levels, but this fluorine was not in the form of fluorinecontaining organic acids; total fluorine in the culture solution decreased rapidly, possibly by volatilization of the amide, the free acid after hydrolysis, or the excreted metabolic product. Horiuchi proposed fluoromethane as a possible plant metabolite, and presented evidence that the C-F bond was not broken to release inorganic fluoride. Soil microorganisms, probably gram-negative bacteria, broke the C-F bond to form inorganic fluoride.

There appears to be no evidence for the formation of monofluorocitric acid in plants. Baker's yeast formed fluorofatty acids, probably butyric and hexanoic, but no fluorocitrate (Aldous, 1963).

Several authors have reviewed the effects of inorganic fluoride on plants and soils (Bartholomew, 1935; Bollard and Butler, 1966; Lundegårdh, 1966), and the amount of available  $F^-$  in soil for uptake by the bean plant has been measured (Applegate *et al.*, 1960).

## EXPERIMENTAL

Foliar Absorption. Four-month-old sugarcane plants of variety H 49-5 were treated with 0.5 ml. of ammonium monofluoroacetate-2-<sup>14</sup>C solution, supplying 50  $\mu$ g. of F or 250  $\mu$ g. of the salt. (The solution was obtained from International Chemical and Nuclear Corp. as a concentrated aqueous solution of 5 mc. per mmole.) The solution was applied to the leaf axil (dewlap) of leaf 3 (counting from the top); excess liquid ran into the adjacent dewlap area of leaves 4 and 5. Laminar tissue samples 0.5 inch wide by 4 inches long, beginning one fourth of the distance from the leaf tip to the axil, were cut periodically for radioactive assay. These samples, and portions of the entire plant taken at the end of a 1-month period, provided the data in Tables I and II. All counts were made with a GM gas flow counter.

**Root** Absorption. Ammonium monofluoroacetate-2-<sup>14</sup>C was added to an aerated standard nutrient solution containing eighteen 4-month-old sugarcane plants, variety H 49-5, to provide a total of 1000  $\mu$ g. of F (5000  $\mu$ g. of the salt). An additional 26,400  $\mu$ g. of unlabeled sodium monofluoroacetate (5000  $\mu$ g. of F) were added to the nutrient medium. The plants remained in the radioactive

Table	I.	Radio	activity	of <i>v</i>	Sugarc	ane	Leaf	Lamin	na a	nd
Sheath	Se	ctions	after A	Appl	ications	of	Ammo	nium	Mor	10-
flu	ioro	acetat	e-2-14C	to A	xils of	Lea	ves 3,	4, and	5	

Interval.	Original Leaf	Final Leaf	Radioactivity, Net C.P.M./G. Dry Weigł	
Days	No.	No.	Lamina	Sheath
2	3	3	636	
10	3	4	2,071	• • •
16	Spindle	1	6,734	
30		Spindle	3,407	
30		1	2,738	647
30	Spindle	2	11,316	798
30	1	3	1,012	1,960
30	2	4	3,146	18,608
30	3	5	556	40,935
30	4	6	436	33,243
30	5	7	1,310	22,213

solutions for 35 days, with water added as necessary. They were then placed in fresh nutrient without radioactivity until completion of the 3-month experiment. Control plants were grown in aerated nutrient solution. Radioactive assay of the culture solutions with time is shown in Table III; sugarcane samples taken at 0.5, 1, 1.5, and 3 months were counted as separate chopped dried portions of leaf, stalk, and root. These data are shown in Figures 1, 2, and 3.

## **RESULTS AND DISCUSSION**

Foliar Application. Sugarcane leaves absorbed monofluoroacetate-2-<sup>14</sup>C ion and translocated a small propor-

Table II.	Radioactivity of Chopped Dried Portions of
Sugarca	ne Plants, One Month after Application of
Ammo	nium Monofluoroacetate-2-14C to Axils of
	Leaves 3, 4, and 5

Plant Part	Radioactivity, Net C.P.M./G. Dry Weight
Spindle	2402
Leaf 1	
Total laminar tissue	2596
Midrib	1036
Sheath	647
Leaf 2 (original spindle)	
Total laminar tissue	3931
Midrib	2539
Sheath	798
Growing point	1484
Upper stalk <sup>a</sup>	3422
Lower stalk <sup>a</sup>	1267
Bagasse fiber from upper stalk	3385
Bagasse fiber from lower stalk	1379
Juice from upper stalk	5036 <sup>b</sup>
Juice from lower stalk	1912
Secondary shoots (suckers)	
Secondary A (26 g. fresh wt.)	
Lamina, leaf 1	737
Midrib, leaf 1	444
Sheath, leaf 1	Trace
Growing point plus stalk	167
Secondary B (1.4 g. fresh wt.),	
entire plant	4279

<sup>a</sup> Upper stalk portion contains green leaves, lower stalk contains dried senescent leaves.

<sup>b</sup> Net c.p.m./ml. of expressed juice reduced to dryness.

## Table III. Radioactivity from Ammonium Monofluoroacetate-2-<sup>14</sup>C Added to Nutrient Solution Containing Sugarcane Plants

Interval, <sup>a</sup>	Radioactivity, Net C.P.M./Ml.			
Days	Untreated control	Treated		
0.12		1122		
2		141		
4	12	154		
7	9	43		
10	14	24		
15	19	35		
28	11	15		
35	11	10		
$35 \pm 0.17^{5}$	0	0		
$35 + 7^{b}$	0	0		
$35 + 42^{b}$	0	0		
<sup>a</sup> pH ranged from 7.2 <sup>b</sup> Nutrient medium r ays.	2 to 7.8 during initial 35- eplaced with fresh unti	day period. eated nutrien		



Figure 1. Apparent change with time of radioactivity in plants grown in nutrient solution treated with ammonium mono-fluoroacetate- $2^{-14}$ C



Figure 2. Apparent change with time in distribution of radioactivity in various parts of plants grown in nutrient solution treated with ammonium monofluoroacetate- $2^{-14}$ C

tion of the radioactivity into new leaf tissue. Approximately 10% of the total radioactivity appeared in tissues younger than those of the application area; little translocation took place from the treated leaf sheath into the blade of the same or lower leaves (Table I). Uptake and movement of the applied radioactivity were restricted, by either poor absorption into the leaf or the high degree of adsorption to the sheath tissue. Most of the translocated activity appeared in the laminas of the younger, rapidly expanding leaves, with less activity in the midribs and relatively little in the leaf sheaths except at or near the point of origin (Table II). From the data it appeared that radioactivity either was translocated from older to younger



Figure 3. Apparent change with time of total radioactivity of plants grown in nutrient solution treated with ammonium monofluoroacetate- $2-^{14}C$ 

leaves after reaching a maximum in each leaf, or was continuously supplied from the source. The growing point did not show particularly high activity, but, in the grass plant, it is located well inside and below the expanding leaf tissue (spindle and first few leaves), although above the site of treatment in these experiments. Fiber and juice from younger stalk portions contained more radioactivity than fiber and juice from older tissue; secondary suckers from the base of the original plant were radioactive, with more activity in the youngest tissue.

In summary, a portion of the active carbon atom, probably with the attached fluorine atom, moved from the site of application in the leaf axil to the lamina of the new leaf, but with a high degree of retention at the treated site. The experiments did not indicate whether monofluoroacetic acid is the translocated substance, but it appears that the  $F^{-14}C$  bond remains in both plant and animal systems.

Root Application. Radioactivity in the nutrient medium containing growing sugarcane plants decreased rapidly with time (Table III). Only 10% of the original count remained after 5 days, and 1 % at the end of 35 days when the radioactive solution was replaced with fresh untreated solution. There was no apparent leakage of radioactivity from the treated plants into the fresh untreated solution. There is considerable evidence of volatility: Radioactivity in the plants is less than can be accounted for as loss from the nutrient; stock solutions of ammonium monofluoroacetate-2-14C or of nutrient solution with added ammonium monofluoroacetate-2-14C lost radioactivity on standing in open containers or on heating; and when the untreated control plants were placed in proximity to treated plants, the radioactivity of the control plants increased to an equilibrium level above background activity and then decreased. The pH range of 7.2 to 7.8 is not high enough to prevent hydrolysis and loss of the free acid.

Most of the radioactivity removed by plants from the nutrient solution remained in the roots (Figures 1 and 2). The early pattern of absorption from solution suggests a strong and relatively complete adsorption by the roots, with translocation of 5 to 8% of the total radioactivity into the

leaves and stalk. The radioactivity bound to the roots decreased rapidly after the source in the nutrient medium was depleted, but even after 3 months (including almost 2 in fresh untreated nutrient) the roots retained 60% of the radioactivity remaining in the plant. During the 3-month period the total apparent radioactivity of the plants decreased from an average of 150,000 to 12,000 c.p.m., with losses in all plant parts except the lower dead leaves. The above-ground portions decreased only from a total of 6700 c.p.m. at 15 days to 4900 at 90 days. During this time the weights of the treated plants increased 300%. It is possible that the losses of radioactivity in the leaf and stalkwhich appear to be approximately 27%-represent only increased self-absorption by the plant fiber. On the other hand, root losses were real, with a 45-fold decrease in counts per minute per gram for a growth weight increase of only twofold.

The data for root uptake suggest a strong adsorption of monofluoroacetate ion on the roots, with only minor translocation to the leaves and stem. This is followed by a slower desorption from the roots and by vaporization, with or without metabolism of the monofluoroacetate to more volatile compounds.

The high degree of adsorption of monofluoroacetate to leaf and root tissue, as well as to other cellulosics such as filter paper, was entirely unexpected in view of its water solubility and volatility. It can be assumed that monofluoroacetate would remain adsorbed to fibrous bait components, probably would not be washed off the bait formulations by moderate rainfall, and would not readily leach into soils, especially into those with considerable organic content.

## LITERATURE CITED

- Aldous, J. G., Biochem. Pharmacol. 12, 627 (1963).
- **47**, 339 (1960).
- Bartholomew, R. P., Soil Sci. 40, 203 (1935). Bollard, E. G., Butler, G. W., Ann. Rev. Plant Physiol. 17, 89 (1966)
- Horiuchi, N., Takamine Kenkyusho Nempo 7, 209 (1955).
- Horiuchi, N., Takamine Kenkyusho Nempo 9, 178 (1957). Horiuchi, N., Takamine Kenkyusho Nempo 11, 215 (1959).
- Horiuchi, N., Takamine Kenkyusho Nempo 12, 310 (1960).
- Horiuchi, N., Kumasawa, N., Takamine Kenkyusho Nempo 8, 215 (1956).
- Horiuchi, N., Yoshimura, N., Takamine Kenkyusho Nempo 10, 272 (1958).
- Lundegårdh, H., "Plant Physiology," pp. 253-60, 314, Elsevier, New York, 1966.
- Marais, J. S. C., Onderstepoort J. Vet. Res. 20, 67 (1944).
- Peters, R. A., *Proc. Roy. Soc. (London)* **B 139**, 143 (1952). Peters, R. A., *Proc. Roy. Soc. (London)* **B 140**, 497 (1953). Peters, R. A., *Endeatour* **13**, 147 (1954).

- Peters, R. A., Shorthouse, M., *Nature* **202**, 21 (1964). Ramsey, L. L., Clifford, P. A., *J. Assoc. Offic. Agr. Chemists* **32**, 788 (1949).

Received for review August 5, 1968. Accepted August 27, 1968. Published with the approval of the Director as Paper No. 210 of the Journal Series, Experiment Station, Hawaiian Sugar Planters' Association, Honolulu. Hawaii.