METHODOLOGY FOR THE DETERMINATION OF TOTAL NITROGEN IN PLANT MATERIALS, AND THE DISTRIBUTION OF FERTILIZER NITROGEN -15 IN WINTER WHEAT UNDER TWO TILLAGE SYSTEMS

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INTRODUCTION

There are two parts to this dissertation pertaining to two separate studies. Both parts are presented in a format suitable for publication in a professional journal.

In the process of carrying out research concerning the use of fertilizer N in a soil-plant system, it is essential to have a reliable procedure for the determination of total N that quantitatively includes NO_3^--N. This is particularly true for experiments which involve the use of nitrogen -15 as a tracer or tag on N fertilizers. Numerous procedures designed for this purpose are present in the literature. However, direct implementation of such procedures are not always successful. Part I provides the description and testing of a procedure which was the end result of an effort to utilize a system developed elsewhere. In this experiment, wheat grain samples were used to test a recommended procedure and two similar procedures involving a pretreatment, digestion, steam distillation, and titration of the samples for total N determination. The procedure recommended provides satisfactory precision and accuracy in the total N analysis.

Part II describes experiments conducted in the field over two consecutive growing seasons utilizing nitrogen -15 fertilizers. The experiments were designed to provide information concerning the effect of tillage system, ^{15}N enrichment level of a labeled N fertilizer, and rate of the labeled N source on the distribution and uniformity of the

labeled N constituents in the fertilized winter wheat plants. Two tillage systems and four rates of fertilizer N were used. Plant samples were taken at physiological maturity and separated into six plant parts. All plant parts were analyzed for total N and ^{15}N content. Amounts of fertilizer N were then calculated for each plant part. Results of these experiments indicated a consistent linear response of each parameter by each plant part to increasing rates of fertilizer N. It was found that the treatments used in these experiments yielded plant parts that had a satisfactory uniformity of fertilizer -^{15}N among plant parts. These results are particularly important with respect to the use of the labeled crop residues in subsequent decomposition and residual fertilizer N studies which represent another facet of the overall research project from which these reported experiments are one part.

PART I

Digestion of Plant Materials for the Determination of Total Nitrogen to Include Nitrate

ABSTRACT

A digestion procedure is described which uses reduced iron and H_2SO_4 pretreatment for the quantitative recovery of NO_3^--N. The procedure utilizes an aluminum block digestor with a programmable control unit. The recommended program employs a stepwise temperature ramp up to 400°C, continues digestion 1 h after clearing for a total digestion time of 7.25 h. Two other digestion programs were compared with the recommended program. One program used a temperature ramp identical to the first but digested an additional 1 h after clearing for a total time of 8.25 h. A third digestion program used a shortened temperature ramp, attained a final temperature of 400°C, and was completed 1 h after clearing for a total time of 5.90 hr. Wheat grain samples with increasing amounts of NH_4^+-N and NO_3^--N up to 0.5 mg N were used to evaluate these procedures. The recommended procedure provides complete recovery of grain N and added N and superior levels of precision in comparison to the other digestion procedures evaluated. The digestion procedure using 5.90 h did not yield accurate analysis of grain N, indicating incomplete digestion. The 8.25 h digestion did not result in any improvement in grain N or added N recovery.

Additional index words: semimicro Kjeldahl, NO_3^--N recovery, tube digestion method, total N analysis, plant N analysis.

INTRODUCTION

A multitude of applications have been found for the standard Kjeldahl method for the determination of total N. Essential aspects of the Kjeldahl method of N analysis were reviewed by Kirk (1950). One of the limitations recognized with the standard Kjeldahl analysis is the inability to quantitatively recover NO_3^--N from the samples. Nitrate - N can be a significant fraction of the total N in soil and plant samples. Therefore, failure to accurately analyze for NO_3^--N can be a source of considerable error. In 1929, Olsen (1929) described a modification of the Kjeldahl procedure which had a pretreatment step to quantitatively include NO_2^--N and NO_3^--N. The pretreatment prior to digestion of the sample utilized acidified permanganate oxidation of NO_2^- to NO_3^-. This was followed by a reduction of NO_3^- to NH_4^+ by use of reduced iron (Fe) and H_2SO_4. The reduced Fe served as an electron donor and the H_2SO_4 provided an acidic medium conducive to the reduction of NO_3^--N to NH_4^+N. After pretreatment, the standard Kjeldahl digestion with H_2SO_4 was followed.

In 1958, Bremner and Shaw (1958) adopted the Olsen method for use in a denitrification study. Since then, Bremner has also described the procedure for total N analysis in soils which employs the modification outlined by Olsen as well as a procedure utilizing salicylic acid and thiosulfate for recovery of NO_2^--N and NO_3^--N (1965). Further evaluations and modifications of these procedures have continued to flourish. This in part is due to the increased interest in quantitative recovery of N species present in soil and plant materials when associated with ^{15}N tracer studies in soil-plant systems.

The use of the salicylic acid - thiosulfate method for inclusion of NO_3^--N is commonly referenced in the literature (Bremner, 1965; Bremner and Mulvaney, 1982). However, it is also recognized that inherent problems exist with this method due to the interference of water in a sample with the nitration reaction (Nelson and Sommers, 1980). Therefore, it is often felt that samples need to be moisture-free for the salicylic acid pretreatment to be successful. To effectively remove all water from a sample, a narrow range in temperatures for drying must be adhered to. The possibility of losing NH_4^+-, NO_2^--, and NO_3^--N exists when samples are dried at 105°C (Bremner, 1965). Several comparisons have been made between modifications of the Kjeldahl procedure to include NO_3^--N, often with conflicting results and conclusions (Goh, 1972; Nelson and Sommers, 1973; Peterson and Chester, 1964; Qashu, 1967). In 1972, Goh found that a semimicro modification of the Olsen method resulted in superior recovery of NO_3^--N compared to the commonly used salicylic acid - thiosulfate method.

In many laboratories, the use of aluminum digestion blocks in both standard and modified Kjeldahl procedures has become a common tool (Nelson and Sommers, 1972; Peterson and Chester, 1964). Many of the aluminum block digestion systems available have programmable capabilities to allow control over temperatures and times utilized in digestion procedures.

The purpose of the study described herein was to develop a semimicro modification of the Olsen method for the automated digestion of plant samples using an aluminum block apparatus with programmable

controls in a effort to obtain total N analysis to quantitatively include NO_3^--N. This digestion procedure is similar to those described by Gallaher et al. (1976) and Douglas et al. (1980). However, in this procedure the early stages of digestion at temperatures less than 200°C (Table 1) are prolonged to ensure removal of water and to prevent loss of samples due to frothing and boil over. In comparison, Gallaher's procedure for digestion calls for the heating of samples to 375°C for 2.5 h, then an additional 1.5 h after clearing (assuming a yellowish-green, transparent appearance). This approach, when used in our laboratory, resulted in difficulties which subsequently necessitated the modifications developed for the procedure described in this paper. Procedures developed to overcome difficulties encountered in other portions of the Kjeldahl analysis, such as distillation, are also described.

MATERIALS AND METHODS

Plant samples used in analysis were dried at 60°C and ground in a Wiley mill to pass a 1 - mm sieve. Subsamples of 100 mg were weighed out for analysis. The procedure described should be suitable for individual samples of 50 to 200 mg of ground plant material. The material used in the evaluation of this procedure was wheat grain containing from 20 to 30 mg g^{-1} total N.

EQUIPMENT

The digestion equipment used in this procedure was a Tecator DS-40 with a Tecator 1008 control unit[1]. The DS-40 is an aluminum block with a capacity of 40 75 ml tubes with constricted necks.

The steam distillation apparatus used was a semimicro system similar to that described by Bremner and Edwards (1965). The apparatus used here consisted of 100 ml distillation flasks held at approximately 45 degree angles. The steam production was provided by a Bellco quartz heating element contained in a 4 liter vessel. A flow of distilled-deionized water was regulated into the steam generator vessel to maintain a constant stream generation process. Steam was supplied to two distillation units from the steam generator via an insulated glass manifold. Distillate condensation columns were supplied with a refrigerated water system to enhance condensation. Waste steam passage was controlled by adjustable clamps placed on tubing that connected each end of the steam delivery manifold to the waste steam condensors. A Fisher auto-titration system was used to titrate distillates collected from each sample. The Fisher system used consisted of the following four components: a Model 380 electrometer, Model 383 titrate demand unit, Model 395 burette/dispenser, and a Model 385 titrate stirrer.

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[1]Trade names are provided for the benefit of the reader and do not imply endorsement by Oklahoma State University.

PROCEDURE

Weigh out a 50 to 200 mg sample of plant material. If samples are destined for isotope - ratio analysis for ^{15}N tracer studies, sample weights may be adjusted to provide a 1.0 to 1.2 mg N in the sample or the amount of N necessary for mass spectrometer specifications to provide minimum N_2 gas pressures. In a given rack of 40 tubes used in this procedure, two tubes should be treated as "blanks" in which all reagents are added but no actual samples are used. If duplicate analysis is desired, this system would then accommodate 19 different samples for analysis. To each sample, add 2 ml distilled-deionized water and 2 ml of 1:1 H_2SO_4. Allow samples to stand for 30 min. Add 2 drops of octyl alcohol and 0.5 g of reduced Fe. Allow the samples to stand for 30 to 40 min or until the effervescence ceases. Next, place the rack of tubes on a pre-heated digestion block at 95 to 100°C for about 45 min. Remove from block and allow samples to cool. Add approximately 1.1 g of the catalyst mixture (containing a 100:10:1 ratio of K_2SO_4, $CuSo_4$, and Se respectively), and add 4 ml of concentrated H_2SO_4. Place rack on digestion block and initiate program outlined in Table 2. Place small funnels with 3 mm diameter stems into the tops of the digestion tubes to encourage thorough refluxing of the samples within the tubes. The early steps of the digestion program are intentionally slow to efficiently drive off any water in the samples and to prevent boil-over problems. The samples should clear approximately six h into the digestion program when using Program No. 1 (Table 2). The additional 1 h after clearing at high temperature is to facilitate complete digestion. After the digestion, remove samples

from the block and allow to cool before transferring to distillation flasks.

A quantitative transfer of the digestion tube contents to the distillation flask is important. Transfers can be carried out by a series of rinses with distilled-deionized water and suspending the remaining contents in the tube by use of a vortex test-tube mixer. Complete transfers should be made with a total volume of approximately 20 ml of water.

The digested samples consist of concentrated acid salts which can react violently when mixed with strong base which is necessary for the distillation step in the Kjeldahl procedure. This reaction can result in the splattering of the contents of the distillation flask throughout the distillation unit or can break the bottom of the distillation flask which causes loss of the sample. This is particularly a problem in a distillation apparatus as designed for the introduction of base to the distillation flask contents via a glass tube that is also used for steam delivery. In this case mixing of these contents in the distillation unit is difficult due to the rigid structure. Therefore, it would be to some advantage to add some base to the distillation flask before placing it on the distillation unit. However, if too much base is added the pH of the solution becomes too high and a potential loss of NH_3 from the sample exists. A series of samples digested and transferred to distillation flasks by this procedure were titrated with 50% (w/w) NaOH. The resultant pH of the solutions after addition of 6 ml NaOH was approximately pH 3 (Figure 1). Considering equilibrium properties concerning the liberation of NH_3 from a solution containing

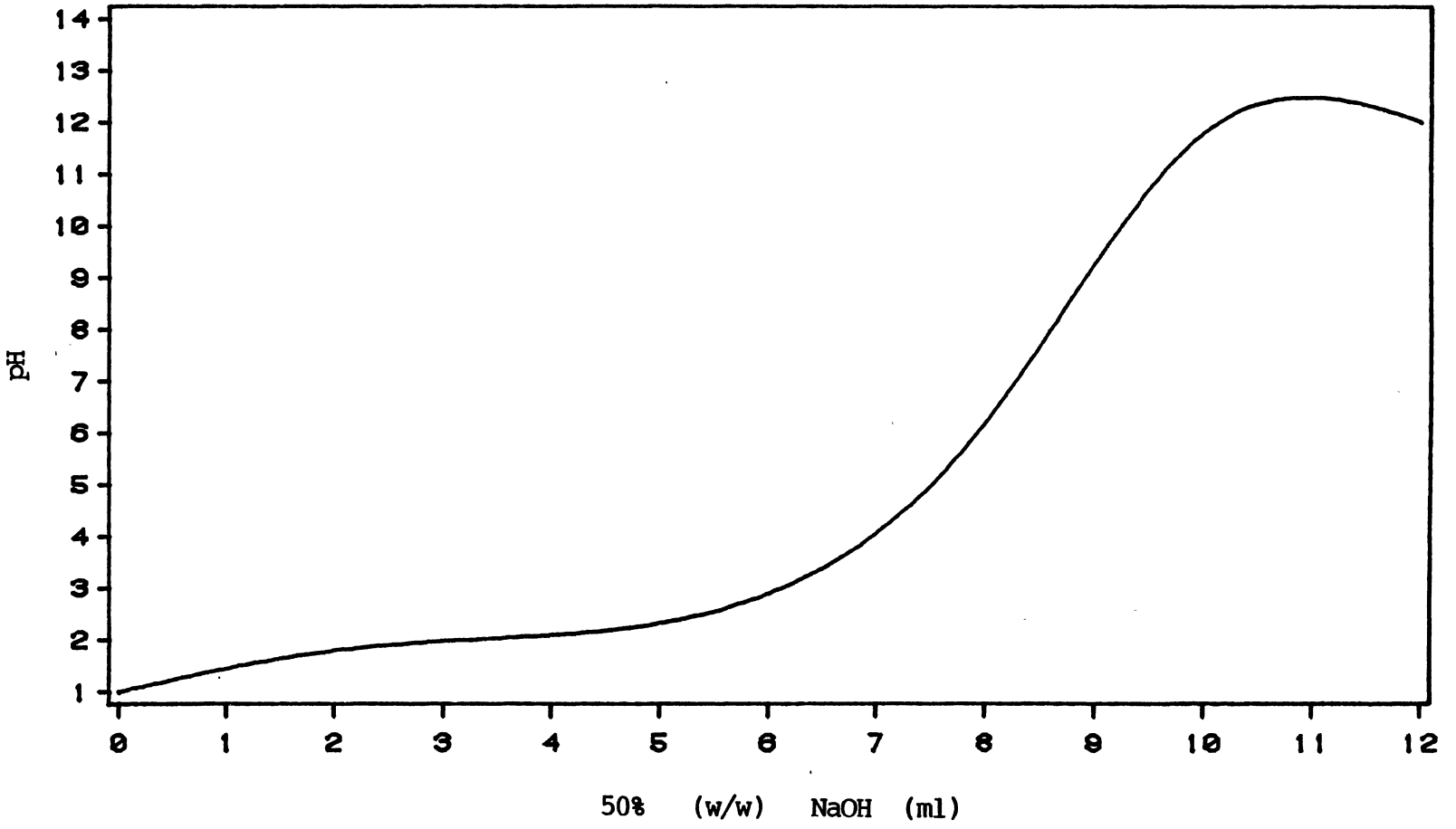

Fig. 1. Titration of sample digests with 50% NaOH

NH_4^+-N as $(NH_4)_2SO_4$ and assuming a total N content of 1.0 to 2.0 mg N, this pH range of 3 to 6 should not pose a problem of NH_3 loss. Therefore in this procedure, an addition of 6 ml of 50% NaOH is made to each sample while gently swirling the contents before placing the flask on the distillation unit. After placement of the flask on the distillation unit, an additional 6 to 8 mls of 50% NaOH can easily be added without encountering violent reactions. The total quantity of base added then provides a high enough pH to facilitate release of NH_3-N during the distillation process. Distillation of the samples was conducted with a distillate flow rate of approximately 6 ml min^{-1}. Samples were distilled into at least 2 ml of 2% H_3BO_3 (adjusted to pH 5). When distillate levels reached 35 ml the steam was shut off, the condensor tip washed, and the sample distillate was removed. The distillation flask was removed, emptied, and placed back on the distillation unit. A 30 ml aliquot of dilute HCl was then added via the funnel port to the steam delivery tube of the distillation unit and the unit was steamed with this solution for 6-to 8 min. This provides a cleansing step for the apparatus that serves to remove the $Fe(OH)_3$ residues formed after the NaOH addition to the sample during distillation. After the HCl washing, the distillation flask was again removed, emptied and replaced on the distillation unit. A 35 ml aliquot of distilled-deionized water was added in a similar fashion as the HCl wash to facilitate complete cleaning of the apparatus. The solution was then steamed for 6-to 8 min before removal. This last washing step with distilled-deionized water was repeated a second time in an identical manner. After these cleansing steps, the distillation

unit was steamed out for 90 to 120 sec before beginning the next sample distillation. The cleaning steps remove deposits left by the sample and are also intended for prevention of cross contamination of N contents between samples. This is particularly important if samples processed in this manner are part of an ^{15}N tracer study.

Actual quantities of N present in each sample distillate were then determined by titration to pH 4.8 with 0.01 $\underline{N}$ H_2SO_4. All titrations were carried out by use of the Fisher auto-titration system previously described.

EXPERIMENTAL

A series of experiments were conducted to evaluate the effectiveness of NO_3^--N recovery of the digestion procedure described. A large sample of wheat grain was used for each experiment. Thirty-six 100 mg subsamples were weighed out and each placed in a clean, dry digestion tube. Four blanks that contained reagents but no wheat grain were included in each digestion rack containing 40 tubes. The remaining 36 grain samples consisted of nine groups, each of which included four increments of N as outlined in Table 1.

All treatments outlined in Table 1 were applied to the dry samples in the digestion tubes as 5 ml aliquots of appropriate solutions. To samples receiving treatment 1, a 5 ml aliquot of distilled-deionized water was added. Treatment 2 consisted of 5 ml additions of a 100 μ g ml^{-1} solution of NH_4^+-N prepared by placing 0.4714 g of reagent grade $(NH_4)_2SO_4$ in a 1 liter volumetric flask and bringing it to volume with distilled-deionized water. Treatment 3 was prepared in a similar manner by using 0.7214 g of reagent grade KNO_3 per liter resulting in a

TABLE 1. TREATMENTS USED IN THE N RECOVERY EXPERIMENTS.

Treatment	N added	N form	Aliquot size
	---mg---		---- ml ----
1	0		5*
2	0.5	NH_4^+	5
3	0.5	NO_3^-	5
4	0.5	$NH_4^+ + NO_3^-$	5

* 5 ml aliquot of distilled-deionized water.

100 μg ml^{-1} NO_3^--N solution. Treatment 3 was then applied as a 5 ml aliquot of this solution. Treatment 4 was prepared by using 0.2357 g $(NH_4)_2SO_4$ and 0.3607 g KNO_3 per liter to give a 50 μg ml^{-1} NH_4^+-N + 50μ g ml^{-1} NO_3^--N. A 5 ml aliquot of this solution was used to give a 0.5 mg total N addition to the sample as a combination of these two mineral forms of N.

The digestion procedure outlined in Table 2 and referred to as Program No. 1 was designed to provide a complete, stepwise temperature ramp to a final temperature of 400°C and a continuation of digestion 1 h after clearing of the samples (Fig. 2). Besides the procedure shown in Table 2, two other digestion formats were tested. Since other published procedures are apparently capable of accomplishing successful digestion and recovery of total N by use of shorter digestion periods than that of Table 1, a shortened digestion procedure was tested. This shortened version referred to as Program No. 2 and outlined in Table 3, utilized a faster temperature ramp in reaching a final temperature of 400°C (Fig. 3). This program then continued for 1 h after clearing of the samples and terminated with a total digestion time of 5.90 h. A third digestion program (Program No. 3) that was tested consisted of an identical temperature ramp as that in Program No. 1. However, this program was continued an additional 1 h past that of Program No. 1 to provide for prolonged digestion of the samples after clearing and a total time of 8.25 h. This digestion program was designed as a comparison to Program No. 1 to evaluate complete digestion of the grain N. The same wheat grain sample was used throughout all experiments testing these three digestion formats with the same treatments shown in

TABLE 2. PROGRAM NO. 1 FOR TEMPERATURE AND TIME CONTROL ON AN ALUMINUM DIGESTION BLOCK.

Program Step	Warm-up Time	Hold Time	Step Time	Total Program Time	Temperature
	---------- min ----------			----h:min---	----°C ----
1	25	20	45	0:45	100
2	15	30	45	1:30	150
3	7	60	67	2:37	175
4	7	30	37	3:14	200
5	25	35	60	4:14	250
6	25	35	60	5:15	300
7	25	35	60	6:15	350
8	25	35	60	7:15	400

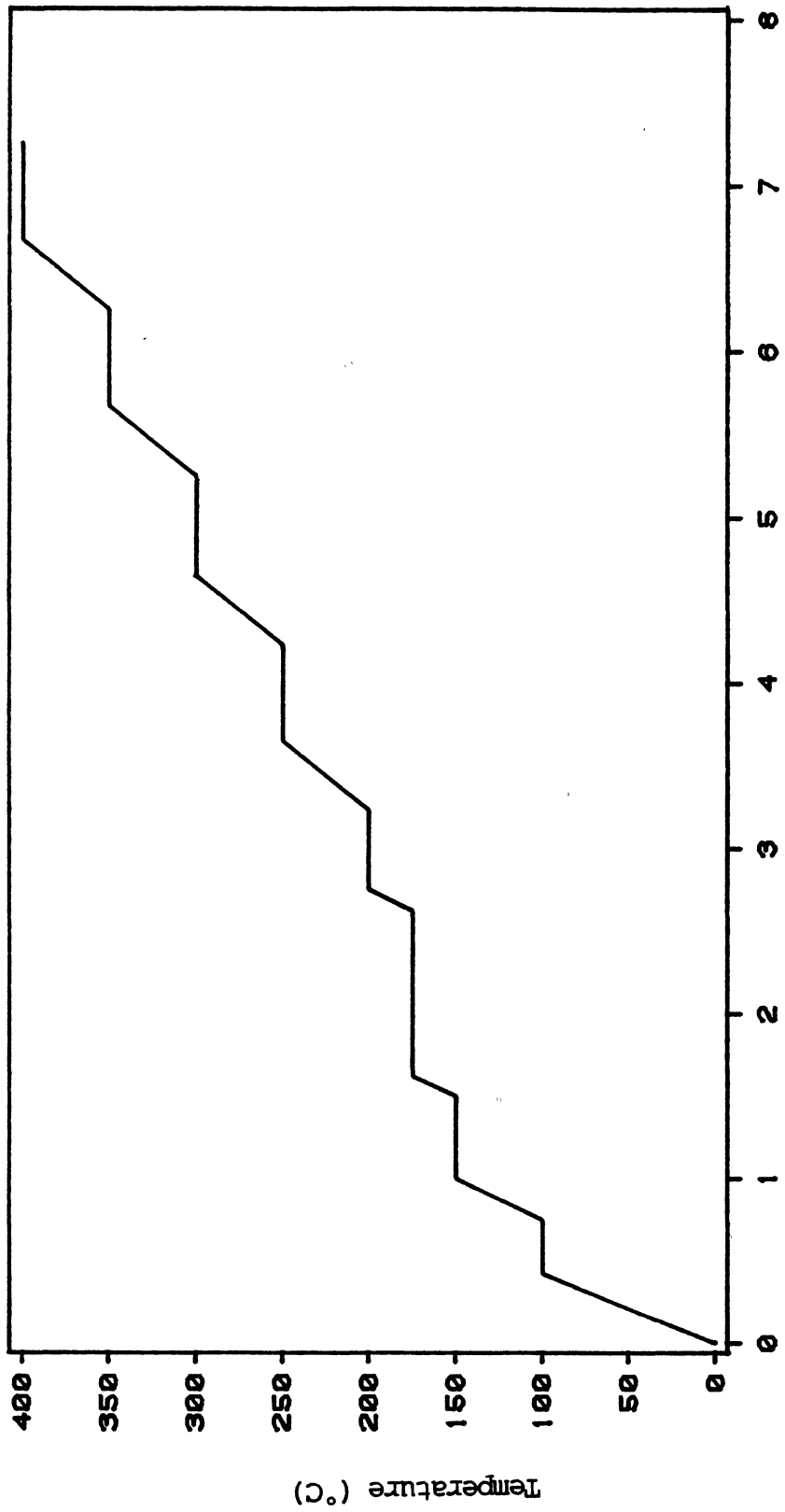

Fig. 2. Temperature ramp for Program No. 1

TABLE 3. PROGRAM NO. 2 FOR TEMPERATURE AND TIME CONTROL ON AN ALUMINUM DIGESTION BLOCK.

Program Step	Warm-up Time	Hold Time	Step Time	Total Program Time	Temperature
	---------- min ----------			----h:min---	----°C ----
1	25	20	45	0:45	100
2	15	30	45	1:30	150
3	7	60	67	2:37	175
4	7	30	37	3:14	200
5	40	120	160	5:54	400

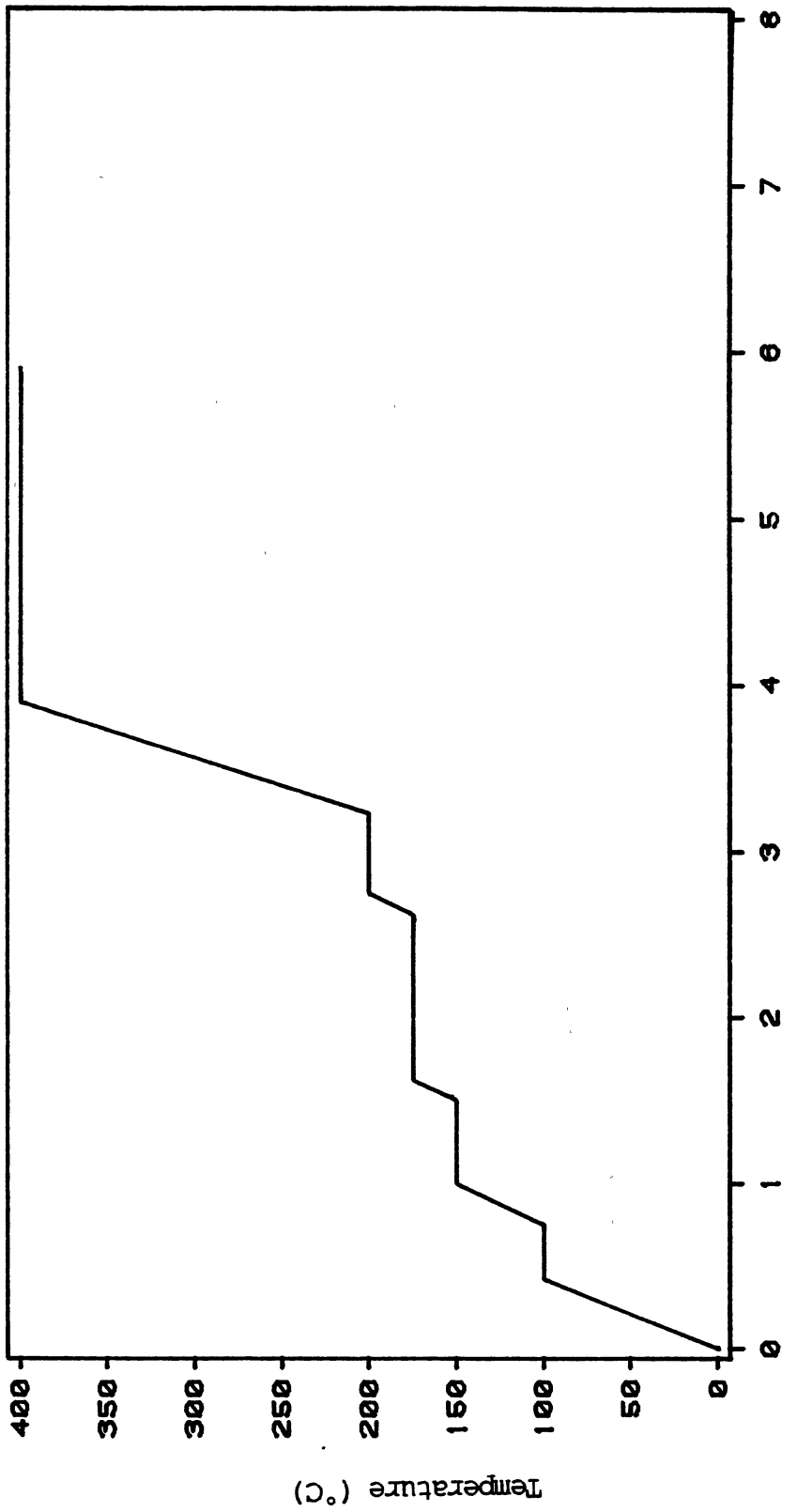

Fig. 3. Temperature ramp for Program No. 2

Table 1. For each of the three programs tested two racks of 40 tubes were digested and analyzed. This consisted then of 18 replications of each group of four treatments for each program tested.

To further evaluate the procedure of digestion shown in Table 2 (Program No. 1), a series of two treatment experiments was conducted. The same grain sample was again used and treatments consisted of treatments number one and three outlined in Table 1. Using 40 digestion tubes per digestion rack and two blank samples, provided 19 replications of treatments number one and two in each rack. Five racks of samples were treated in this manner, digested in the format of Program No. 1 and analyzed via distillation and titrations of all samples as previously described.

Grain samples were used since the content of NO_3^--N was expected to be negligible and the result of the reduction step should be apparent. All groups of samples were then pre-treated and digested according to the procedure described. The digested samples were then analyzed by completing the final two steps of Kjeldahl analysis: steam distillation and titration.

Experimental data were analyzed statistically using analysis of variance procedures and an LSD multiple comparison test outlined by the SAS Institute (1982).

RESULTS AND CONCLUSIONS

The use of the modified Olsen procedure for total N analysis previously described was evaluated through the use of several different digestion procedures. Faster digestion procedures than that outlined in Table 2 (Program No. 1), such as that of Gallaher et al. (1976), and

Douglas et al. (1980), resulted in various difficulties. Problems of excessive frothing and boil-over as well as poor recoveries were encountered with faster digestions. The results from a series of experiments consisting of the treatments outlined in Table 1 and digested according to Procedure No. 1 of Table 2 are presented in Table 4. For Program No. 2 an identical series of treatments were applied to the common wheat grain sample having a digestion procedure similar to that of Program No. 1, but having a terminal step at No. 5 going from 200°C directly to 400°C then holding at this temperature until clearing occurred and then continuing an additional h before completing the digestion. This shortened version resulted in a total program time of 1 h and 20 min (1:20) less than Program No. 1 (Table 3). Nitrogen analysis results are shown in Table 5 for the shorter digestion program (Program No. 2). Considering the CV values as a measure of precision in the procedure, it is apparent that treatment Nos. 2 and 3 in Program No. 2 had much closer agreement between samples (Table 5) than those same treatments following a longer digestion as in Program No. 1 (Table 4). However, the samples from treatments No. 1 (no added N) exhibited much higher variation between samples for the Program No. 2 compared to those from the longer digestion of Program No. 1. This was attributed to incomplete digestion of materials containing organic N compounds (such as N in cyclic ring structures) in the shortened program of Program No. 2. However the procedure obviously allowed for complete recovery of the added mineral N treatments (treatments 2 to 4) under Program No. 2. In review of the results obtained from a version of the digestion procedure of Table 2 in which the final step was prolonged an

TABLE 4. TOTAL N ANALYSIS OF GRAIN SAMPLES AND RECOVERY OF N TREATMENTS WHEN PROGRAM NO. 1 WAS USED.

Treatment	N added	mean	sd	cv	Recovery
		------ mg N ------		-------- % --------	
1	0	2.83a*	±0.07	2.49	
2	0.5 mg NH_4^+ - N	3.34b	±0.14	4.28	100.3
3	0.5 mg NO_3^- - N	3.37b	±0.11	3.16	101.2
4	0.5 mg (NH_4^+ + NO_3^-)-N	3.38b	±0.11	3.30	101.5
LSD (0.05)		0.11			

*Treatments followed by the same letter are not significantly different at the 0.05 probability level (LSD multiple comparison test).

TABLE 5. TOTAL N ANALYSIS OF GRAIN SAMPLES AND RECOVERY N TREATMENTS WHEN PROGRAM NO. 2 WAS USED.

Treatment	N added	mean	sd	cv	Recovery
		------ mg N ------		-------- % --------	
1	0	2.30a*	±0.24	10.55	
2	0.5 mg NH_4^+ - N	2.96b	±0.06	1.99	105.7
3	0.5 mg NO_3^- - N	2.95b	±0.05	1.81	105.4
4	0.5 mg (NH_4^+ + NO_3^-)-N	2.97b	±0.04	1.50	106.1
LSD (0.05)		0.14			

*Treatments followed by the same letter are not significantly different at the 0.05 probability level (LSD multiple comparison test).

additional h (8:15 total time referred to as Program No. 3) illustrated in Table 6, it was concluded that additional digestion past 7:15 total time does not yield improved efficiency in N recovery to warrant extended digestions.

The difference in the actual digestion times between Program No. 1 and Program No. 2 is small (1:20). However, the analytical differences are considered to be significant in terms of the objectives of utilizing such a procedure. Therefore, based upon the improved ability to recover the N forms that are apparently more resistant to digestion in materials such as wheat grain, the digestion program of No. 1 was chosen as a superior digestion format.

The information in Table 7 is from the results of the two treatment digestion experiments utilizing treatments number one and three listed in Table 1 and Program No. 1 of Table 2. These analyses verify the ability of the digestion procedure of Program No. 1 to render accurate determinations of total N including NO_3^--N in plant materials such as wheat grain.

These results illustrate not only the importance and effectiveness of the modified Olsen procedure and digestion of Program No. 1 to recover NO_3^--N but also the importance in evaluation of the digestion step itself in the use of a modified Kjeldahl procedure.

TABLE 6. TOTAL N ANALYSIS OF GRAIN SAMPLES AND RECOVERY N TREATMENTS WHEN PROGRAM NO. 3 WAS USED.

Treatment	N added	mean	sd	cv	Recovery
		------ mg N ------		-------- % --------	
1	0	2.84a*	±0.21	7.52	
2	0.5 mg NH_4^+ - N	3.45b	±0.06	1.69	103.3
3	0.5 mg NO_3^- - N	3.43b	±0.07	2.05	102.7
4	0.5 mg (NH_4^+ + NO_3^-)-N	3.47b	±0.05	1.50	103.9
LSD (0.05)		0.11			

*Treatments followed by the same letter are not significantly different at the 0.05 probability level (LSD multiple comparison test).

TABLE 7. RESULTS OF TOTAL N RECOVERY EXPERIMENTS FROM WHEAT GRAIN USING TWO TREATMENTS AND PROGRAM NO. 1.

Group	N added	mean	sd	cv	Recovery
		------ mg N ------		-------- % --------	
1	0	2.84	±0.24	8.52	
1	0.5 mg NO_3^- - N	3.42	±0.11	3.21	102.4
2	0	2.83	±0.10	3.37	
2	0.5 mg NO_3^- - N	3.34	±0.08	2.49	100.3
3	0	2.81	±0.10	3.58	
3	0.5 mg NO_3^- - N	3.33	±0.08	2.32	100.6
4	0	2.77	±0.09	3.16	
4	0.5 mg NO_3^- - N	3.27	±0.09	2.68	100.0
5	0	2.81	±0.09	3.36	
5	0.5 mg NO_3^- - N	3.32	±0.08	2.57	100.3
overall	0	2.81	±0.14	4.84	
overall	0.5 mg NO_3^- - N	3.34	±0.10	2.99	100.9

REFERENCES

Bremner, J.M. 1965. Total nitrogen. In C.A. Black (ed.) Methods of soil analysis, part 2. Agronomy 9:1149-1178. Am. Soc. Agron. Madison, Wis.

Bremner, J.M., and A.P. Edwards. 1965. Determination and isotope-ratio analysis of different forms of nitrogen in soils: Apparatus and procedure for distillation and determination of ammonium. Soil Sci. Soc. Am. Proc. 29:504-507.

Bremner, J.M., and C.S. Mulvaney. 1982. Total nitrogen. In A.L. Page (ed.) Methods of soil analysis, part 2, 2nd ed., Agronomy 9:619.

Bremner, J.M., and K. Shaw. 1958. Denitrification in soil. I. Methods of investigation. J. Agric. Sci. 51:22-39.

Douglas, L.A., A. Riazi, and C.J. Smith. 1980. A semi-micro method for determining total nitrogen in soils and plant material containing nitrite and nitrate. Soil Sci. Soc. Am. J. 44:431-433.

Gallaher, R.N., C.O. Weldon, and F.C. Boswell. 1976. A semiautomated procedure for total nitrogen in plant and soil samples. Soil Sci. Soc. Am. J. 40:887-889.

Goh, K.M. 1972. Comparison and evaluation of methods for including nitrate in the total nitrogen determination of soils. J. Sci. Food Agric. 23:275-284.

Kirk, P.L. 1950. Kjeldahl method for total nitrogen. Anal. Chem. 22:354-358.

Nelson, D.W., and L.E. Sommers. 1972. A simple digestion procedure for estimation of total nitrogen in soils and sediments. J. Environ. Qual. 1:423-425.

Nelson, D.W., and L.E. Sommers. 1973. Determination of total nitrogen in plant material. Agron. J. 65:109-112.

Nelson, D.W., and L.E. Sommers. 1980. Total nitrogen analysis for soil and plant tissues. J. Assoc. Off. Anal. Chem. 63:770-778.

Olsen, C. 1929. On determination of nitrogen in soils. With special reference to presence of nitrates and nitrites. C.R. Trav. Lab. Carlsberg Vol. 17, no. 3.

Peterson, L.A., and G. Chester. 1964. A reliable total nitrogen determination on plant tissue accumulating nitrate nitrogen. Agron. J. 56:89-90.

Qashu, H.K. 1967. A micro-digestion procedure for the determination of aminoid nitrogen in plant material. Agron. J. 59:486.

Statistical Analysis Systems Institute. 1982. The ANOVA procedure p. 119-138. In H.A. Ray (ed.) SAS user's guide: Statistics. Statistical Analysis Systems Institute, Inc., Raleigh, NC.

PART II

Distribution of Fertilizer Nitrogen -15 in Winter Wheat Under Clean and Reduced Tillage Systems

ABSTRACT

A field experiment utilizing nitrogen -15 labeled fertilizer was initiated in the fall of 1982 at the Agronomy Research Station at Stillwater, Oklahoma using winter wheat (Triticum aestivum L.) as a test crop. Objectives of this portion of the research project were (1) to determine fertilizer -^{15}N content in wheat plants due to N rate, enrichment level of the labeled fertilizer, and tillage system, and (2) to determine the uniformity of ^{15}N content among different plant parts of winter wheat due to fertilizer -^{15}N and tillage treatments. The experiment was conducted during two consecutive growing seasons (1982-83 and 1983-84). Treatments included two tillage systems (clean and reduced), and four rates of fertilizer N (0, 56, 112, and 168 kg ha^{-1}). Fertilizers were applied post-emergence using $^{15}NH_4NO_3$ as the labeled N source with 7.2 and 1.2 atom % ^{15}N respectively in 1982 and 1983. Plant samples were taken at physiological maturity and separated into the following parts: roots, crowns, leaves, stems, grain, and chaff. All plant parts were analyzed for total N (TN) and ^{15}N content. Fertilizer N (FN) content was subsequently calculated from the analytical results. Tillage treatments were not significantly different ($P < 0.05$) for any of the variables tested, either year. Excess atom % ^{15}N data for the 1983 samples indicated significant differences among several plant parts within some rates of fertilizer N. No differences were found among above ground plant parts at any

rate of fertilizer N. In 1984 excess atom % ^{15}N values were not significantly different among plant parts at any rate of fertilizer N. Both TN and FN concentrations in plant parts followed similar trends to each other in 1983 and 1984. Grain contained consistent significantly higher concentrations of TN and FN often followed by two groups of plant parts in the following order: grain > leaves, chaff > crowns, stems, and roots.

Analysis of the ratios of FN:TN revealed no differences among plant parts in 1984 and only slight differences in 1983. All plant parts exhibited a significant linear response to increasing N rates. The use of above ground components of winter wheat plants labeled with ^{15}N from treatments used in these experiments appear to be satisfactory for use in subsequent decomposition and mineralization studies.

Additional Index Words: Excess Atom % ^{15}N, Triticum aestivum L., N distribution, plant parts, total N, fertilizer N.

INTRODUCTION

The processes of nitrogen (N) cycling in the soil are directly related to the management of crop residues. The mineralization of soil N is an important process which includes the mineralization of N contributed from decomposing crop residues. Therefore, it is important to understand the release of N from crop residues as a function of decomposition and mineralization in order to effectively manage N inputs into a given agricultural system. This requires specific knowledge and information concerning the unique circumstances involving the environmental and soil-plant system under study.

The use of various residue management systems other than conventional clean-tillage approaches are becoming increasingly more prevalent (Magleby et al., 1985). Different residue management systems may impart changes in physical, chemical, and biological properties of a soil-plant system. For example, higher bulk densities and penetrometer resistances have been reported for corn belt soils under no-tillage (NT) compared to similar soils under conventional-clean tillage (CT) (Lindstrom et al., 1984). However, Hill and Cruse (1985) reported recently that there were no significant differences in soil bulk density values among CT, reduced-tillage (RT), or NT tillage systems. Denitrification has also been reported for soils under NT management (Doran, 1980; Rice and Smith, 1982). Also leaching of N may increase under NT (McMahon and Thomas, 1976). Mineralization of N in CT systems has been shown to be greater than in NT systems (Dowdell and Cannell, 1975; Powlson, 1980; and House et al., 1984). Wilson and Hargrove (1986) have reported a faster rate of N release in residues of

crimson clover under CT than NT conditions.

In the comparison of any soil properties between different residue management systems it is important to consider the time or duration of the tillage systems under study. This point was well illustrated in the evaluation of a 16 yr study comparing CT and NT systems of corn production in Kentucky (Rice, Smith, and Blevins, 1986). In this study the mineralization rates of soil N between CT and NT differed initially. After 10 to 16 cropping years under these residue management systems, soil N mineralization rates apparently reached a steady state and no differences were observed between mineralization rates under CT and NT treatments.

One means of monitoring the fate of fertilizer N incorporated into crop residues is by use of labeled N fertilizer with ^{15}N. This technique has been used to some extent in previous experiments with winter wheat (Triticum aestivum L.). Myers and Paul (1971) used ^{15}N - labeled oat residues with 0.514 atom % ^{15}N in a field experiment with winter wheat being grown as a crop. This level of ^{15}N enrichment was found to be satisfactory for measuring the release of the N in the residues through decomposition and mineralization over two cropping seasons. This enabled Myers and Paul (1971) to determine that 11% of the N in the oat residues was taken up by the winter wheat. Fredrickson et al. (1982) utilized spring wheat straw with 5.29 atom % ^{15}N and 1.2% total N applied to a winter wheat crop to measure the availability of the straw N. From the use of the labeled residues, Fredrickson et al. (1982) found that approximately 9% of the N in the spring wheat straw was taken up by the subsequent winter wheat crop in

one cropping year.

The use of ^{15}N labeled crop residues to evaluate the fate of residual fertilizer N usually implies recognition of several underlying assumptions and considerations as outlined by Hauck and Bremner (1976). One of these assumptions considers the ^{15}N from labeled fertilizer N sources incorporated in the crop plant to be uniform among all parts of the plant. Wagger et al. (1985a) recognized the need to verify this assumption on the basis of producing labeled crop residues to study decomposition and mineralization of the N constituents. They considered three points fundamental to the aim of producing labeled crop residues containing satisfactory uniformity and levels of enrichment of ^{15}N among all plant parts. These points were (a) the level of ^{15}N enrichment of the fertilizer N to be used, (b) the rate of N to be applied as fertilizer to the crop, and (c) the stage of growth at which the crop should be fertilized. Wagger et al. (1985a) utilized a rather low rate of fertilizer N application (13.3 kg N ha^{-1}) with a high level of ^{15}N enrichment (84.5 atom % ^{15}N) which was applied at one stage of growth (jointing developmental stage) to provide satisfactory labeling of the resultant wheat plant residues which contained 7.0 to 8.2 atom % ^{15}N. In later experiments, Wagger et al. (1985b) found these residues to be adequate for use in the measurement of mineralization of the residual N present in the ^{15}N - labeled wheat residues for two crop seasons.

Apparently there is a limited amount of information available concerning the use of ^{15}N - labeled fertilizers with less than 10.0 atom % ^{15}N and the effect of varying rates of ^{15}N - labeled fertilizer

N on the distribution and level of ^{15}N enrichment on crop residues fertilized with ^{15}N - labeled fertilizers. This information may be of particular interest in field experiments which require larger amounts of ^{15}N - labeled crop residues to study the mineralization of residual N. If found to provide adequate uniformity and enrichment levels of ^{15}N among plants parts, use of ^{15}N labeled fertilizers with lower levels of ^{15}N enrichment could provide considerable savings of research resources.

The present study was part of a larger experimental project designed to investigate several aspects of fertilizer N use by winter wheat under different tillage systems and rates of fertilizer N. The objectives were (1) to determine fertilizer - ^{15}N content in wheat plants due to N rate and tillage system treatments, and (2) to determine the uniformity of ^{15}N content among different wheat plant parts as effected by N rate and tillage system.

MATERIALS AND METHODS

A field experiment was established in the fall of 1982 on Norge loam (fine-silty, mixed, thermic Udic Paleustoll) at the Agronomy Research Station at Stillwater, Oklahoma. Initial soil pH, NO_3^--N, P, and K indices were determined by use of 1:1 H_2O, specific ion electrode, Bray and Kurtz (1945) no. 1 extract (1:20 dilution), and 1M neutral NH_4OAc (Knudsen et al., 1982) respectively. Initial soil pH was 5.6. Initial NO_3^--N, P, and K indices were found to be 30, 81, and 545 kg ha^{-1} respectively. It was determined that the soil contained sufficient P and K based on Oklahoma State University soil test calibrations (Johnson and Tucker, 1982) and application of P and K was not necessary.

The experiment was conducted during two consecutive growing seasons (1982-83 and 1983-84). Two tillage systems (clean and reduced), and four rates of fertilizer N (0, 56, 112, and 168 kg N ha^{-1}) were used in the study. Clean-till (CT) plots were double-disked while reduced-till (RT) plots were tilled with a V-blade twice to a depth of 12 cm which only partially incorporated crop residues. Weed control was carried out on an as needed basis by the application of paraquat (1, 1, - dimethyl - 4, 4, bipyridinium ion), bromoxynil plus (3, 5 - dibromo - 4 - hydroxybenzonitrile and [(4 - chloro - otolyl) oxy] acetic acid), and glyphosate [N - (phosphonomethyl) glycine] at recommended rates.

A split-plot design was employed with four replications. Tillage treatments were the main units and N rates were the subunits. Tillage main plots were each 12 by 33 m. Individual N plots were each 6 by 6

m. Each N plot was divided into nine 2 by 2 m subplots. The center-most subplot was further divided into four 1 by 1 m sub-subplots. Each of the sub-subplot areas were then further sub-divided into four 0.5 by 0.5 m microplots to help insure the uniform application of ^{15}N-enriched fertilizer. Appropriate amounts of the ^{15}N fertilizer were dissolved in 500 ml of distilled-deionized water and sprayed uniformly over each microplot. In the fall of 1982, four microplots were fertilized within each N plot in this manner and in the fall of 1983 another four microplots were fertilized within each N plot.

In each N plot, only the center-most subplot area received ^{15}N-enriched N fertilizer while the remainder of the plot received appropriate rates of N as the same N source without ^{15}N-enrichment. Fertilizers were applied after a uniform stand of wheat had been established to prevent physical movement of the fertilizer outside of the designated plots. The labelled N fertilizer used was $^{15}NH_4NO_3$ with 7.2 and 1.2 atom % ^{15}N respectively in 1982 and 1983.

All plots were planted with 100 kg ha^{-1} certified seed of the semidwarf cultivar TAM W-101 in 25.4 cm rows by use of a Tye-drill Pasturepleaser[1] adapted with rolling coulters and double disk openers capable of seeding in residues. With this spacing, two rows crossed each microplot.

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[1]Trade names are provided for the benefit of the reader and do not imply endorsement by Oklahoma State University.

The wheat plants were harvested from microplots at physiological maturity (Feekes stage 11.5 as illustrated by Large, 1954). Entire plants were removed at harvest including the adhering large roots. All roots and crowns were lightly washed at sampling to remove any adhering soil and the soil plus water solution was placed back into each microplot. Plant samples were then separated into the following parts: roots, crowns, leaves (plus sheaths), stems, grain, and chaff. Plant part samples were dried at 60°C for 24 hr, then ground to pass a 1-mm sieve. Precautions were taken to prevent cross-contamination between each sample.

Total N in plant part samples was determined in duplicate by use of a modified semimicro - Kjeldahl method to include nitrate (Douglas et al., 1980; Bremner, 1965). To minimize cross contamination of ^{15}N between samples the distillation apparatus was rinsed in succession with dilute HCl and twice with double-distilled, deionized water, then finally steamed after each sample. After titration with 0.01 $\underline{N}$ H_2SO_4, distillates were acidulated with 0.2 ml of 0.8 $\underline{N}$ H_2SO_4, and evaporated to dryness. The N isotope ratio analyses were determined with a VG Micromass 602D by the method of Porter and O'Dean (1977). Amounts of fertilizer N were calculated by use of the Eq. [1].

$$X = \frac{TN\ (C - B)}{A} \qquad \text{Eq. [1]}$$

where: X = amount of fertilizer N present (mg g^{-1})

TN = total N in sample (mg g^{-1})

A = excess atom % ^{15}N in the fertilizer

B = atom % ^{15}N in the standard

C = atom % ^{15}N in the sample

All data were subjected to analysis of variance using procedures outlined by the SAS institute (SAS Institute, 1985) and Steel and Torrie (1980). Analysis of variance was performed using a split-split plot design. In this manner tillage treatments were considered as whole units, N rates as subunits, and plant parts as sub-subunits.

RESULTS AND DISCUSSION

Information from analysis of variance concerning the main effects due to tillage system, N rate, and plant parts, as well as interaction terms, is shown in Table 1. The response variables analyzed and presented include excess atom % ^{15}N, total N (mg g^{-1}), fertilizer N (mg g^{-1}), and the ratios of fertilizer N / total N present. The response of each of these variables to the increasing fertilizer N rates used was statistically significant in both years (Table 1). The pattern of response associated with each variable to the rates of fertilizer N averaged over replications, tillages, and plant parts is shown in Fig. 1 and Fig. 2 for the 1982 to 1983 and the 1983 to 1984 cropping years respectively with the pertinent linear regression equations shown accordingly in Table 2. Main effects due to tillage system were not significant for any of these variables either cropping year (Table 1).

Excess Atom % ^{15}N in Plant Parts

There was a significant three-way interaction among tillage system, N rate, and plant parts with respect to excess atom % ^{15}N content. Comparisons of excess atom % ^{15}N values among different plant parts within rates of fertilizer-^{15}N by tillage system for 1983, are shown in Table 3. Under clean-tillage there were no significant differences among plant parts at either the 0 or the 56 kg N ha^{-1} rate of fertilizer N. Leaves were significantly higher in excess atom % ^{15}N than the roots at the 112 and 168 kg N ha^{-1} rates. Crowns were significantly higher in excess atom % ^{15}N than the roots at the 168 kg

TABLE 1. ANALYSIS OF VARIANCE FOR TILLAGE SYSTEM, N RATE, AND PLANT PART EFFECT ON EXCESS ATOM % ^{15}N, TOTAL N, FERTILIZER N, AND RATIOS OF FERTILIZER N : TOTAL N IN 1983 AND 1984.

		1983		1984	
Source	df.	Ex. at. % ^{15}N	Total N	Ex. at. % ^{15}N	Total N
Tillage	1	NS	NS	NS	NS
N Rate	3	***	***	***	***
Tillage X N Rate	3	NS	NS	NS	*
Plant Part	5	***	***	NS	***
Tillage X Plant Part	5	NS	NS	NS	NS
N Rate X Plant Part	15	**	***	NS	*
Tillage X N Rate X Plant Part	15	**	NS	NS	NS

*,**,*** Indicates significance at 0.05, 0.01, and 0.001 probability levels respectively;
NS = nonsignificant.

TABLE 1. (continued)

Source	1983		1984	
	Fertilizer N	Fert. N : Total N	Fertilizer N	Fert. N : Total N
Tillage	NS	NS	NS	NS
N Rate	***	***	***	***
Tillage X N Rate	NS	NS	NS	NS
Plant Part	***	**	***	NS
Tillage X Plant Part	NS	NS	*	NS
N Rate X Plant Part	***	**	***	NS
Tillage X N Rate X Plant Part	NS	***	NS	NS

*,**,*** Indicates significance at 0.05, 0.01, and 0.001 probability levels respectively; NS = nonsignificant.

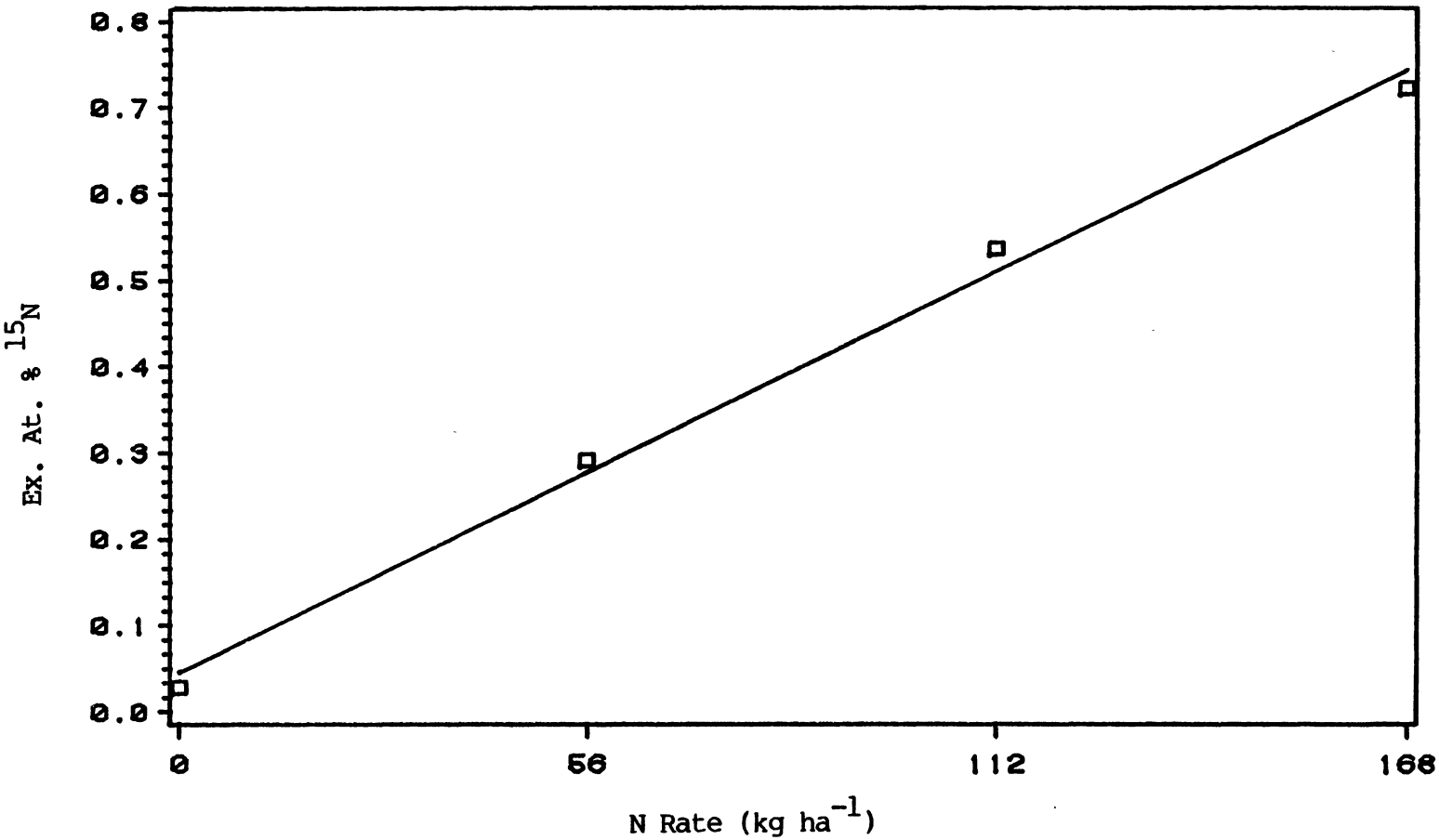

Fig. 1a. Means of ex. at. % ^{15}N by N rate in 1983 averaged over reps, tillages, and plant parts

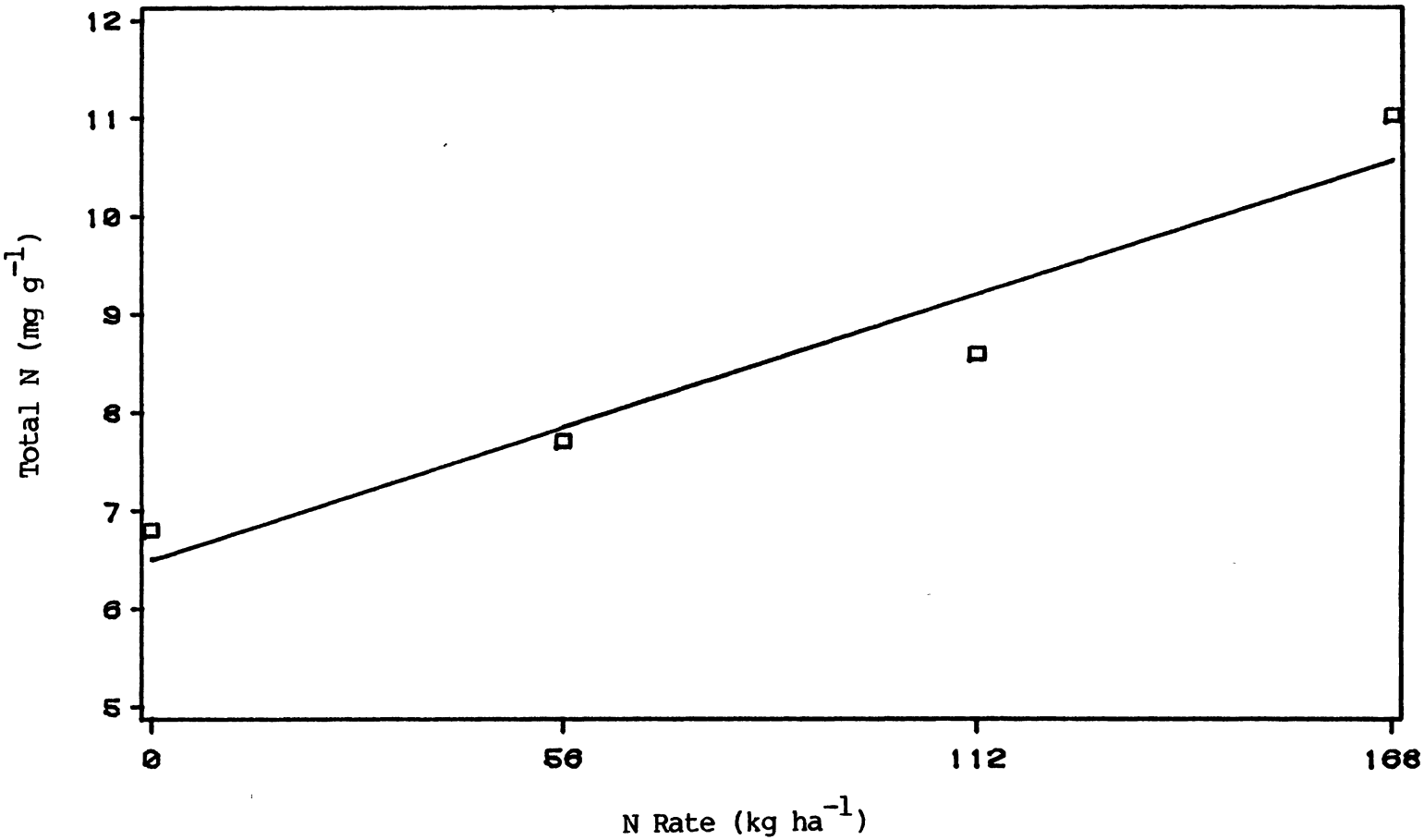

Fig. 1b. Means of total N by N rate in 1983 averaged over reps, tillages, and plant parts

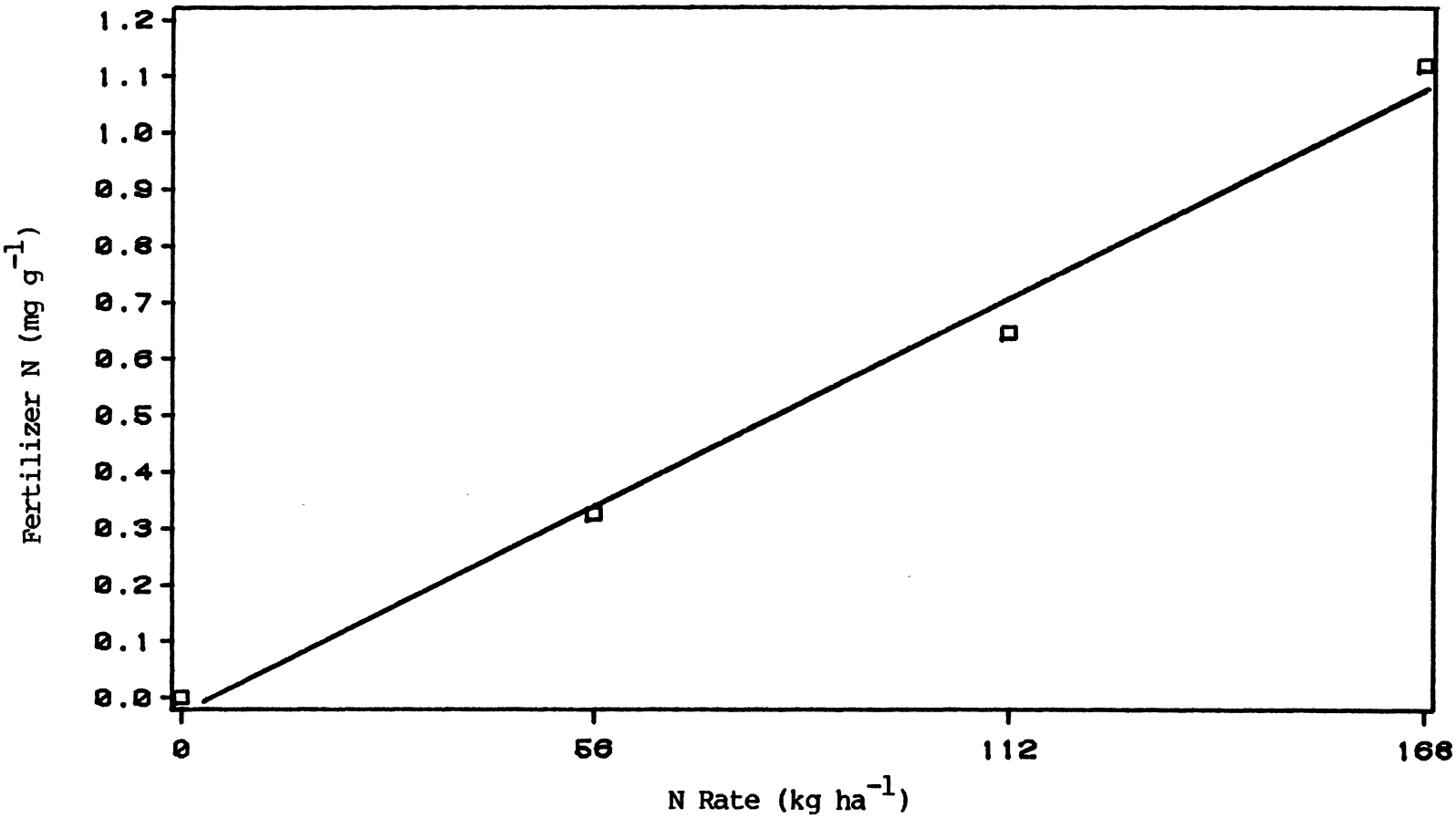

Fig. 1c. Means of fertilizer N by N rate in 1983 averaged over reps, tillages, and plant parts

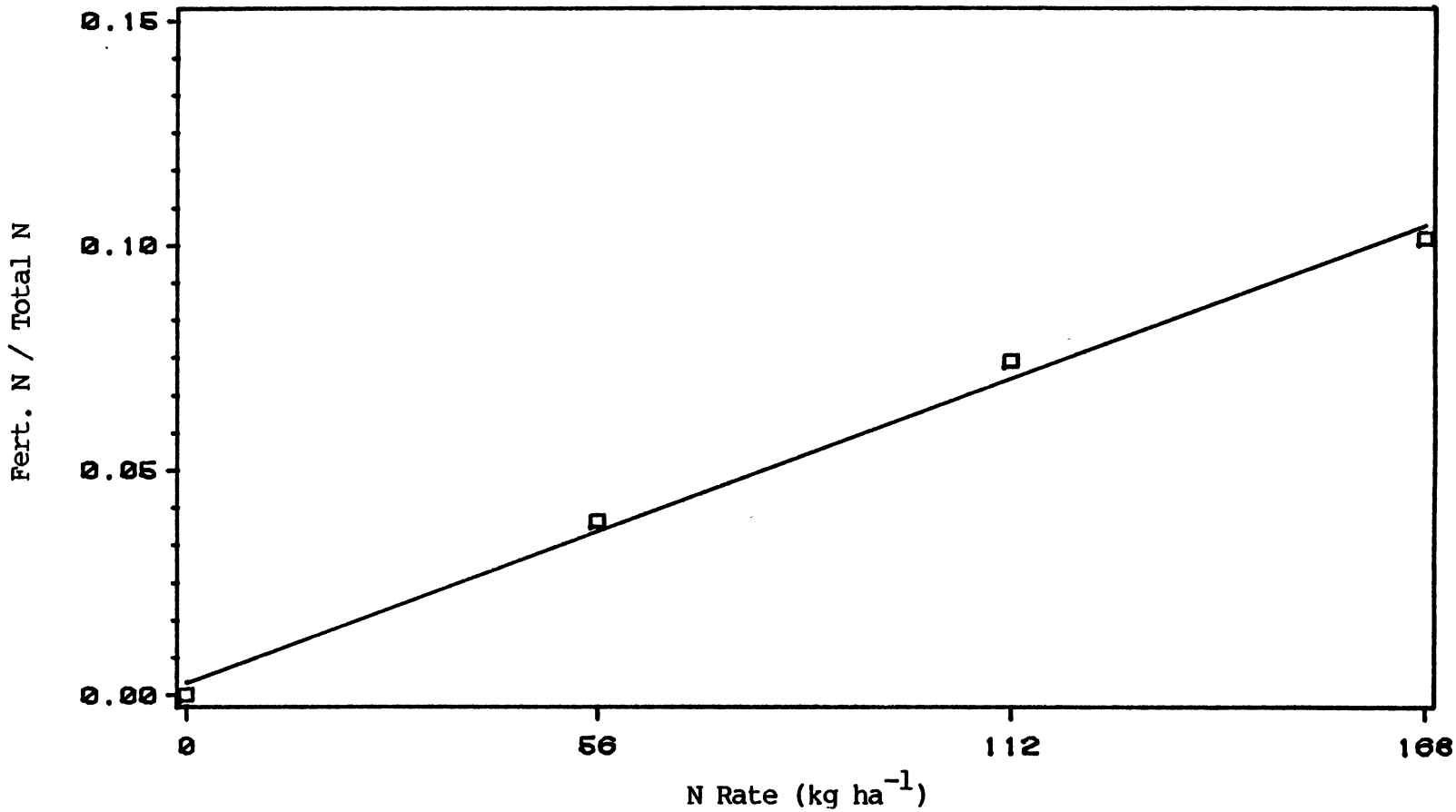

Fig. 1d. Means of ratios of fertilizer N : total N by N rate in 1983 averaged over reps, tillages, and plant parts

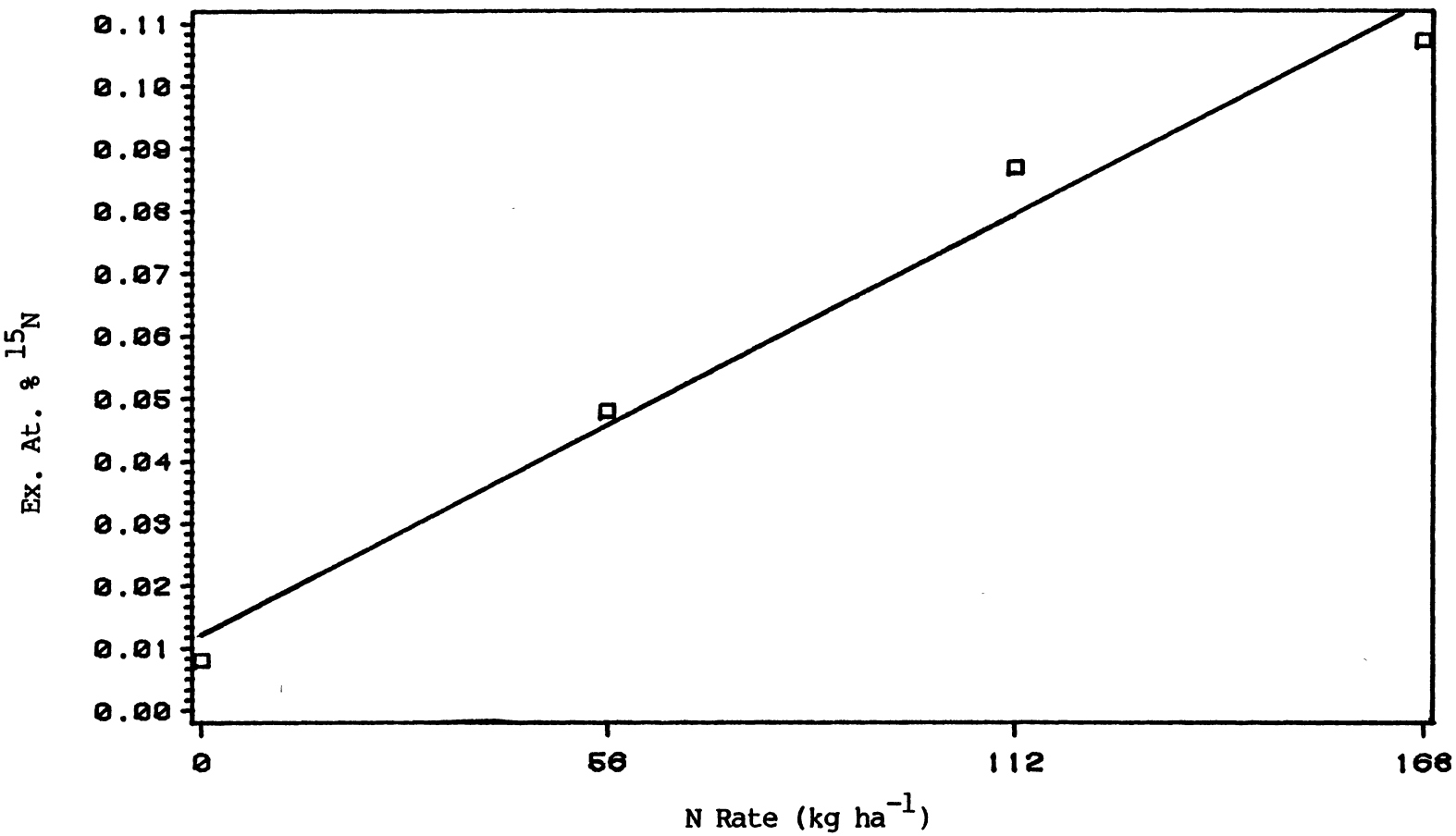

Fig. 2a. Means of ex. at. % ^{15}N by N rate in 1984 averaged over reps, tillages, and plant parts

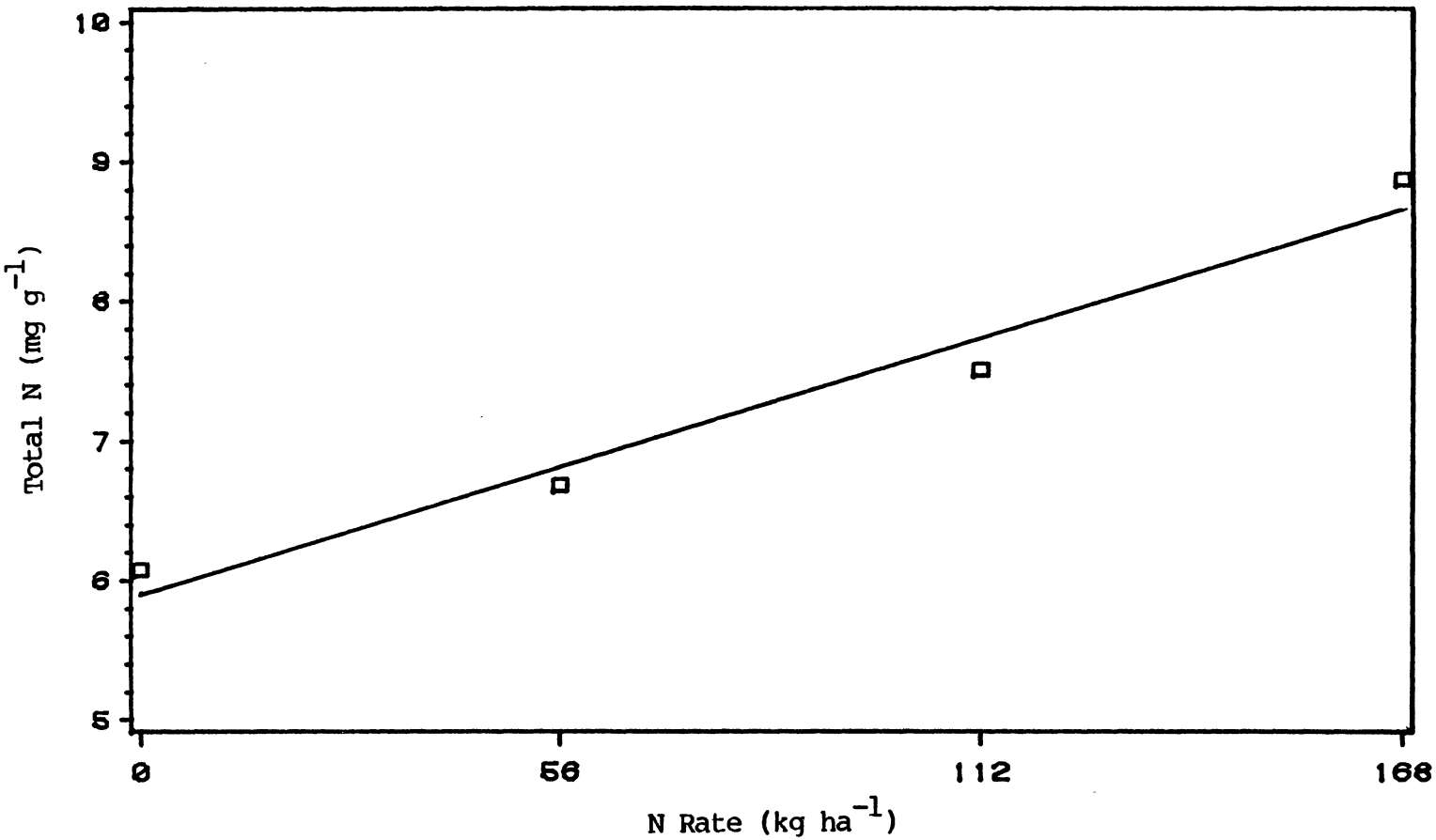

Fig. 2b. Means of total N by N rate in 1984 averaged over reps, tillages, and plant parts

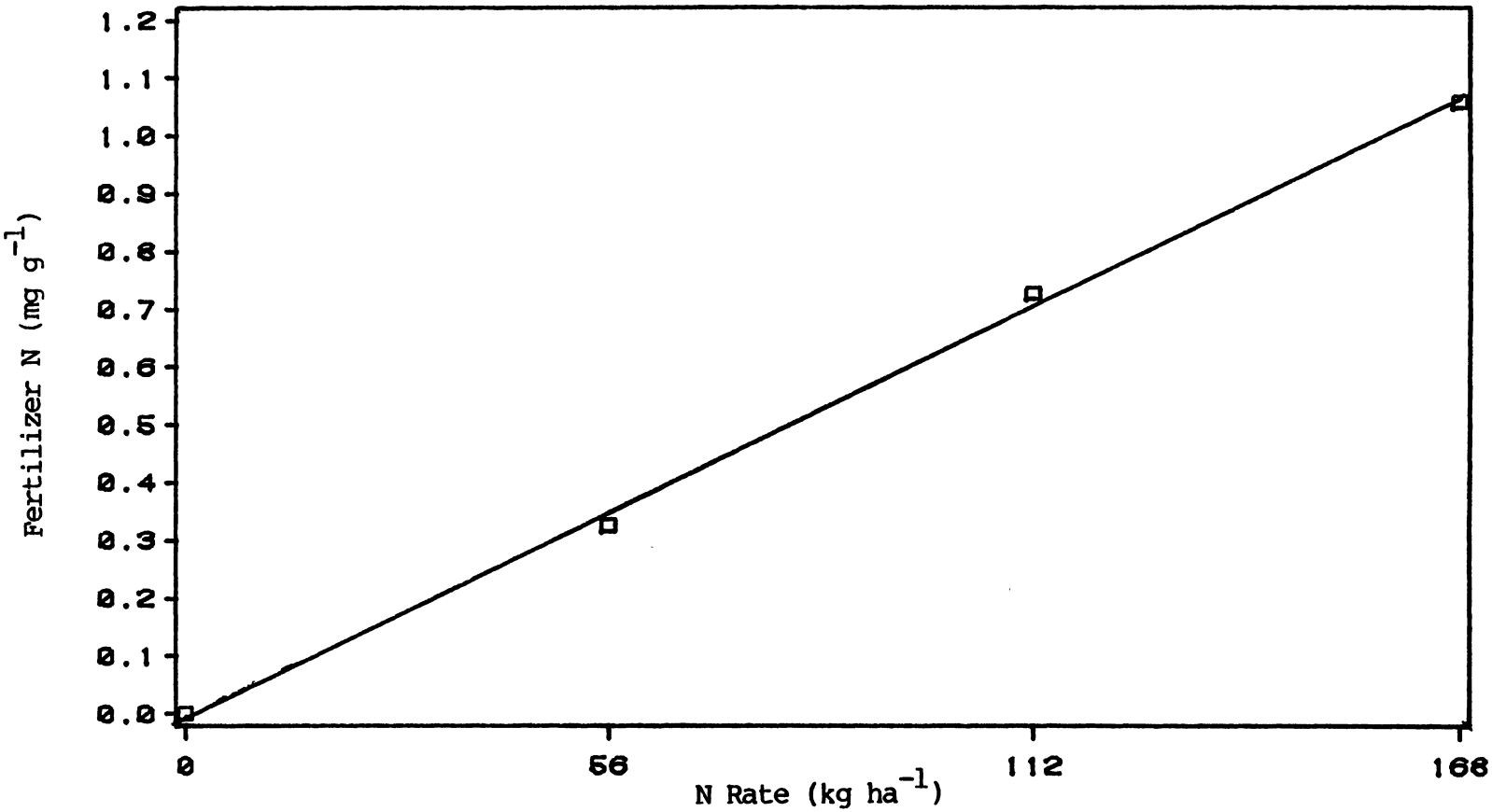

Fig. 2c. Means of fertilizer N by N rate in 1984 averaged over reps, tillages, and plant parts

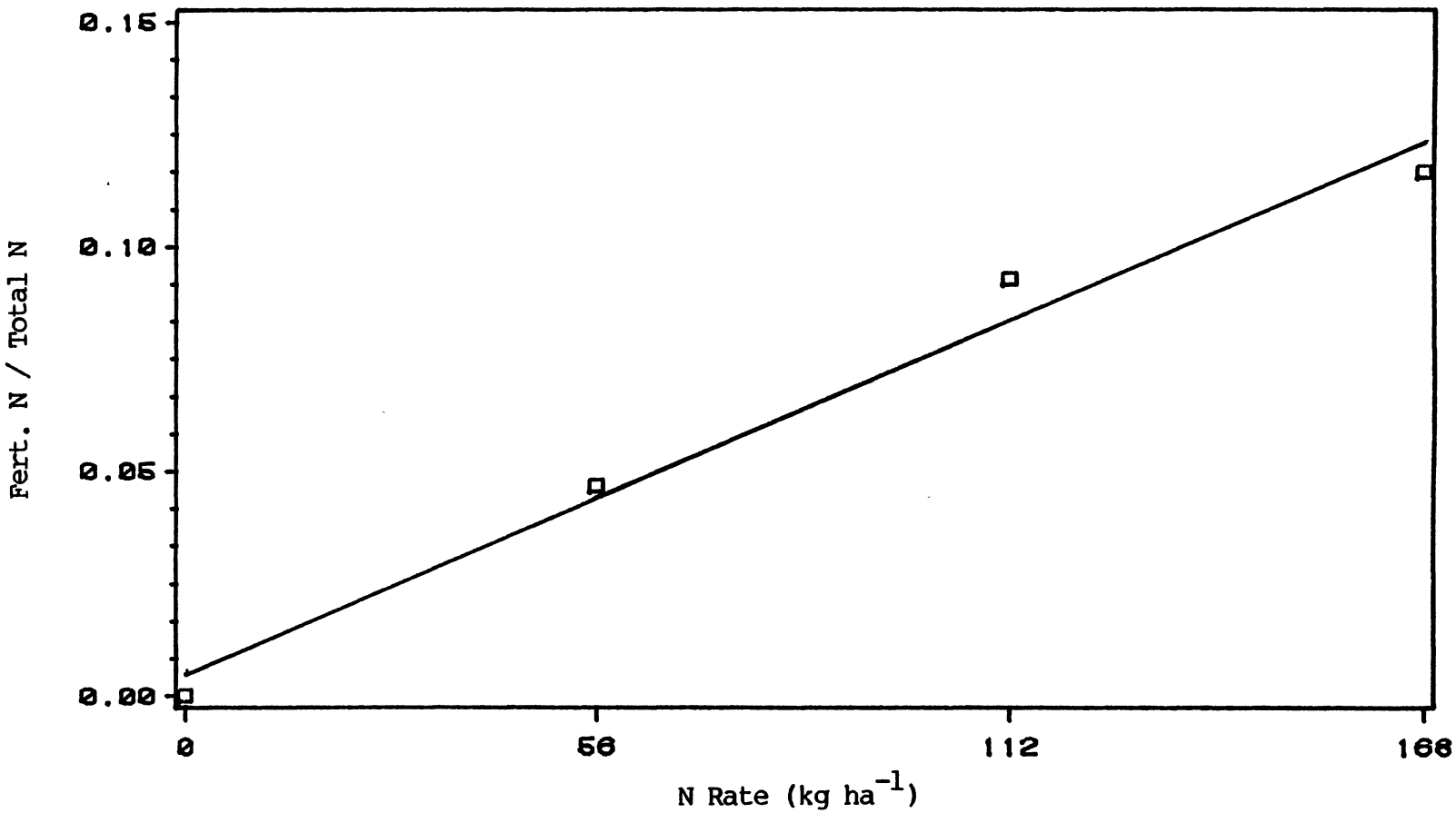

Fig. 2d. Means of ratios of fertilizer N : total N by N rate in 1984 averaged over reps, tillages, and plant parts

TABLE 2. LINEAR REGRESSION EQUATIONS FOR (a) EXCESS ATOM % ^{15}N, (b) TOTAL N, (c) FERTILIZER N, AND (d) RATIOS OF FERTILIZER N : TOTAL N AS A FUNCTION OF FERTILIZER N RATE IN 1983 AND 1984.

	1983		1984	
Dependent Variable	Equation[†]	r^2	Equation	r^2
(a) Ex. at. % ^{15}N	Y = 0.046 + 0.004 X	0.99**	Y = 0.012 + 0.0006 X	0.98**
(b) Total N	Y = 6.501 + 0.024 X	0.93*	Y = 5.90 + 0.016 X	0.97*
(c) Fertilizer N	Y = -0.029 + 0.007 X	0.99**	Y = -0.009 + 0.006 X	0.99***
(d) Fert N/Total N	Y = 0.003 + 0.001 X	0.99**	Y = 0.005 + 0.001 X	0.98**

*,**,*** Indicates significance at the 0.05, 0.01, and 0.001 probability levels respectively.

[†]Y = dependent variable; X = N rate (kg ha^{-1})

TABLE 3. EFFECT OF TILLAGE SYSTEM AND RATE OF FERTILIZER - N ON EXCESS ATOM % ^{15}N IN PLANT PARTS IN 1983.

	Clean Tillage				Reduced Tillage			
	N Rate (kg ha^{-1})				N Rate (kg ha^{-1})			
Plant Part	0	56	112	168	0	56	112	168
	Ex. at. % ^{15}N							
Roots	0.023a*	0.193a	0.461b	0.654b	0.031a	0.222ab	0.344b	0.634a
Crowns	0.025a	0.258a	0.621ab	0.907a	0.026a	0.308ab	0.522ab	0.714a
Leaves	0.016a	0.367a	0.718a	0.910a	0.015a	0.372ab	0.629a	0.617a
Grain	0.101a	0.319a	0.605ab	0.797ab	0.014a	0.443a	0.409ab	0.686a
Stems	0.025a	0.277a	0.541ab	0.717ab	0.022a	0.291ab	0.516ab	0.606a
Chaff	0.020a	0.290a	0.544ab	0.762ab	0.022a	0.162b	0.525ab	0.658a
FLSD (0.05)	0.217	0.217	0.217	0.217	0.227	0.227	0.227	0.227

*Values followed by the same letter within a column are not significantly different at the 0.05 probability level according to pairwise comparisons using Fisher's LSD.

N ha^{-1} rate. There were no significant differences among plant parts under reduced-tillage in 1983 at the 0 or 168 kg N ha^{-1} rates of fertilizer N. Grain was significantly higher in ^{15}N content than the chaff at the 56 kg N ha^{-1} rate but at the 112 kg N ha^{-1} rate only the leaves had a significantly higher ^{15}N content than roots in 1983.

In 1984 significant differences in excess atom % ^{15}N were only observed among N rates averaged over replications, tillage treatments, and plant parts (Table 1). The response of excess atom % ^{15}N to increasing rates of fertilizer -^{15}N is depicted in Fig. 1a. The excess atom % ^{15}N values for 1984, separated by plant part and N rate are shown in Table 4. Single degree of freedom orthogonal contrasts were employed to characterize the response by plant part to increasing rates of fertilizer -^{15}N with respect to ^{15}N content. Contrasts indicated a consistent significant linear increase in excess atom % ^{15}N in all plant parts with respect to increasing N rates in both 1983 and 1984 (Table 4).

There were no differences in excess atom % ^{15}N in leaves and stems which represent the largest amounts of above ground plant residues that may be subject to decomposition and immobilization. The only exception was in the chaff at 56 kg N ha^{-1} under reduced tillage in 1983 (Table 3). Thus, under the factors present in these experiments, it is apparent that the use of such labeled plant materials should prove to be satisfactory for use in further studies evaluating the fate of the residual fertilizer N present in the residues. The information presented in Tables 3 and 4 indicate both satisfactory uniformity and levels of ^{15}N enrichment among plant parts for use in this manner.

TABLE 4. EFFECT OF FERTILIZER - N ON EXCESS ATOM % ^{15}N IN PLANT PARTS AND SINGLE DEGREE OF FREEDOM ORTHOGONAL CONTRASTS FOR INCREASING FERTILIZER N RATES.

	1983			1984						
	Contrast			N Rate (kg ha^{-1})				Contrast		
Plant Part	N linear	N quad.	N cubic	0	56	112	168	N linear	N quad.	N cubic
	F values			Excess Atom % ^{15}N				F values		
Roots	44.50***	2.77	1.99	0.006	0.051	0.079	0.106	35.25***	0.49	0.07
Crowns	62.16***	0.12	0.06	0.006	0.044	0.083	0.094	30.60***	1.18	0.24
Leaves	57.69***	3.10	0.24	0.009	0.054	0.102	0.125	51.93***	0.78	0.30
Grain	42.12***	0.35	0.83	0.005	0.048	0.092	0.108	40.83***	1.20	0.26
Stems	41.41***	0.72	0.08	0.009	0.044	0.083	0.102	33.39***	0.40	0.20
Chaff	50.16***	0.04	0.50	0.010	0.046	0.083	0.108	36.74***	0.15	0.06

*** Indicates significance at the 0.001 probability level.

Total N in Plant Parts

Significant differences of total N in plant parts due to N rates were observed in both 1983 and 1984. This is indicated by the significant interaction for N rate and plant parts in both years (Table 1). Differences among plant parts within given rates of fertilizer N are shown in Table 5 for 1983 and 1984.

An identical pattern of partitioning of the total plant N was found among plant parts at all rates of fertilizer N in 1983 (Table 5). Grain was significantly higher in total N concentration than chaff and leaves which were significantly higher in total N than roots, crowns, and stems. The nature of the response of each plant part in total N concentration to increasing rates of fertilizer N was characterized by use of single degree of freedom orthogonal contrasts (Table 6). In 1983 the total N of leaves, chaff, and grain each responded in a significant linear manner with respect to increasing rates of fertilizer N.

In 1984 total N concentrations among plant parts followed the same pattern at the 0 and 56 kg N ha^{-1} rates of fertilizer N as shown in Table 5. This pattern consisted of grain being significantly higher in N concentration than all other parts, followed by leaves which were significantly higher than stems. The pattern of total N partitioning among plant parts at the 112 kg N ha^{-1} rate found grain to contain significantly more N than all other plant parts. Leaves were significantly higher in total N than crowns and stems. The partitioning of total N among plant parts at the 168 kg N ha^{-1} in 1984 found grain to contain higher total N concentrations than all other

TABLE 5. EFFECTS OF FERTILIZER N RATES ON TOTAL N CONCENTRATION AMONG PLANT PARTS.

	1983				1984			
	N Rate (kg ha^{-1})							
Plant Part	0	56	112	168	0	56	112	168
	Total N (mg g^{-1})							
Roots	2.18c*	2.97c	3.43c	4.18c	3.64bc	4.08bc	5.09bc	5.86c
Crowns	3.82bc	4.27c	4.84c	6.75c	3.44bc	3.98bc	4.58cd	5.84c
Leaves	6.51b	7.88b	10.19b	14.06b	4.96b	5.76b	6.77b	8.85b
Grain	18.08a	19.41a	19.47a	21.57a	18.88a	19.65a	20.55a	22.38a
Stems	3.44c	4.02c	4.47c	5.83c	1.92c	2.23c	2.65d	3.52d
Chaff	6.77b	7.73b	9.20b	13.75b	3.59bc	4.34b	5.36bc	6.71c
FLSD (0.05)	2.99	2.99	2.99	2.99	1.94	1.94	1.94	1.94

*Values followed by the same letter within a column are not significantly different at the 0.05 probability level according to pairwise comparisons using Fisher's LSD.

TABLE 6. SINGLE DEGREE OF FREEDOM ORTHOGONAL CONTRASTS FOR TOTAL N RESPONSES TO INCREASING RATES OF FERTILIZER N.

	F Values					
	1983			1984		
	Contrast					
Plant Part	N linear	N quad.	N cubic	N linear	N quad.	N cubic
Roots	1.74	0.0003	0.02	7.15**	0.07	0.08
Crowns	3.64	0.44	0.06	7.38**	0.30	0.42
Leaves	25.96***	1.31	0.02	19.51***	0.99	0.09
Grain	4.63*	0.12	0.45	15.70***	0.68	0.08
Stems	2.42	0.13	0.04	3.27	0.19	0.02
Chaff	20.96***	2.68	0.28	13.09***	0.21	0.0004

*,**,*** Indicates significance at 0.05, 0.01, and 0.001 probability levels respectively.

plant parts. Leaves were higher in total N concentration than roots, crowns, and chaff. Stems contained lower total N amounts than all other plant parts at the 168 kg N ha^{-1} in 1984 (Table 5). The orthogonal contrasts in 1984 for total N concentration of each plant part indicated a significant linear response for all plant parts except for stems (Table 6).

Partitioning of total N among plant parts was not identical for all rates of fertilizer N in 1983 compared to 1984. However, general trends in partitioning of total N among plant parts at all rates of fertilizer N was indicative of the source-sink relationship within a wheat plant as it approached physiological maturity. The information from these experiments support the final result suggested from a model of translocation and utilization of N in wheat plants proposed by Simpson, Lambers, and Dalling (1983). This is particularly distinct at the higher rates of fertilizer N in 1984 where there was greater segregation of plant parts with respect to total N content.

The model described by Simpson indicated that redistribution of N within winter wheat during grain filling consisted mainly of symplastic transport from the leaves (particularly the flag leaf) and the glumes to the grain. They also measured some translocation in the phloem from the leaves to the roots. However root N was considerably depleted due to apoplastic transport of N to the flag leaf and the glumes. The grain was found to be the ultimate sink for most N by symplastic translocation. This relationship between grain and other plant parts with respect to sink and source characteristics of plant N is also described by Smith, Peterson, and Sander (1983), which reinforces

trends recognized in the present experiments. Smith et al. (1983) reported a steady depletion of N concentration in the shoots (total aboveground portion of the plant minus heads) of winter wheat plants after anthesis. There was also a concomitant increase in both N concentration in the heads and N uptake by the plant from anthesis to complete physiological maturity.

The total N concentrations for the present experiments were in the same range as those presented by Daigger, Sander and Peterson (1976) for wheat plants fertilized with 112 kg N ha^{-1} and separated into leaves, stems, heads, and roots in their experiments.

As indicated in Table 1, there was a significant interaction between tillage system and N rates for total N concentration values in 1984. Both tillage systems were evaluated with regard to response to increasing fertilizer N rates by use of orthogonal contrasts. Results of these contrasts in Table 7 indicated each tillage system responded in a significant linear manner to increasing increments of fertilizer N.

Fertilizer N in Plant Parts

Concentrations of fertilizer N present in the plant parts analyzed followed a pattern similar to that of the total N concentrations. Differences in fertilizer N among plant parts compared within the rates of fertilizer N applied are listed in Table 8 for both 1983 and 1984.

Partitioning of fertilizer N followed a similar pattern for the 56 and 112 kg N ha^{-1} rates of fertilizer N in both experiments (1983 and 1984). Grain contained the largest concentration of fertilizer N which

TABLE 7. SINGLE DEGREE OF FREEDOM ORTHOGONAL CONTRASTS FOR RESPONSES OF TOTAL N CONCENTRATION TO TILLAGE SYSTEM AND INCREASING RATES OF FERTILIZER N IN 1984.

	Tillage	
Contrast	Clean	Reduced
	---------- F values ----------	
N linear	37.75***	24.40***
N quadratic	3.34	0.04
N cubic	0.40	0.06

*** Indicates significance at 0.001 probability level.

TABLE 8. EFFECTS OF FERTILIZER - N RATES ON FERTILIZER N CONCENTRATIONS IN PLANT PARTS.

	1983			1984		
	N Rate (kg ha^{-1})					
Plant Parts	56	112	168	56	112	168
	Fertilizer N (mg g^{-1})					
Roots	0.08b*	0.20b	0.40c	0.21b	0.44b	0.70c
Crowns	0.16b	0.39b	0.77c	0.18b	0.40b	0.60c
Leaves	0.41ab	0.98ab	1.54b	0.31b	0.74b	1.22b
Grain	0.92a	1.28a	2.14a	0.99a	2.08a	2.68a
Stems	0.15b	0.33b	0.53c	0.09b	0.23b	0.38c
Chaff	0.23b	0.67b	1.34b	0.17b	0.47b	0.77bc
FLSD (0.05)	0.54	0.54	0.54	0.52	0.52	0.52

*Values followed by the same letter within a column are not significantly different at the 0.05 probability level according to pairwise comparisons using Fisher's LSD.

was significantly greater than that found in all other plant parts in 1983 and 1984. The leaves were somewhat intermediate in fertilizer N concentration to the other two general groupings of plant parts at these rates of fertilizer N in 1983.

There was a further partitioning of fertilizer N into 3 groups of plant parts at the 168 kg N ha^{-1} rate of fertilizer N in 1983 and 1984. Grain was again significantly higher than all other plant parts in fertilizer N concentration followed then by leaves and chaff which were significantly greater than roots, crowns and stems. In 1984, the chaff component was somewhat intermediate to the two other groups of plant parts (leaves vs. roots, crowns, and stems).

Patterns revealed by the partitioning of the fertilizer N among plant parts was an indication of the source-sink relationship that exists between various components of the wheat plant culminating in these concentrations found at physiological maturity. It was also interesting to note that the general pattern of fertilizer N partitioning was similar to that of the total N among plant parts. This suggested a rather consistent use of fertilizer N with indigenous soil N forms since the final distribution of N in the plant is a function of both N redistribution within the plant and late season uptake of N from the soil regardless of the N source.

All plant parts responded in a significant linear fashion to increasing rates of fertilizer N in both years as revealed by orthogonal contrasts (Table 9). The only exceptions to this occurred with roots in 1983 and the stems in 1984. Both of these components appear to be N sources for redistribution to plant parts such as grain

TABLE 9. SINGLE DEGREE OF FREEDOM ORTHOGONAL CONTRASTS FOR FERTILIZER N RESPONSE TO INCREASING RATES OF FERTILIZER N IN PLANT PARTS.

	F Values					
	1983			1984		
	Contrast					
Plant Part	N linear	N quad.	N cubic	N linear	N quad.	N cubic
Roots	2.36	0.08	0.0006	7.23**	0.02	0.0006
Crowns	9.14**	0.35	0.01	5.47*	0.001	0.012
Leaves	38.26***	0.18	0.03	22.75***	0.22	0.007
Grain	64.98***	0.03	1.52	112.32***	1.04	1.13
Stems	4.48*	0.02	0.0002	2.25	0.02	0.003
Chaff	28.35***	1.19	0.004	9.15**	0.10	0.02

*,**,*** Indicates significance at 0.05, 0.01, and 0.001 probability levels respectively.

TABLE 10. TILLAGE EFFECTS ON FERTILIZER N CONCENTRATION OF PLANT PARTS IN 1984.

Plant Part	Tillage System	
	Clean	Reduced
	------- Fertilizer N (mg g^{-1}) ------	
Roots	0.34bc*	0.34b
Crowns	0.28bc	0.30b
Leaves	0.64b	0.50b
Grain	1.61a	1.26a
Stems	0.19c	0.16b
Chaff	0.35bc	0.36b
FLSD (0.05)	0.37	0.37

*Values followed by the same letter within a column are not significantly different at the 0.05 probability level according to pairwise comparisons using Fisher's LSD.

and chaff as indicated by the patterns of both total N and fertilizer N partitioning (Tables 5 and 8).

In 1984, there was a statistically significant interaction between tillage systems and plant parts with regard to final fertilizer N concentrations (Table 1). The differences between tillage systems in the distribution of fertilizer N among plant parts averaged over all replications and N rates are shown in Table 10. Grain contained significantly higher fertilizer N concentrations than all other plant parts regardless of tillage system. Leaves in plants grown under clean tillage had significantly higher fertilizer N concentrations than stems with roots, crowns, and chaff being somewhat intermediate. There were no significant differences in fertilizer N among parts of plants grown under reduced tillage other than grain which contained higher concentrations of fertilizer N.

Ratios of Fertilizer N : Total N in Plant Parts

The ratios of fertilizer N : total N (FN/TN) were calculated to evaluate the effects on increasing rates of fertilizer N on the proportions of fertilizer N present in each plant part, realizing of course that such a ratio is a direct reflection of the ^{15}N content. In 1983 a significant three-way interaction existed with tillage systems, N rates, and plant parts (Table 1). The information presented in Table 11 describes the ratio of FN/TN present in each plant part under each fertilizer N rate for both tillage systems separately in 1983. There were no significant differences among plant parts at the 56 kg N ha^{-1} rate of fertilizer N under the clean-tillage system. However, at the

TABLE 11. EFFECT OF TILLAGE SYSTEM AND FERTILIZER N RATE ON THE RATIO OF FERTILIZER N : TOTAL N IN PLANT PARTS IN 1983.

	Tillage					
	Clean			Reduced		
	N Rate (kg ha^{-1})					
Plant Parts	56	112	168	56	112	168
	Fert. N : Total N					
Roots	0.025a	0.064b*	0.092b	0.028ab	0.046b	0.089a
Crowns	0.034a	0.087ab	0.129a	0.041ab	0.073ab	0.101a
Leaves	0.051a	0.103a	0.131a	0.052ab	0.090a	0.088a
Grain	0.032a	0.074ab	0.102ab	0.063a	0.058ab	0.098a
Stems	0.037a	0.075ab	0.101ab	0.039ab	0.072ab	0.085a
Chaff	0.040a	0.077ab	0.109ab	0.020b	0.074ab	0.093a
FLSD (0.05)	0.036	0.036	0.036	0.036	0.036	0.036

*Values followed by the same letter within a column are not significantly different at the 0.05 probability level according to pairwise comparisons using Fisher's LSD.

112 and 168 kg N ha^{-1} rates, leaves had a significantly higher ratio of FN/TN than roots. The ratio FN/TN for crowns was also greater than roots at the 168 kg N ha^{-1} level. There were only slight differences in the FN/TN ratio among plant parts under reduced tillage with respect to rate of fertilizer N. Grain ratios of FN/TN were significantly greater than the chaff ratios at the 56 kg N ha^{-1}. Leaves had significantly higher ratios than roots at the 112 kg N h^{-1}. There were no significant differences in FN/TN ratios among any of the plant parts at the 168 kg N ha^{-1} rate of fertilizer N.

In 1984 there was only a significant response in the ratios of FN/TN for N rate as a source of variation (Table 1). The response pattern of these ratios to increasing rates of fertilizer N is shown in Fig. 2d. There was a significant linear response due to increasing rates of fertilizer N for all plant parts in the ratios of FN/TN (Fig. 2d and Table 13). This was true for both 1983 and 1984.

Some differences in ratios of FN/TN existed among plant parts within rates of fertilizer N averaged over replications and tillage treatments for 1983 and 1984 (Table 12). The presentation of these ratios in the format of Table 12 provides an opportunity to review the trends and proportions of fertilizer N present in each plant part for both experiments. It is interesting to note that the proportions of fertilizer N were relatively low. One must consider the fact that in both experiments (both years) $^{15}NH_4NO_3$ was used as the labeled source of fertilizer N with 7.2 and 1.2 atom % ^{15}N in 1982-83 and 1983-84 crop seasons respectively. This level of enrichment is expressed as atom % ^{15}N of the NH_4^+-N only. Therefore an assumption could be made that if

TABLE 12. EFFECTS OF FERTILIZER N RATES ON RATIOS OF FERTILIZER N : TOTAL N IN PLANT PARTS.

	1983			1984		
	N Rate (kg ha^{-1})					
Plant Parts	56	112	168	56	112	168
	Fert. N : Total N					
Roots	0.026b*	0.055b	0.090a	0.052	0.085	0.116
Crowns	0.038ab	0.080a	0.115a	0.045	0.090	0.104
Leaves	0.052a	0.096a	0.109a	0.052	0.109	0.136
Grain	0.047ab	0.066b	0.100a	0.050	0.101	0.120
Stems	0.038ab	0.074ab	0.093a	0.041	0.086	0.109
Chaff	0.030ab	0.075ab	0.101a	0.041	0.085	0.116
FLSD (0.05)	0.025	0.025	0.025	NS	NS	NS

*Values followed by the same letter within a column are not significantly different at the 0.05 probability level according to pairwise comparisons using Fisher's LSD; NS = nonsignificant.

TABLE 13. SINGLE DEGREE OF FREEDOM ORTHOGONAL CONTRASTS FOR RESPONSE OF THE RATIOS OF FERTILIZER N : TOTAL N CONCENTRATIONS TO INCREASING RATES OF FERTILIZER N IN PLANT PARTS.

	F Values					
	1983			1984		
	Contrast					
Plant Part	N linear	N quad.	N cubic	N linear	N quad.	N cubic
Roots	40.00***	0.19	0.01	27.84***	0.39	0.06
Crowns	66.56***	0.02	0.06	24.17***	0.93	0.19
Leaves	61.74***	3.33	0.64	40.97***	0.62	0.24
Grain	45.06***	0.38	0.89	32.21***	0.94	0.20
Stems	44.38***	0.77	0.09	26.40**	0.32	0.16
Chaff	53.67***	0.04	0.53	29.21***	0.10	0.08

,* Indicates significance at 0.01 and 0.001 probability levels respectively.

all of the NO_3^--N present in the $^{15}NH_4NO_3$ fertilizer was utilized by the wheat plants in a similar manner as $^{15}NH_4^+$, then all ratios of FN/TN could be multiplied by two. This provides higher proportions of fertilizer N present but fertilizer N recovery still appears to be low since there was a consistent linear response to increasing rates of fertilizer N for most plant parts. This suggests a large amount of the N present in the plant was derived from indigenous soil N. This also suggests a relatively large amount of soil N to be present in an easily mineralizable N pool which was apparently converted and utilized by the wheat plants. This was particularly apparent since residual soil NO_3^--N tests prior to the initiation of the experiments revealed no carryover of available N in the field study area.

This discrepancy with respect to the ratios of FN/TN warrants further consideration. A mass balance approach to assess fertilizer N uptake and recovery from the soil-plant system in question would be a reasonable approach to begin to resolve this situation. Such an approach is another facet of the overall research project of which the experiments described herein represent one part.

SUMMARY

No significant differences were found due to tillage systems with respect to any variable studied in either 1983 or 1984. However, increasing rates of fertilizer N resulted in increases in (a) ^{15}N from labeled fertilizer sources, (b) total N concentration, (c) concentration of fertilizer N, or (d) ratios of FN/TN in a consistent linear manner for all plant parts of winter wheat. This conclusion was

supported by evidence from the single degree of freedom orthogonal contrasts used to test the nature of the response of each plant part to increasing N rate by the parameters studied. The four rates of fertilizer N and two levels of ^{15}N enrichment of the labeled fertilizer sources tested caused only slight differences in ^{15}N content among plant parts when compared within rates of fertilizer N included experimentally.

From these results it is concluded that the use of above ground plant parts labeled with ^{15}N by use of enriched fertilizer materials in the range of 1.2 - 7.2 atom % ^{15}N, as used in these experiments, should provide adequate uniformity in labeling with ^{15}N among plant parts. Satisfactory uniformity and levels of ^{15}N enrichment among plant parts was realized for both CT and RT systems. Therefore, these wheat crop residues would be satisfactory for use in subsequent studies of residual fertilizer N present as constituents of the residues that are subject to decomposition and mineralization processes.

REFERENCES

Bray, R.H., and L.T. Kurtz. 1945. Determination of total, organic, and available forms of phosphorus in soils. Soil Sci: 59:39-45.

Bremner, J.M. 1965. Total nitrogen. In C.A. Black et al. (ed.) Methods of soil analysis, Part 2. Agronomy 9:1149-1178.

Daigger, L.A., D.H. Sander, and G.A. Peterson. 1976. Nitrogen content of winter wheat during growth and maturation. Agron. J. 68:815-818.

Doran, J.W. 1980. Soil microbial and biochemical charges associated with reduced tillage. Soil Sci. Soc. Am. J. 44:518-524.

Douglas, L.A., A. Riazi, and L.J. Smith. 1980. A semi-micro method for determining total nitrogen in soils and plant material containing nitrite and nitrate. Soil Sci. Soc. Am. J. 44:431-433.

Dowdell, R.J., and R.Q. Cannell. 1975. Effect of plowing and direct drilling on soil nitrate content. J. Soil Sci. 26:53-61.

Fredrickson, J.K., F.E. Koehler, and H.H. Cheng. 1982. Availability of ^{15}N - labeled nitrogen in fertilizer and in wheat straw to wheat in tilled and no-till soil. Soil Sci. Soc. Am. J. 46:1218-1222.

Hauck, R.D. and J.M. Bremner. 1976. Use of traces for soil and fertilizer nitrogen research. Adv. Agron. 28:219-266.

Hill, R.L., and R.M. Cruse. 1985. Tillage effects on bulk density and soil strength of two mollisols. Soil Sci. Soc. Am. J. 49:1270-1273.

House, G.F., B.R. Stinner, D.A. Crossley, Jr., E.P. Odum, and G.W. Langdale. 1984. Nitrogen cycling in no-tillage agroecosystems. J. Soil Water Conserv. 39:194-200.

Johnson, G.V., and B.B. Tucker. 1982. OSU soil test calibrations. OSU Fact Sheet 2225. Coop. Ext. Service, Div. of Agric., Oklahoma State University, Stillwater, OK.

Knudsen, D., G.A. Peterson, and P.F. Pratt. 1982. Lithium, sodium, and potassium. In A.C. Page et al. (ed.) Methods of soil analysis, part 2, 2nd ed. Agronomy 9:225-246.

Large, E.C. 1954. Growth stages in cereals. Plant Pathol. 3:128-129.

Lindstrom, M.J., W.B. Voorhees, and C.A. Onstad. 1984. Tillage system and residue cover effects on infiltration in north- western corn belt soils. J. Soil Water Conserv. 39(1):64-68.

Magleby, R., D. Gadsby, D. Colacicco, and J. Thigpen. 1985. Trends in conservation tillage use. J. Soil Water Conserv. 40(3):274-276.

McMahon, M.A., and G.W. Thomas. 1976. Anion leaching in two Kentucky soils under conventional tillage and no-till culture. Agron. J. 68:437-442.

Myers, R.J.K., and E.A. Paul. 1971. Plant uptake and immobilization of ^{15}N labelled ammonium nitrate in a field experiment with wheat. p. 55-64. In Nitrogen -15 in soil-plant studies. IAEA, Vienna.

Porter, L.K., and W.A. O'Dean. 1977. Apparatus for preparing nitrogen from ammonium chloride for nitrogen -15 determination. Annal. Chem. 49:514-516.

Powlson, D.S. 1980. Effect of cultivation on the mineralization of nitrogen in soil. Plant Soil 57:151-153.

Rice, C.W., and M.S. Smith. 1982. Denitrification in no-till and plowed soils. Soil Sci. Soc. Am. J. 46:1168-1173.

Rice, C.W., M.S. Smith, and R.L. Blevins. 1986. Soil nitrogen availability after long-term continuous no-tillage and conventional tillage corn production. Soil Sci. Soc. Am. J. 50:1206-1210.

Simpson, R.J., H. Lambers, and M.J. Dalling. 1983. Nitrogen redistribution during grain growth in winter wheat (Triticum aestivum L.). Plant Physiol. 71:7-14.

Smith, T.L., G.A. Peterson, and D.H. Sander. 1983. Nitrogen distribution in roots and tops of winter wheat. Agron. J. 75:1031-1036.

Statistical Analysis System Institute. 1982. The ANOVA procedure. p. 119-138. In A.A. Roy (ed.) SAS user's guide:Statistics. Statistical Analysis System Institute, Inc. Raleigh, NC.

Steel, R.G.D. and J.H. Torrie. 1980. Principles and procedures of statistics, 2nd ed. McGraw-Hill Book Company, Inc., New York.

Wagger, M.G., D.E. Kissel, and S.J. Smith. 1985a. Uniformity of nitrogen -15 enrichment in different plant parts and subsequent decomposition of labeled crop residues. Soil Sci. Soc. Am. J. 49:1205-1208.

Wagger, M.G., D.E. Kissel, and S.J. Smith. 1985b. Mineralization of N from nitrogen -15 labeled crop residues under field conditions. Soil Sci. Soc. Am. J. 49:1220-1226.

Wilson, D.O., and W.L. Hargrove. 1986. Release of nitrogen from crimson clover residue under two tillage systems. Soil Sci. Soc. Am. J. 50:1251-1254.

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