loss of $^{15}NO_3$-N during incubation is given in Table 4. Losses without glucose ranged from approximately one-tenth of added nitrate to more than one-half from different soils and depths. With glucose addition losses increased until two-fifths to more than two-thirds of added nitrate could not be recovered.

The surface 0-5 cm depths of all the soil profiles were checked for the presence of denitrifying organisms (Table 5). Denitrifying organisms were positively identified in all soils using procedures outlined by Alexander (1965a).

Table 3. Soil nitrogen forms in Sonoran desert profiles — DSCODE A 3UTH03

		Soil N Forms								
		NH_4^+ - N			NO_2^- + NO_3^- - N			Organic - N		
			Amended*			Amended			Amended	
Soil	Depth(cm)	Initial	NO_3	NO_3 + Glucose	Initial	NO_3	NO_3 + Glucose	Initial	NO_3	NO_3 + Glucose
					- µg of N/g of soil -					
Sonoita sandy loam (SR-1)	0-5	0.7	1.2	0.0	0.5	52.1	0.6	265.0	243.2	274.2
	15-20	1.2	2.4	1.6	1.1	99.5	2.0	256.7	218.1	293.1
	30-35	0.7	1.4	0.3	0.2	90.0	1.8	169.1	207.2	269.9
	60-65	0.6	0.3	1.1	0.8	89.3	1.7	152.3	128.8	274.8
	90-95	0.6	0.9	2.1	0.4	88.4	0.2	65.2	114.6	205.5
Anthony sandy loam (SR-2)	0-5	1.1	0.3	1.2	0.6	87.4	53.9	151.3	168.0	165.6
	15-20	0.7	0.4	2.0	1.3	91.7	2.1	59.6	112.6	160.4
	30-35	0.6	0.6	0.0	0.4	86.5	0.0	79.3	226.0	158.8
	60-65	0.9	3.1	0.3	1.7	83.4	1.0	244.7	419.6	392.6
	90-95	1.0	1.2	0.8	1.6	83.4	0.3	244.9	290.0	315.3
Rillito loam (SB-1)	0-5	2.2	0.7	0.8	2.1	70.9	0.8	191.2	209.9	242.0
	15-20	0.8	2.8	0.0	0.2	87.2	0.3	146.5	126.3	216.5
	30-35	4.6	2.7	3.6	1.0	89.2	0.9	171.7	248.1	253.6
	60-65	3.4	1.8	2.6	0.4	89.2	0.0	209.1	310.5	278.7
	90-95	2.6	0.8	0.3	0.6	93.9	0.2	194.9	215.1	217.4
Rillito loam, eroded (SB-2)	0-5	9.1	1.7	6.6	1.2	71.4	0.0	354.9	460.3	407.3
	15-20	1.6	8.1	16.4	0.3	96.3	0.0	407.0	447.0	543.2
	30-35	0.5	6.2	5.9	0.1	88.6	0.3	362.7	374.9	474.3
	60-65	0.6	2.8	9.4	2.0	93.0	0.1	262.8	360.0	450.4

*Amended with NO_3 or NO_3 + glucose and incubated 18 days @ 36 C under argon atmosphere.

Table 4. Calculated loss of added $^{15}NO_3$ in soil samples incubated under argon for 18 days at 36 C — DSCODE A3UTH01

Depth(cm)	SR-1		SR-2		SB-1		SB-2	
	NO_3	NO_3 + Glucose	NO_3	NO_3 + Glucose	NO_3	NO_3 + Glucose	NO_3	NO_3 + Glucose
	% loss							
0-5	52.8	66.9	13.5	62.7	36.3	66.2	17.9	60.3
15-20	19.8	57.1	36.2	48.6	32.9	65.0	11.8	39.1
30-35	21.8	41.6	28.0	55.0	23.7	69.5	20.3	47.1
60-65	27.7	39.2	25.3	49.7	22.5	52.4	16.4	42.1
90-95	36.1	45.9	23.9	46.6	13.6	60.0	-	-

Table 5. Detection of denitrifying organisms in the 0-5 cm depth of Sonoran desert soils

Soil Condition	SR-1	SR-2	SB-1	SB-2
Dry	+	+	+	+
Wet	+	+	+	+

+ indicates presence of denitrifiers

In order to differentiate between biological and non-biological gaseous losses of N, a soil sterilization procedure had to be employed that would not destroy the physical structure or the chemical constituents of the soil. Standard steam sterilization procedures were inadequate because of the gross destruction of the clay minerals and changes in chemical constituents that occur when soils are subjected to high temperatures and pressures. A sterilization procedure using methyl bromide has been adopted and compared with steam sterilization and unsterilized soils for the presence of viable organisms on specific denitrifican agar plates. Data of a portion of the study are shown in Table 6. Agar plates inoculated with methyl bromide sterilized soil remained sterile for five days and longer. It is interesting

to note that steam sterilization of soil samples for 30 min at 121 C and 15 psi did not reduce the viable denitrifiers. The methyl bromide procedure is being used to differentiate between biological and non-biological gaseous losses of nitrogen.

Table 6. Detection of denitrifiers in steam sterilized and methyl bromide sterilized soils from the 0-5 cm depth of an eroded Rillito loam (SB-2)

	Unsterilized Days					Steam sterilized Days					CH_3Br Days				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Wet soil	+	+	+	-	+	+	+	+	+	-		no wet soil			
Dry soil	+	+	+	+	+	+	+	+	+	+	-	-	-	-	-
Wet soil extract 10:100	+	+	-	-	-	+	+	+	+	+		no wet soil			
Dry soil extract 10:100	+	+	+	+	+	+	+	+	+	-	-	-	-	-	-
Blank	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

+ indicates presence of denitrifiers

Data for a detailed oxygen exchange study with time at all depths of two soils from Santa Rita and two soils from Silverbell are reported in Table 7. It is interesting to note that in the lower depths of the Sonoita sandy loam (SR-1) and the upper depths of the Anthony sandy loam (SR-2) gases are evolved during the first 5 hr rather than oxygen being consumed. However, oxygen was consumed in all depths of all profiles except the Anthony sandy loam (SR-2) which has a buried A horizon at approximately 60 cm.

Oxygen exchange data in a Sonoita sandy loam (site 2, different location than SR-1) that had been steam or methyl bromide sterilized are reported in Table 8. Oxygen consumption increased with time in the unsterilized soil with the highest amount of oxygen being consumed in the surface soil. Both steam and methyl bromide sterilization resulted in gaseous evolution. In the case of steam sterilized soils, the oxygen evolution probably resulted from broken chemical bonds and clay lattices that were collapsed during the heating with pressure in the autoclave. Only the lower depths of the soil showed oxygen consumption at 4 and 24 hr. Methyl bromide sterilization resulted in gaseous evolution from all depths with time. The gas evolved was thought to be CH_3Br adsorbed to clay surfaces which was released upon wetting and/or oxygen as a result of bromination of chemical constituents in the soil.

Table 7. Oxygen exchange in four Sonoran Desert soil profiles after wetting DSCODE— A3UTH04

Depth, cm	Sonoita sandy loam (SR-1) hours				Anthony sandy loam (SR-2) hours				Rillito loam (SB-1) hours				Rillito loam-eroded (SB-2) hours			
	1.0	3.0	5.0	24.0	1.0	3.0	5.0	24.0	1.0	3.0	5.0	24.0	1.0	3.0	5.0	24.0
	µl O_2/10g of soil															
0- 5	52.2	76.4	35.8	414.3	+8.0	+11.7	+19.5	160.5	19.8	59.9	88.1	402.1	76.9	90.3	62.7	463.6
15-20	43.4	71.3	35.5	340.3	+3.9	+27.8	+47.8	71.5	4.0	23.9	21.8	119.2	59.6	107.5	59.8	322.2
30-35	+8.0	+19.8	+4.0	175.1	+11.9	12.3	40.4	92.0	4.0	44.1	80.0	204.6	60.9	100.7	68.1	284.8
60-65	+7.7	+11.9	+12.1	127.7	0.2	31.7	47.5	198.0	+11.9	11.9	43.7	182.9	77.7	103.0	51.2	317.5
90-95	+11.9	+39.8	+71.6	107.6	0.0	18.8	39.6	266.4	15.7	3.7	+0.2	111.7	--	--	--	--

+ indicates oxygen released from the soil.

Table 8. Oxygen exchange in a Sonoita sandy loam after steam sterilization and methyl bromide treatment — DSCODE A3UTH04

Depth(cm)	Unsterilized hours			Steam sterilized hours			CH_3Br hours		
	0.5	6.0	26.0	1.0	4.0	24.0	1.0	6.0	24.0
	µl O_2/10g of soil								
0-5	15.9	131.1	401.2	+ 11.6	+ 7.8	+244.5	+43.5	+63.4	+126.7
10-15	11.8	70.9	201.0	+104.5	+128.7	+101.4	+35.3	+58.7	+ 93.7
25-30	4.0	23.9	91.7	+ 87.4	+119.3	+135.5	+19.6	+27.3	+ 43.0
50-55	4.1	20.3	32.4	+ 43.1	15.8	82.6	+39.3	+39.2	+ 35.4
75-80	+11.9	11.9	127.2	+ 24.0	12.1	88.0	+58.8	+58.8	+ 46.6

+ indicates gas released from the soil

Most probable numbers (MPN) of denitrifying organisms in the initial air-dry soil and soils saturated and partially saturated that were incubated 5, 10, and 15 days at 37 C are shown in Table 9. The MPN of denitrifiers in the initial air-dry samples were low but increased with depth. A cholla cactus root was decaying in the 25-30 cm depth and the organic carbon energy source was the apparent reason for the increased MPN of denitrifiers. The MPN of denitrifiers in the unsaturated soils were always less than the MPN of denitrifiers in the saturated soils. In unsaturated soils MPN of denitrifiers increased in 10-day incubation but decreased with 15-day incubation. In saturated soils MPN of denitrifiers increased with depth and time in all cases except in the 15-day, 25-30 cm depth incubation. With the large number of denitrifiers, energy may have been limiting.

Table 9. Number of denitrifiers in a Sonoita sandy loam (site-2) incubated at 37 C aerobically under partially saturated and saturated conditions — DSCODE A3UTH03

Depth(cm)		Days t0	t5	t10	t15
		denitrifiers/g of soil			
0-5	Air Dry	0.2	---	---	---
	53% Sat.	---	4.9	11.0	17.0
	Sat.	---	22.0	220.0	790.0
10-15	Air Dry	0.6	---	---	---
	53% Sat.	---	7.9	79.9	46.0
	Sat.	---	140.0	280.0	1,100.0
25-30	Air Dry	42.0	---	---	---
	53% Sat.	---	17.0	35.0	13.0
	Sat.	---	490.0	2800.0	2,200.0

2.3.4.3.-14

During the period October 14-24, 1972, we cooperated with and supported Dr. Roy Cameron (JPL and Utah State University) in a series of broadscale field experiments at the Silverbell site to gather data that would be representative of actual field conditions. Data pertaining to algae activity, gaseous composition of the soil atmosphere, fluxes in the microbial population, and nitrogen transformations were collected before, during, and after an intensive rainstorm. The nitrogen transformation data were processed in our laboratory and have been forwarded to Dr. Cameron to be included in his report.

DISCUSSION

Results of 1971 demonstrating the potential for gaseous N losses were expanded in 1972 to include three additional soil profiles. Gaseous loss of N occurred at all depths of all soil profiles and was enhanced by additions of nitrate and organic carbon. The total N loss can be appreciable when conditions are favorable, as indicated by the low amount of NO_3-N remaining in the soils after incubation.

The oxygen consumption technique was useful in evaluating sterilization methods which will be used to distinguish between potential biological and non-biological N losses.

Methyl bromide and steam sterilization methods were compared and the methyl bromide technique was adopted for further study of biological vs. non-biological gaseous N losses. Methyl bromide treated soil samples were void of denitrifying organisms.

Studies on the rate of gaseous N loss have been initiated that include variables of moisture, temperature, soil depth, organic carbon, NO_3-N concentration, and time. Other studies include methyl bromide sterilization in relation to several variables mentioned above as well as complete anaerobic incubation (Helium atmosphere).

Apparently not all of the denitrifying activity is confined to the upper surface of the desert soils, as indicated by the high number of denitrifying organisms in the lower depths of the profiles studied. The presence of organic material from decaying roots may have contributed to the higher number of denitrifiers in the lower depths of the observed profiles.

EXPECTATIONS

It is anticipated that the contribution of gaseous loss of N from non-biological and biological sources will be resolved in the immediate future, since adequate sterilization techniques have been developed. Preliminary indications are that non-biological contributions are negligible. Experiments have been initiated to determine rate of gaseous N loss as influenced by moisture, temperature, soil depth, organic carbon, and NO_3-N concentration. Experiments are in progress and data for one profile are almost complete. After evaluation of these results, additional profiles will be studied with modifications of variables deemed appropriate.

In order to relate results obtained under laboratory conditions more closely to field conditions a series of field experiments will be conducted in 1973. Metal cylinders will be driven into the soil to various depths and known amounts of organic material with $^{15}NO_3$ will be incorporated in the soil. These cylinders will be closed at the top with plastic or metal foil. Acid and alkaline traps will be placed on the surface of the soil inside the cylinders to trap NH_3 and CO_2 gases evolved. Decomposition rates of organic carbon and nitrogen and the transfer of organic N to soil N as well as N loss will be measured at scheduled time intervals throughout the year.

Rates of gaseous loss of N as influenced by moisture, temperature, soil depth, organic carbon, and NO_3 concentration, in the laboratory studies as well as data pertaining to gaseous losses, decomposition of organic carbon and transfer of organic N to soil N under actual field conditions, will be very useful in the modelling of the ecosystems in semi-arid regions.

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