In situ measurements of sulfate incorporation into forest floor and soil organic matter

TIMOTHY C. STRICKLAND[1] AND JOHN W. FITZGERALD
Department of Microbiology, University of Georgia, Athens, GA, U.S.A. 30602
AND
WAYNE T. SWANK
Southeastern Forest Experiment Station, Coweeta Hydrologic Laboratory, Otto, NC, U.S.A. 28763

Received July 29, 1985[2]
Accepted December 19, 1985

STRICKLAND, T. C., J. W. FITZGERALD, and W. T. SWANK. *In situ* measurements of sulfate incorporation into forest floor and soil organic matter. Can. J. For. Res. 16: 549-553.

Litter and soil from a mixed mature hardwood forest were examined for the capacity to incorporate ^{35}S-labelled sulfate into organic matter *in situ*. Amounts of sulfate incorporated within 48 h of field incubation were 70, 49, and 18% of added ^{35}S per gram of substrate in the O1, O2, and A horizons, respectively. These potentials increased in the respective horizons to 74, 61 and 29% after 7 days. The incorporated ^{35}S was predominately in the form of carbon-bonded S (17-48% of added ^{35}S). *In situ* incorporation rates exceeded rates previously estimated by laboratory incubations and the former rates showed a positive response to increase sulfate loading.

STRICKLAND, T. C., J. W. FITZGERALD et W. T. SWANK. *In situ* measurements of sulfate incorporation into forest floor and soil organic matter. Can. J. For. Res. 16: 549-553.

Les auteurs ont étudié *in situ* la capacité d'incorporation de $^{35}SO_4$ dans la matière organique de la litière et du sol d'une forêt feuillue mixte adulte. Les quantités de SO_4 incorporé durant 48 h d'incubation au champ s'élevaient à 70, 49 et 18% de ^{35}S ajoute par gramme de stubstrat dans les horizons O1, O2 et A, respectivement. Les potentiels d'incorporation dans les memes horizons apres 7 jours s'élevaient respectivement a 74, 61 et 29%. Le ^{35}S incorporé était principalement sous forme S lie au carbone (17 a 48% de ^{35}S ajouté). Les taux d'incorporation *in situ* furent plus élevés que les taux estimés antérieurement au cours d'incubation en laboratoire et ils ont montré une réponse positive a une charge accrue de sulfate.

[Traduit par la revue]

Introduction

Acid rain and snow represent major input sources for the sulfate anion in forested ecosystems and, at the Coweeta Hydrologic Laboratory in North Carolina, sulfate accounts for 68% of the anionic composition of precipitation (Swank et al. 1984.) Many forests exhibit export discrepancies in sulfate concentration, which suggests that the anion is retained within these ecosystems (Heinrichs and Meyer 1977; Shriner and Henderson 1978; Johnson et al. 1979). Swank and Douglass (1977) reported that the Coweeta watersheds accumulated up to 13 kg ha^{-1} $year^{-1}$ of sulfate S. Sulfate adsorption may account for some of the observed S retention (Johnson and Henderson 1979; Johnson et al. 1980; Johnson et al. 1982) and laboratory studies indicated that the incorporation of sulfate into soil organic matter also plays an important role in the S-retention process in agricultural (Freney et al. 1971; Saggar et al. 1981) as well as forested ecosystems (Fitzgerald et al. 1982; Strick et al. 1982; Fitzgerald et al. 1983; Strickland and Fitzgerald 1984; Swank et al. 1984). Although much effort has been expended to determine steady-state levels of S in soils (Williams and Steinbergs 1959; Freney 1961; Lowe 1964; Freney et al. 1969; Jones et al. 1972; Tabatabai and Bremner 1972), to our knowledge, very little, other than the work of Freney et al. (1971), has been done to determine the direct *in situ* contribution of microflora to the formation of extracellular organic S. In the current work, evidence is presented that documents the *in situ* incorporation of [^{35}S]sulfate into organic matter. Moreover, the forms of organic sulfur generated in these field incubations have been delineated.

Materials and methods

In 1982, a transect was established on watershed 18 (Coweeta Hydrologic Laboratory, Otto, NC) consisting of ten 0.01-ha plots located about 40 m apart. The transect is at midelevation (780-820 m) on this mixed mature hardwood forest and runs from ridge to stream to ridge. Since 1982, laboratory experiments have continued in an effort to characterize S-transformation processes in samples of forest floor and soil collected from this transect. Plot 9 of the transect was chosen to examine *in situ* sulfate incorporation processes. The soil type on this plot is of the sandy loam Ashe series, a Typic Dystrochrept. The vegetation is dominated by a rhododendron understory and a mixed hardwood overstory that includes hickory, red maple, chestnut oak, scarlet oak, sourwood, and black birch (Day and Monk 1974). The *in situ* fate of sulfate was monitored during July 1984 using a ^{35}S label ($Na_2{}^{35}SO_4$, Amersham, approx. 3.3 x 10^{10} Bq $mmol^{-1}$).

Pretreatment of radioactive sulfate to remove organic S

The sodium sulfate preparation received from Amersham was contaminated with organic sulfate. Therefore, it was necessary to hydrolyze the sulfate ester linkages containing this organic S to yield inorganic sulfate (Fitzgerald 1976). This was accomplished by making the commercial preparation 3 M in hydrochloric acid and autoclaving for 2 h.

Sample treatment and incubation

Two samples (25 x 25 x 25 cm) were dug out and placed intact into containers that allowed adequate drainage. One side of each container was hinged to allow insertion of the sample with minimum disturbance and care was taken to maintain the integrity and orientation of the Ol and O2 forest floor layers and the soil. Sampling in this manner included the A1 (0–10 cm) and Ap (10-25 cm) components of the solum. The samples and containers were then placed in larger individual containers (catch basins), which had been sealed to prevent water loss. The interior containers were placed on supports to yield a 5 cm elevation from the catch basin bottom to ensure drainage.

Approximately 1.5 x 10^8 Bq of ^{35}S-labelled sulfate (0.78 nmol SO_4 S) was added to each sample. Additions of ^{35}S were made in

[1]Present address: Department of Soil Science, Oregon State University, Corvallis, OR, U.S.A. 97331.
[2]Revised manuscript received December 16, 1985.

TABLE 1. Pool sizes of endogenous sulfur (milligrams per kilogram of litter or soil) in samples used for *in situ* [^{35}S]sulfate incorporation

Sulfur pool	2-day incubation O1	2-day incubation O2	2-day incubation A	7-day incubation O1	7-day incubation O2	7-day incubation A
Total S	804.8	1047.0	247.8	971, ,9	768.,7	188.7
ffl-reducible S	209.9	221.7	46.2	210, ,6	328.,0	125.6
	(26.1)	(21.2)	(18.6)	(21, .7)	(42. .7)	(66.6)
Non-HI-reducible S	595.0	835.2	201.6	761.,3	440.,7	63.2
	(73.9)	(79.8)	(81.4)	(78, ,3)	(57.,3)	(33.5)
Raney Ni reducible S	394.4	63.9	6.6	501, ,4	309.,3	28.5
	(49.0)	(6.1)	(2.7)	(51..6)	(40.2)	(15.1)
Sulfonate S	200.6	771.4	195.0	260, .0	131. .3	34.7
	(24.9)	(73.7)	(78.7)	(26, .8)	(17, .1)	(18.4)

NOTE: Values in parentheses are percentages of total S in the horizon.

10-mL increments as evenly as possible over the surface of each sample. Each aliquot was washed into the sample with **10 mL** of deionized water. The final volume added to each sample was 200 **mL.** The sample containers were then left open on the watershed for 2 or 7 days, as appropriate. There was **2.3** cm of rainfall during the initial 2-day incubation and **7.8** cm of rain accumulated between days 2 and **7.** During this period, the average **throughfall** input was **3.45 ppm (0.108 mmol** L^{-1}**)** SO_4^{2-} **S.**

Sample analysis

The **O1,** O2 and A horizons were separated from each sample following incubation. Roots were removed by hand and maintained separately. The O2 and A horizons were passed through a **1-cm** sieve and mixed well. Eight **subsamples** of each horizon were then extracted by the method of Fitzgerald et **al.** (1983), which completely recovers water-soluble ^{35}**S, salt-extractable** ^{35}**S,** and acid-alkali **extractable** ^{35}**S** (insoluble organic **S).** The amount of ^{35}**S** present in each of the above fractions was determined by liquid scintillation counting and combined yields from the three fractions were taken as **100%. Aliquots** (80 **µL)** of each extract were then subjected to **electrophoresis** in **0.1** M KH_2PO_4–K_2HPO_4 buffer, pH 8.0 **(2 h** at 200V) and individual ^{35}**S-labelled** components were located on dried strips by scanning. The area of each peak on the chart paper, representing a particular ^{35}**S-labelled** component, was determined by **triangulation.** The quantity of each component was determined as a percent of the total radioactivity on each strip.

Characterization of organic sulfur pools

The **sulfur** status of samples was estimated for each horizon after field incubation was completed. Total S was determined by the alkaline oxidation method of **Tabatabai** and Bremner **(1970),** total **hydriodic** acid reducible S (Hi-reducible S) by the method of Johnson and **Nishita** (1952), and total S not reducible with hydriodic acid **(non-HI-reducible** S) was calculated as the difference between total S and Hi-reducible **S.** Hydriodic acid reduction converts S in sulfate and ester sulfate to **sulfide,** which is determined **colorimetrically,** while forms of S not reduced by hydriodic acid include **amino** acid and **sulfonate** sulfur (Johnson and Nishita 1952; Freney 1958). Raney nickel reducible S (amino acid S) was determined by the method of Freney et **al. (1970)** and sulfonate S was determined as the difference between non-HI-reducible S and Raney nickel reducible **S.**

In** situ* levels of soluble and adsorbed S were determined as **follows:** subsamples of each horizon (4 g wet weight) were mixed with 0.5 M Na_2HPO_4 (w/v, 1:5), shaken for 30 **min,** and **centrifuged** to remove **particulates.** All samples were treated 3 times successively in this manner and supernatant fluids were combined and filtered **(2.0-µm retention).** The amount of Hi-reducible S extracted per **gram** dry weight was determined. Subsamples of the extract were **tnen** filtered successively through 0.45- and **0.22-µm** membranes and sulfate S (per gram dry weight) was determined by **anion chromatography** (Dionex 2010i CMA-6) after column elution with **2.8 m*M $NaHCO_3$ - **2.2 m*M*** Na_2CO_3 at a flow rate of 2.5 mL min^{-1}**. Phosphate-extractable** ester sulfate was taken as the difference between Hi-reducible S and sulfate; **nonphosphate-extractable** ester sulfate was taken as the difference between total Hi-reducible S and phosphate-ex**tractable,** Hi-reducible S.

Results

Sulfur status of litter and soil after field incubation

Examination of the S status of the samples (Tables 1 and 2) indicates that in the upper 28 cm of the forest litter and soil, S is concentrated in organic matter. In no instance did inorganic sulfate make up greater than **18%** of the total S (Table 2) and this anion may constitute as little as 2% (at least in the O2 horizon). Carbon-bonded S (non-HI-reducible S) was the dominant form of organic S in the litter layers (Table **1),** while ester sulfate predominated in the A horizon (Table **2).** Data in Table 2 also show that the majority of the ester sulfate was present in a form that was not extractable with phosphate. Increased field incubation from 2 to 7 days resulted, except for the Ol horizon, in decreased levels of sulphonate S with concomitant increases in amino acid S (Raney Ni reducible **S;** Table **1)** and ester sulphate (nonphosphate extractable; Table 2).

Incorporation of $^{35}SO_4^{2-}$ *into organic matter*

Figure 1 illustrates the **electrophoretic** patterns of extracts obtained after 2 or 7 days incubation of the Ol horizon with ^{35}**S-labelled** sulfate. Extraction was carried out with *(a)* deionized water, *(b)* **1.0** M salt solutions, **(*c*) 6.0** M hydrochloric acid, and **(*d*)**2.0 M sodium hydroxide. Similar patterns were observed in the O2 and A horizons. The component that exhibited a mobility of **12–13** cm was identified as sulfate; all other ^{35}**S-labelled** components were some form of organic **S.** Figure 1 indicates the presence of at least two different organic S components that were in a soluble (Fig. **1*a*)** or absorbed form (Fig. **1*b*).** These particular components in all horizons examined constituted less than 6% of the ^{35}**S** present (Table 3). Figures **1*c*** and **1*d*** show the ^{35}**S-labelled** organic components present in the acid and alkali extracts, respectively. The levels of these components fluctuated depending on incubation time and horizon, ranging from **18** to 74% of the total extractable ^{35}**S.** Table 3 also shows that the organic S formed during *in situ* incubation contained ester sulfate as well as carbon-bonded **S,** but the latter was the dominant form of ^{35}**S-labelled** organic S found in all cases except in the O2 horizon after 7 days incubation. In terms of the levels of available sulfate S, the

TABLE 2. Endogenous levels of inorganic sulfate and ester sulfate (milligrams per kilogram of litter or soil) in samples used for *in situ* [^{35}S]sulfate incorporation

	2-day incubation			7-day incubation		
Sulfur pool	O1	O2	A	O1	O2	A
Phosphate-extractable						
HI-reducible S	72.8	72.5	36.2	205.8	55.9	39.7
	(9.0)	(62.9)	(14.6)	(21.2)	(7.3)	(21.0)
Inorganic sulfate	86.9	25.0	23.0	177.2	16.1	32.3
	(10.8)	(2.4)	(9.3)	(18.2)	(2.1)	(17.1)
Ester sulfate	0	47.6	13.2	28.6	39.7	7.5
	(0)	(4.5)	(5.3)	(2.9)	(5.2)	(4.0)
Nonphosphate **extractable**						
Ester sulfate	137.1	149.2	10.0	4.8	272.2	85.9
	(17.0)	(14.3)	(4.0)	(0.5)	(35.4)	(45.5)

NOTE: Values in parentheses are percentages of total S in the horizon (see Table 1 for total S values).

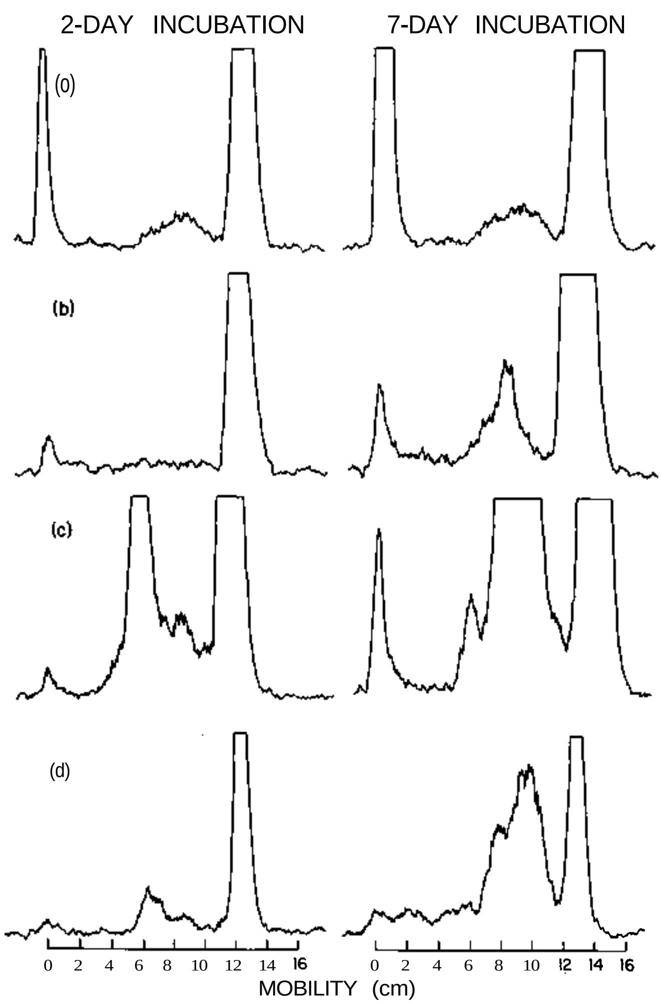

FIG. 1. Separation by electrophoresis of ^{35}S-labelled metabolites present in extracts after field incubation of the O1 forest floor with [^{35}S]sulfate. Electrophoretogram scans are shown of (a) water-soluble S, *(b)* salt-extractable (adsorbed) S, (c) acid-extractable (organic) S, and *(d)* alkali-extractable (organic) S. Similar scans obtained for the O2 and A1 horizons are omitted for purposes of clarity.

amount of ^{35}S-labelled sulfate incorporated into insoluble organic matter, as determined by acid-alkali extractability, was greatest in the Ol layer and least in the A horizon (Table 4).

As expected from results of laboratory incubations carried out by Johnson et al. (1980), substantial amounts of added sulfate became adsorbed during field incubation, especially in the A horizons (81% of total extractable ^{35}S; Table 3). The level of salt-extractable sulfate in these horizons decreased after 7 days, indicating that some desorption of added sulfate can take place. In agreement with results of laboratory incubations of samples taken from this watershed (Fitzgerald et al. 1983), the O1, O2, and A horizons exhibited progressively increasing capacities to adsorb added sulfate in field incubations (Table 3).

Discussion

The predominance of organic S in the upper horizons of temperate forests is well documented (Johnson et al. 1980; McGill and Cole 1981; David et al. 1982). Since large inputs of the sulfate anion occur in many forested areas, it is reasonable to expect biological accumulation of ^{35}S into organic forms, and this was observed after laboratory incubations (Strick et al. 1982; Fitzgerald et al. 1983; Swank et al. 1984). Although mineral soils of the Coweeta watersheds have a large capacity to adsorb inorganic sulfate (Johnson et al. 1980), the low percentage of sulfate extracted from samples on watershed 18 suggests that inorganic S is rapidly taken up and converted to organic forms *in situ*. Moreover, the low levels of phosphate-extractable organic sulfur found in these samples is consistent with the observation (Fitzgerald et al. 1982) that relatively small amounts of soluble organic S could be extracted with salt after laboratory incubation of samples with ^{35}S-labelled sulfate. Collectively, these results suggest a stepwise process involving formation from sulfate S of low molecular weight organic S components with subsequent polymerization of these to form a larger insoluble matrix.

Examination of extracts by electrophoresis allows the separation of the organic S forms into which sulfate was incorporated. Sulfur incorporated into organic matter that was only extractable after acid hydrolsis or alkaline oxidation can be characterized on the basis of bond type by referring to the electrophoretic patterns

TABLE 3. Fate of sulfate ^{35}S after field incubation for 2 and 7 days with forest floor and soil

Incubation time and horizon	Recovered form of ^{35}S (% of total extractable ^{35}S)						
	Water-soluble		Salt-extractable		Acid-extractable		Alkali-extractable,
	Organic S	Sulfate	Organic S	Sulfate	Carbon-bonded S	Ester sulfate[a]	carbon-bonded S
2 days							
O1	1.46	2.83	1.97	23.21	21.89	32.21	15.44
O2	2.04	4.84	5.86	38.40	24.30	16.37	8.20
A	0.55	0.95	0	80.60	15.08	0	2.82
7 days							
O1	1.65	2.16	3.40	18.38	39.49	26.19	8.74
O2	1.40	3.51	1.11	33.09	19.62	34.74	6.55
A	0.57	1.09	0	69.66	23.58	0	5.10

[a]Quantitated from the amount of $^{35}SO_4^{2-}$ released after acid hydrolysis.

TABLE 4. *In situ* incorporation of sulfate S into insoluble organic matter

Horizon	2-day incubation		7-day incubation	
	SO_4^{2-} S available[a] (mg kg^{-1})	S incorporated* (mg S kg^{-1})	SO_4^{2-} S available (mg kg^{-1})	S incorporated* (mg S kg^{-1})
O1	86.9	61.3+1.5 (70.54)	177.2	131.9±3.1 (74.44)
O2	25.0	12.2±0.3 (48.80)	16.1	9.8±0.4 (60.87)
A	23.0	4.1±0.3 (17.83)	32.3	9.3+0.8 (28.79)

[a]Calculated from values shown in Table 2.
[b]Values are means ± SE (N = 8). The amount of S incorporated as a percentage of the amount of sulfate S available is given in parentheses.

of these extracts. After extraction of samples with 1.0 *M* salt solution, all free and adsorbed sulfate will be removed. Any ^{35}S-labelled sulfate generated from acid hydrolysis following salt extraction originates from the hydrolysis of organic ester sulfate linkages. Therefore, ^{35}S-labelled sulfate in the acid hydrolyzate represents the relative ester sulfate content of the organic S recovered during acid treatment. Since all sulfate from ester sulfate was removed from the samples after hydrolysis, the remaining organic S was likely present as carbon-bonded S because of the extreme stability of the C—S linkage to acid (Fitzgerald 1976). Sulfate released by this subsequent alkali extraction may result from the oxidation of carbon-bonded S linkages.

Referring again to Table 3, the proportion of ^{35}S-labelled organic sulfur that was present in a water- or salt-extractable form remained low and did not vary substantially as field incubation time increased. The amount of water-soluble sulfate present also did not vary appreciably over time. However, the amount of ^{35}S incorporated into the acid-alkali extractable organic S increased with a corresponding decrease in adsorbed sulfate levels. From data based on laboratory incubations, Strickland and Fitzgerald (1984) suggested that inorganic sulfate may be incorporated into low molecular weight organics, which, subsequently, might polymerize to form larger, insoluble organic matrices. The data obtained from field incubations suggest that such a formation process does indeed occur *in situ* and that this process ultimately draws sulfate from the adsorbed sulfur pool. This would mean that during steady-state conditions (i.e., no precipitation and corresponding sulfate input), the sulfate adsorption–desorption equilibrium, specifically the rate of desorption, is an important rate-limiting process in organic sulfur formation. In addition to desorption, the rate of sulfate removal by biological uptake would exert a secondary regulatory effect on the organic S formation process. During periods of high biological activity, the desorption rate would play the major regulatory role.

One of the most pertinent questions asked of laboratory studies is the validity of extrapolating results to field conditions. Fitzgerald et al. (1983) reported levels of sulfate incorporation into acid-alkali extractable organic matter to be 49.5, 26.4, and 15.1% of added ^{35}S after 48 h in laboratory incubations at 20°C for the O1, O2, and A horizons, respectively. Although laboratory and field assays were performed in different years and, in the case of laboratory incubations, on samples collected in July instead of September, *in situ* incubation yielded incorporation levels of 70.5, 48.8, and 17.8% of the available sulfate after 48 h in the same respective horizons. If anything, these results suggest that the laboratory investigations may underestimate watershed organic S formation potentials. During the 5-day interval between the 2- and 7-day field incubations, there was a period of intense rainfall that increased the available sulfate levels *in situ*. Nevertheless, the relative proportion of ^{35}S incorporated into organic matter increased under these conditions by 4, 12, and 11% in the O1, O2, and A horizons, respectively. This capacity to respond positively to elevations in sulfate input was predicted by results of laboratory investigations reported by Swank et al. (1985). Results presented here indicate that laboratory studies do provide realistic views of S incorporation processes occurring in an ecosystem. Although these studies must rely on the artificial production of ecosystem conditions and although it is impossible to recreate all the interrelating variables of a system in the laboratory, these studies do lend important insight into the process-level parameters and regulatory mechanisms in a system.

Acknowledgements

The authors would like to thank Jam T. Ash and Dorothy D. Hale for expert technical assistance. In addition we thank Mary E. Watwood and W. T. Scott Swank for sampling assistance. This research was supported by the National Science Foundation (grant BSR-82-15259) and through a cooperative agreement with the United States Department of Agriculture, Forest Service.

DAVID, M. B., M. J. MITCHELL, and J. P. NAKAS. 1982. Organic and inorganic sulfur constituents of a forest soil and their relationship to microbial activity. Soil. Sci. Soc. Am. J. 46: 847-852.

DAY, F. P., and C. D. MONK. 1974. Vegetation patterns on a southern Appalachian watershed. Ecology, 55: 1064-1074.

FITZGERALD, J. W. 1976. Sulfate ester formation and hydrolysis: a potentially important yet often ignored aspect of the sulfur cycle of aerobic soils. Bacteriol. Rev. 40: 698–721.

FITZGERALD, J. W., J. T. ASH, T. C. STRICKLAND, and W. T. SWANK. 1983. Formation of organic sulfur in forest soils: a biologically mediated process. Can. J. For. Res. 13: 1077-1082.

FITZGERALD, J. W., T. C. STRICKLAND, and W. T. SWANK. 1982. Metabolic fate of inorganic sulphate in soil samples from undisturbed and managed forest ecosystems. Soil. Biol. Biochem. 14: 529-536.

FRENEY, J. R. 1958. Determination of water soluble sulfate in soils. Soil Sci. 86: 241-244.

———1961. Some observations on the nature of organic sulphur compounds in soil. Aust. J. Agric. Res. 12: 424-432.

FRENEY, J. R., G. E. MELVILLE, and C. H. WILLIAMS. 1969. Extraction, chemical nature and properties of soil organic sulfur. J. Sci. Food Agric. 20: 440-445.

———1970. The determination of carbon bonded sulfur in soil. Soil Sci. 109: 310-318.

———1971. Organic sulphur fractions labelled by addition of ^{35}S-sulphate to soil. Soil Biol. Biochem. 3: 133–141.

HEINRICHS, H., and R. MEYER. 1977. Distribution and cycling of major and trace elements in two central European forest ecosystems. J. Environ. Qual. 6: 402-407.

JOHNSON, D. W., D. W. COLE, and S. P. GESSEL. 1979. Acid precipitation and soil sulfate adsorption properties in a tropical and temperate forest soil. Biotropica, 11: 38-42.

JOHNSON, D. W., and G. S. HENDERSON. 1979. Sulfate adsorption and sulfur fractions in a highly weathered soil under a mixed deciduous forest. Soil Sci. 128: 34-40.

JOHNSON, D. W., G. S. HENDERSON, D. D. HUFF, S. E. LINDBERG, D. D. RICHTER, D. S. SHRINER, D. E. TODD, and J. TURNER. 1982. Cycling of organic and inorganic sulphur in a chestnut oak forest. Oecologia, 54: 141-148.

JOHNSON, D. W., J. W. HORNBECK, J. M. KELLY, W. T. SWANK, and D. E. TODD. 1980. Regional patterns of soil sulfate accumulation: relevance to ecosystem sulfur budgets. *In* Atmospheric sulfur deposition: environmental impact and health effects. *Edited by* D. S. Shriner, C. R. Richmond, and S. E. Lindberg. Ann Arbor Science Publishers Inc., Ann Arbor, MI. pp. 507-520.

JOHNSON, C. M., and N. NISHITA. 1952. Microestimation of sulfur in plant material, soil, and irrigation waters. Anal. Chem. 24: 736-742.

JONES, L. H. P., D. W. COWLING, and D. R. LOCKYER. 1972. Plant available and extractable sulfur in some soils of England and Wales. Soil Sci. 114: 104-114.

LOWE, L. E. 1964. An approach to the study of the sulphur status of soils and its application to selected Quebec soils. Can. J. Soil. Sci. 44: 176-179.

MCGILL, W. B., and C. V. COLE. 1981. Comparative aspects of cycling of organic C, N, S and P through soil organic matter. Geoderma, 26: 267-286.

SAGGAR, S., J. R. BETTANY, and J. W. B. STEWART. 1981. Sulfur transformations in relation to carbon and nitrogen in incubated soils. Soil Biol, Biochem. 13: 499–511.

SHRINER, D. S., and G. S. HENDERSON. 1978. Sulfur distribution and cycling in a deciduous forest watershed. J. Environ. Qual. 7: 392-297.

STRICK, J. E., S. C. SCHINDLER, M. B. DAVID, M. J. MITCHELL, and J. P. NAKAS. 1982. Importance of organic sulfur constituents and microbial activity to sulfur transformations in an Adirondack forest soil. Northeast. Environ. Sci. 1: 161–169.

STRICKLAND, T. C., and J. W. FITZGERALD. 1984. Formation and mineralization of organic sulfur in forest soils. Biogeochemistry, 1: 79-95.

SWANK, W. T., and J. E. DOUGLASS. 1977. Nutrient budgets for undisturbed and manipulated hardwood forest ecosystems in the mountains of North Carolina. *In* Watershed research in eastern North America. Vol. 1. *Edited by* D. L. Correll. Smithsonian Institution Press, Washington, DC. pp. 343-363.

SWANK, W. T., J. W. FITZGERALD, and J. T. ASH. 1984. Microbial transformation of sulfate in forest soils. Science (Washington, D.C.), 233: 182-184.

SWANK, W. T., J. W. FITZGERALD, and T. C. STRICKLAND. 1985. Transformations of sulfur in forest floor and soil of a forest ecosystem. *In* Hydrological and hydrogeochemical mechanisms and model approaches to the acidifation of ecological systems. International Hydrological Programme Workshop, Uppsala Report No. 10. *Edited by* I. Johansson. Coordinating Committee for Hydrology, Norden, Sweden pp. 137-145.

TABATABAI, M. A., and J. M. BREMNER. 1970. An alkaline oxidation method for determination of total sulfur in soils. Soil Sci. Soc. Am. Proc. 34: 62-65.

———1972. Forms of sulfur and carbon, nitrogen and sulfur relationships in Iowa soils. Soil Sci. 114: 380-386.

WILLIAMS, C. H., and S. STEINBERGS. 1959. Soil sulphur fractions as chemical indices of available sulphur in some Australian soils. Aust. J. Agric. Res. 10: 340-352.