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EFFECT OF AMMONIUM NITRATE ON THE Sr90
CONCENTRATION IN TOMATO ORGANS

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Author's name and affiliation:

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TABLE 1
Selected properties of experimental soils

Soil No.	pH 1:1 Suspend- sion	E. C.	Lime as CaCO ₃ *	Organic Matter*	Total Nitrogen	NO ₃ Nitrogen	CEC	Exchange- able Na	Texture		
		Sat. Extr.							Sand	Silt	Clay
		mmhos./ cm.	%		ppm		m. e./100 g		%		
7	5.1	0.16	-	0.35	263	2.6	14.4	0.13	39.7	23.1	37.2
5	8.7	0.65	1.09	0.86	88.3	0.48	6.4	0.30	89.5	4.7	5.9
1	7.8	2.47	9.0	1.9	782	66	11.0	5.90	39.4	31.0	29.6
10	7.9	1.76	18.2	1.9	585	5.1	12.5	0.20	46.0	36.5	17.6
4	8.6	0.65	33.2	0.45	125	3.0	7.6	0.40	76.3*	15.1*	8.9*

* Analyses provided by H. Nishita (10)

TABLE 2

Mean squares and probabilities ($P <$) from analysis of variance upon organ yields (weight, Sr90), relative distribution of plant (weight, Sr90) among organs, shoot to root (weight, Sr90) ratios, and concentration of Sr90 in leaves, stems and roots of tomato plants, from soil in pots.

Source of variation	df	Leaves		Stems		Roots		Leaves		Stems		Roots	
		Mean square	P<	Mean square	P<	Mean square	P<	Mean square	P<	Mean square	P<	Mean square	P<
Organ weight, g (dry weight)						Organ Sr90, μ c.							
Nitrogen rate (N)	3	.311	NS	.326	.001	.016	NS	.435	NS	.150	NS	.028	NS
Soils (S)	4	.609	.001	.329	.001	.162	.001	.781	.005	.522	.001	.074	.001
Interaction (NxS)	12	.243	.001	.135	NS	.031	NS	.584	.005	.122	NS	.014	NS
Residual	38	.062	-	.049	-	.018	-	.168	-	.056	-	.007	-
Relative organ weight, % of plant g						Relative accumulation, % of plant Sr90							
Nitrogen rate (N)	3	.012	NS	.009	NS	.008	NS	.010	NS	.007	NS	.001	NS
Soils (S)	4	.059	.001	.005	NS	.044	.001	.034	.005	.023	.005	.018	.001
Interaction (NxS)	12	.013	.001	.013	.001	.004	NS	.023	.005	.014	.005	.002	NS
Residual	38	.003	-	.003	-	.002	-	.006	-	.004	-	.001	-

(cont'd)

(Table 2, cont'd)

		Shoot/Root ratios									
		Mean square	P <	Mean square	P <						
		g		µc.		Sr90 concentration, µc./g					
Nitrogen rate (N)	3	3.86	NS	23.42	NS	.887	.001	.471	.001	.103	.005
Soils (S)	4	28.71	.001	334.96	.001	.579	.001	.163	NS	.075	NS
Interaction (N x S)	12	1.28	NS	21.83	NS	.232	.001	.049	NS	.046	NS
Residual	38	.97	-	9.33	-	.053	-	.041	-	.020	-

* Numbers rounded after F-ratios were calculated; NS - Probability of chance occurrence less than 0.005

Footnotes:

- ¹ These studies were supported by Contract AT(04-1) GEN-12 between the U. S. Atomic Energy Commission, Division of Biology and Medicine, and the University of California.

I am indebted to Dr. K. H. Lu for statistical advice, to J. F. Irwin for technical assistance, to S. D. Zellmer for programming, and Dr. R. H. Moore for contributing through a stimulating discussion.

- ² Sr90 was obtained from Oak Ridge National Laboratory, Oak Ridge, Tenn., in HCl and held one year to reduce any short-lived radioactive impurities.
- ³ Neel, J. W. et al. 1953. Soil-plant interrelationships with respect to the uptake of fission products. I. The uptake of Sr90, Cs137, Ru106, Ce144 and Y91. UCLA-247. University of California, Los Angeles.

FIGURE LEGENDS

Figure 1. Sr90 concentrations in tomato plant leaves, stems, and roots after four nitrogen fertilization rates were applied to four calcareous soils and to one acid soil (No. 7).

Figure 2. Dry weights of tomato plant organs after 0, 100, 200 and 400 parts per million nitrogen applications to four calcareous and one acid soil (No. 7), with the percent Sr90 contributions of each organ to plant Sr90 (lower row).

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EFFECT OF AMMONIUM NITRATE ON THE Sr90 CONCENTRATION OF TOMATO ORGANS

Global distribution of radioactive debris from nuclear detonations is well documented, so are the reasons for watching the environmental 'behavior' of Sr90. Pragmatic interest should accrue to any production practice that may alter Sr90 transfer rates from the soil reservoir into mans' food chains. Various experimental methods to inhibit root uptake were devised and tested, but little effort is expended upon the effects contemporary field practices may have on Sr90 uptake by cultivars.

A recent technological development in meat and milk production is the application of nitrogen fertilizers at very high rates to enhance the quality of plants used in animal feeding programs, without expecting incremental increases in crop yield. These much higher applications of ammonium nitrate, a preferred nitrogen vector, inject ammonium into the fertilized zone at rates close to symmetry concentration for sandy soils. No elaboration is needed about the effectiveness of ammonium in solution for displacing cations from exchange sites in agricultural soils.

Cation exchange theory, and empirical verification, show plant uptake of Sr90 from neutral soils and fertilized, acid soil inversely proportional to exchangeable calcium; comparable guidelines for system models of calcareous soils are lacking. These soils, by definition, have soil-solution calcium at equilibrium with mineral calcium carbonate. Such soils may compensate similarly for reactions that shift the concentration of calcium in solution until all the mineral carbonate is dissolved. The amount of free calcium carbonate present is not relevant while any remains undecomposed.

This experiment was designed to test the hypothesis that plant uptake of tracer-level Sr90 is similar from calcareous soils ^{with} ~~or~~ free calcium carbonate.

METHODS AND MATERIALS

Five mineral soils were selected to provide a wide range in calcium content and in salt concentration of the saturated-soil extract. All of the soils were from sites previously used for cattle range, but one was in irrigated agriculture 28 years (No. 1). Some of the chemical and physical properties of the soil fractions passed by a 2-mm. round hole sieve are indicated in Table 1, and are the fractions used experimentally.

The experimental design was a randomized complete block (6) with three replications of four nitrogen rates on five soils. The nitrogen rates were additions at 0, 100, 200 and 400 parts per million (ppm.). A standard computing program was used for the analysis of variance (3).

Air-dry soil was weighed out in 4500-g lots from stock. Each lot was quartered repeatedly and separated into three parts. An amount equivalent to 3500 g, on an oven-dry basis, was removed and one-seventh spread an inch deep on waxed paper. Carrier-free $\text{Sr}90^2$ tracer solution was sprinkled over the soil from a 10 ml. pipette and air-dried 10 days. The radioactive aggregates formed in the wetted soil were reduced by hand and incorporated into the 500 g aliquot, which was mixed with the 3 kg remainder to obtain a $\text{Sr}90$ dose of 57 $\mu\text{c}/\text{kg}$ soil. The entire 3500 g was batch-mixed in a V-chambered McClellan mixer 30 minutes and spread onto a protected bench for fertilization. The remaining uncontaminated kg was held for analytical determinations.

Commercial ammonium nitrate 'prills' were dried 60 minutes at 105°C , weighed, hand-pulverized, and incorporated into contaminated soil by hand according to the nitrogen treatment being instituted. The nitrogen content of the dry 'prills' was 35.55%.

The Sr90-traced, fertilized soil was put into tared, cylindrical, metal, plastic-lined (Amercoat Corp. No. 33) containers 16.7 cm. in diameter by 19.0 cm. high. The soils were wet slightly in excess of 0.1 bar moisture content to assure penetration, were held covered for 24 hours, then allowed to dry by evaporation 10 days until use.

Tomato plants (Lycopersicon esculentum Mill. 'Pearson Improved') were used as the extraction agent for Sr90. These maintain a continuous stress upon salts in the soil solution by their indeterminate, vegetative growth habit. This characteristic was exploited to circumvent the progressive changes in the Sr90 concentration of plants resulting from physiological maturation, as observed in early radionuclide studies³.

Seedlings were started at midsummer in flats of sand, irrigated with Hoagland solution, cultured in a smog-free glasshouse for seven weeks, and put into a growth chamber (14). On the third day they were weighed, grouped by weight in bundles of three until 30 bundles were collected. The two largest and the two lightest bundles were discarded, and positions within treatments were assigned to bundles from random numbers, as was replicate status to single plants. These were transplanted to contaminated soil, which was brought to 0.1 bar moisture content by weight (10). They were kept at 22°C continuously, under 16-hour daylength to promote vegetative growth (7, 9) and harvested after 36 days. The containers were weighed frequently, and irrigated with glass-distilled water as they approached 1.0 bar moisture tension.

Shoots were separated into leaves (including petioles) and stems, dried 24 hours at 70°C, weighed, ground in a Wiley mill to pass a 40-mesh screen, and five counting replicates were weighed out in 50-mg quantities to three significant digits. Soil in the containers was brought to

saturation, covered four hours, and the roots were carefully removed manually. The intact root systems were washed in tap water, rinsed 10 seconds in 0.1 N HCl (12), washed in distilled water, blotted dry and processed by drying, weighing, and grinding. Counting was done after secular equilibrium was reached between Sr90 and Y90, on a beta counting system having a window density of $150 \mu\text{g}/\text{cm}^2$ (Nuclear-Chicago, Model C115 Low Background). Counting times were varied to maintain less than 2% counting error at 0.95 confidence level. Corrections were made for backscatter, geometry, and plant background radioactivity. Plant backgrounds were obtained from plants grown in uncontaminated soil, but otherwise identical to experimental units and within the randomized complete blocks.

Standard methods were used to characterize soil properties. Nitrate-nitrogen, soil suspension pH, and exchangeable sodium analyses followed Jackson (8). Sodium was determined, after soil extracts were digested in perchloric acid, by flame photometer (Beckman, Model DU). Cation exchange capacity determinations were those adapted to calcareous soils (1); Salinity Laboratory methods (11) were used for mechanical analysis and saturation extract conductivity, determined by conductivity bridge (Industrial Instruments, Model RC-12C1P). Total-nitrogen of soils was determined with combustion apparatus (Coleman, Model 29A). One hundred-g aliquots of soil were treated with C. P. grade KNO_3 or NH_4NO_3 solution, made from fertilizer 'prills', to obtain the nitrogen levels shown in Figure 1, were air-dried, and brought to saturation moisture content. They were equilibrated overnight before extraction and determination of the conductivity of the saturation extracts.

RESULTS AND DISCUSSION

Saturation extracts from soils fertilized with potassium nitrate, in general, had higher specific conductance than their counterparts fertilized with ammonium nitrate. Differences observed were less than 10 percent. Chapin et al. (2) reported similar results using calcareous soils fertilized with both nitrate salts.

Each increment of fertilizer added to the alkali soil (No. 1) reduced plant growth rates. Addition of nitrogen at 200 ppm. reduced the dry weights of plants from this soil by 50 percent, compared to the weights of control plants. Plants treated with 400 ppm. nitrogen showed obvious, apparently irreversible, damage at harvest. The electrolyte concentrations of saturation extracts from fertilized alkali soil were more than 8 atmospheres, calculated in osmotic pressure (11). These specific conductances may indicate that the saline soil solutions created were calcium-saturated, under the solvent action of the neutral salts generated from ammonium nitrate upon precipitated calcium minerals.

Osmotic pressures of saturation extracts from the remaining calcareous soils were less than 0.8 atmospheres, and those from the acid soil (No. 7) less than 0.2 atmospheres. Nevertheless, additions of nitrogen at 200 ppm. reduced the mean dry weights of plants as compared to those fertilized with 100 ppm. nitrogen ($P < .001$ computed from residual mean squares as a standard error of a difference).

Statistical criteria

Positive statements about the results of treatments imply statistical significance at the 99.5% confidence level ($P < .005$), unless the 99.9% confidence level is stated ($P < .001$). All the data for dependent variables

in Table 2 were interpreted by analysis of variance for randomized complete blocks. The mean squares, with their confidence levels based on F-ratios, were partitioned into appropriate sources of variation.

Significant interaction terms constitute a priori proof that the basic analysis of variance presumption was violated, i.e., treatment effects are additive. Dependent variables, for which the analysis of variance had significant interaction terms, were subjected to various transformations, including square root, $\log_{10}$, $\log_e$ and $\log_2$, without diminishing the significance of the interaction term. This is in agreement with Scheffe (15), that transformations are ineffective in changing interaction when one factor is quantitative and the other is qualitative, as in this experiment.

Significant interaction in the analysis of variance with non-significant nitrogen effects verifies that significant but non-additive differential plant response to nitrogen occurred among cells within the treatment matrix. On the average, these significant nitrogen effects cancelled one another across the soils and were specific to a nitrogen-soil treatment for direction and magnitude. Those dependent variables with statistically significant interaction (nitrogen x soils) in the analysis of variance are dealt with individually as they occur in the text.

Concentrations of Sr90

The mean effect of increasing nitrogen applications was an increase in the concentration of Sr90 in roots and stems (Figure 1), but the interaction term for leaves was high ($P < .001$, Table 2) and nullified this analysis of variance for randomized complete blocks.

Nitrogen effects on the Sr90-concentration of leaves were interpreted by one-way analysis of variance within each soil. There were no appreciable

differences in the Sr90 concentration of leaves from plants on the four soils with free CaCO_3 (Figure 1), although upward trends were present. The difference in Sr90 concentration of leaves from plants grown in unfertilized, acid soil (No. 7) and leaves from the 100 ppm. nitrogen rate was highly significant ($P < .001$).

Leaves from plants grown in calcareous soils had Sr90 concentrations that were between those of leaves from plants in acid soil treated at 0 or at 100 ppm. nitrogen additions. It was unexpected for both the minimum and maximum concentrations of Sr90 to occur in plants from acid soil. Concentrations of Sr90 in tomato plant organs were: leaves > stems > roots, in the approximate ratios 5 : 3 : 2.

The minimum concentration of Sr90 in leaves occurred in plants from acid soil (calcium-saturation of the exchange complex was less than 20 percent). In view of these results, some question about the high uptake of strontium reported for plants grown in acid soil seems justifiable. Reports in the literature cite acid soils as having plant-available strontium in high concentrations, on both experimental and theoretic grounds. Pretreatment fertilization of acid soil "in sufficient amounts to ensure normal development of the plants" seems universal. In many experiments reported, it seems likely that pretreatment fertilization effects were confounded with effects from the treatments imposed. In essence, prefertilization invalidated the acid soil "control" in those experiments. In an experiment, where the treatments were depth of Sr90 placement, higher concentrations of plant Sr90 resulted where fertilizer and Sr90 tracer were combined, compared to those where each was installed separately (4). That finding is in accord with these suggestions. There is a high probability that some conclusions about plant uptake of strontium from nitrogen-fertilized, acid soil apply to an artifact of soil management rather than to the properties of acid soil per se.

Plant yield

Analysis of variance for roots (Table 2) showed that differences in dry weights were randomly distributed across the treatment matrix and were soil-dependent. Root weights increased with nitrogen increases in calcareous soil (No. 5) and concurrently decreased in saline soil (No. 1, Figure 2). Stem tissue was adversely affected by high nitrogen rates and the differences in weight between control and 200 ppm. was substantial ($P < .001$ calculated from residual mean squares).

The high interaction term for dry weights of leaves (Table 2) invalidates the analysis of variance. These effects of nitrogen on leaf weight were not additive but had resulted in differential weight increments, soil by soil (Figure 2). The effects of nitrogen increases upon saturation extract conductivity were discussed, in terms of plant growth rates for the salinized, alkali soil. Those comments apply, in general, to the weight of leaves from plants on this soil (r^2 , the coefficient of determination, was 0.833 for leaf to plant weights).

Interactions for percent of plant in leaves and in stems were high ($P < .001$) but the mean effect of nitrogen on the percent of plant in roots was randomized. Alkali soil produced plants with the lowest percent of weight in roots, and had the highest soluble nitrate content (Table 1); these seem related. Ratios of shoot to root weights were insensitive to nitrogen applications on the average, but were influenced greatly by which soil was plant substrate ($P < .001$). The range among soils in mean shoot/root weight ratios was from 2.5 for plants from acid soil (No. 7) to 6.3 for plants from alkali soil (No. 1).

Distribution of Sr90 among organs

Ratios for shoot/root Sr90 followed the same pattern as the weight ratios. Nitrogen level had little effect on the average Sr90 ratio, and substrate soil determined the ratio (Table 2). Mean Sr90 shoot/root ratios by soils were similar to weight ratios, though approximately three times larger, i.e., from 6.5 in plants on acid soil to 18.3 for plants on alkali soil. Differential translocation of Sr90 to the shoots, under the influence of nitrogen, was of small consequence in this study.

Nitrogen additions had little effect on the rate of accumulation of Sr90 by roots and stems; again the dominant response was that induced by the particular soil from which the plants absorbed Sr90 (Table 2). The high interaction term for accumulation of Sr90 by leaves contradicts the verdict of non-significant response projected by the analysis of variance. Accumulation of Sr90 in leaves varied within soils, was minimal in those from plants grown on unfertilized acid soil, and was maximum in plants on unfertilized alkali soil. The relative amounts accumulated by organs were: leaves > stems > roots, in the approximate ratio 9 : 5 : 2, respectively.

Internal distributions of Sr90 within individual plants were expressed as percent of plant uptake of Sr90, collated in replications, then averaged by cells of the treatment matrix (Figure 2). The percent of plant Sr90 held in roots on the average was not affected by nitrogen application (Table 2). High interaction terms for the percent of plant Sr90 in stems and in leaves requires the interpretation accorded the rate of Sr90 accumulation by leaves.

The awkwardness in dealing with large interaction levels encourages use of methods that treat plant organs as integral parts of a single entity, the plant.

Loss of statistical sensitivity to accommodate large mean squares for interaction is a further encouragement to adopt unitized analytic methods of data evaluation for a plant from data about its exfoliated organs. Intense shrinkage of statistical sensitivity occurred at the partition of the analysis of variance into multiple sub-analyses, one for each soil in the treatment matrix. In degrees of freedom for residual error, the drop in sensitivity was from 38 degrees per dependant characteristic to 8 degrees per soil. Such arrays of data from excised organs, where interpretive information is elicited by handling every soil-organ combination as a separate statistical problem, fractionate the yield of information. This separate but equal statistical structure pretends that leaves grew in pots apart from stems or roots. An additional analytic method is needed to ascertain how a characteristic under scrutiny interacts from organ to organ along the system named "plant". Lumping observations across several organs to acquire a summation, i. e. plant $\bar{g}$ or plant μ_c , disowns the central issue of concurrent but differential response among organs of a plant.

Data from this experiment that could be handled by regression analysis is limited. It is confined to assessments "within plants". These are the measurements made upon the dependant variables, organ by organ. Variance within the plant could be partitioned into larger-than, smaller-than rank among organs through partial correlation coefficients of multiple regression, or accounted for as the percent of concurrent variation by organs in the multiple coefficient of determination, but these are not measures of the contributions of the separated organs to the variance of the "intact" plant.

Soils are a qualitative variable and are not accessible as independant variables for regressions. Numerical ratings are demanded from soils as input for regression analysis, and these are not available.

SUMMARY

Four calcareous and one acid soil were contaminated with Sr90, fertilized with ammonium nitrate, and used as substrate for tomato plants in a growth chamber. The free CaCO_3 content of the soils ranged from 0 to 33 percent; ammonium nitrate levels were 0, 100, 200, and 400 parts per million, as nitrogen.

Significantly lower ($P < .001$) Sr90 concentrations occurred in leaves of plants grown in unfertilized acid soil compared to those from acid soil fertilized with 100 parts per million nitrogen; these two nitrogen levels and soil resulted in plants with leaves having the minimum and the maximum observed concentrations of Sr90. Plants grown in calcareous soils, fertilized or unfertilized, had leaves with intermediate Sr90 concentrations.

The concentration of Sr90 in stems was increased ($P < .001$) by increasing rates of nitrogen fertilization, as was the concentration in roots ($P < .005$). The concentrations in the plants were leaves $>$ stems $>$ roots in the approximate ratio of 5 : 3 : 2, respectively.

The amounts of Sr90 recovered from soils were variable because plant growth rates and tissue concentrations of Sr90 were confounded, as shown by the high interaction term (nitrogen x soil) in the analysis of variance ($P < .005$).

The minimum concentrations of Sr90 occurred in plants from unfertilized acid soil, instead of in plants from high-calcium soils. This observation suggests that Sr90 studies, in which acid soils were fertilized with nitrogenous materials before experimental treatments were instituted, may benefit from reassessment. The conclusions drawn from such studies may apply to an artifact of soil management, rather than to acid soils per se.

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LEAVES

STEMS

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