ROOT AND FOLIAR UPTAKE OF ^{134}Cs BY THREE TOBACCO PLANT VARIETIES

G. D. ARAPIS[1]

RÉSUMÉ. — *Prélèvement racinaire et foliaire de Césium (^{134}Cs) dans trois variétés de tabac.* — Le prélèvement racinaire de Césium (^{134}Cs) et sa distribution subséquente dans les pousses, fleurs et feuilles, de même que son dépôt direct sur ces parties de la plante, ont été étudiés expérimentalement par culture en pot en utilisant trois variétés de tabac (McNair, Kaba Koulac et Katerinis) très cultivées en Grèce. La migration verticale du ^{134}Cs dans le sol et la teneur en cet élément de la fumée produite par la combustion de feuilles radio-polluées ont également été étudiées. Une différence variétale significative a été observée dans le prélèvement racinaire de ^{134}Cs, tant dans le Facteur de Transfert (TF) calculé que dans la distribution du ^{134}Cs dans les différentes parties de la plante. Une différence variété-dépendante de la teneur en ^{134}Cs a aussi été mesurée pour la fixation foliaire (dépôt direct). On a observé que, par rapport aux autres parties (tiges, feuilles supérieures et fleurs), les feuilles du bas de la plante absorbaient ou interceptaient la plus grande quantité de ^{134}Cs. Durant les deux mois de culture du tabac, le ^{134}Cs résultant d'un dépôt humide se retrouvait dans les cinq centimètres supérieurs du sol mais aucune migration de radionucléide n'a été détectée dans les dépôts à sec. Enfin la teneur en ^{134}Cs de la fumée était inférieure à 1 % de la quantité totale dans les cendres des feuilles.

SUMMARY. — The root uptake of Caesium (^{134}Cs) and its subsequent distribution into the shoots, flowers and leaves, as well as the direct deposition of ^{134}Cs on these parts of the plant, were studied in pot experiments, using three tobacco plant varieties (McNair, Kaba Koulac and Katerinis) that are widely cultivated in Greece. The vertical migration of ^{134}Cs into the soil and the quantity of this element contained in the smoke of burning radio-polluted leaves were also studied. For the root uptake of ^{134}Cs, a significant varietal difference was observed, both in calculated Transfer Factor (TF) and in distribution of ^{134}Cs in the different plant parts. A variety dependent difference in ^{134}Cs content was also measured for foliar uptake (direct deposition). It was observed that the main quantity of ^{134}Cs was absorbed or intercepted by the lower leaves of the plants, compared to the quantity absorbed or intercepted by the other plant parts (stems, upper leaves and flowers). During the two-month period of tobacco cultivation, the wet-deposed ^{134}Cs was contained in the upper five centimeters of soil, while no radionuclide migration was detected in dry deposition. Finally, the amount of ^{134}Cs detected in the smoke was measured to be less than 1% of the total amount in the ashes of the leaves.

The cultivation of tobacco and the production of cigarettes are spread widely all over the world. Since tobacco production is of great importance to the Greek economy, ^{134}Cs contamination of tobacco plants was studied in pot experiments, under field conditions. The objective of the study was to improve knowledge on the consequences of radioactive pollution on tobacco crops.

Several studies have been conducted on: (i) mechanisms of Caesium uptake by plants (Cheshire & Shand, 1991; Rauret *et al.*, 1995; Hinton *et al.*, 1996; Gerzabec *et al.*, 1998;

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[1] Laboratory of Ecology and Environmental Sciences, Agricultural University of Athens, 75 Iera Odos, Botanikos, GR 11855 Athens, Greece. E-mail: mani@aua.gr

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White & Broadley, 2000; Zhu & Smolders, 2000; Absalom *et al.*, 2001; Goncharova *et al.*, 2002), (ii) foliar and root uptake of Caesium by crops used mainly in food production (Cline & Hungate, 1960; Smolders & Shaw, 1995; Carini & Lombi, 1997; Smolders *et al.*, 1997; Zhu *et al.*, 2000), and (iii) application of different countermeasures (Barber, 1964; Shaw & Bell, 1991; Bergeijk *et al.*, 1992; Shaw *et al.*, 1992; Maubert *et al.*, 1993; Prister *et al.*, 1993; Shaw, 1993; Arapis & Perepelyatnikova, 1995; Nisbet, 1995; Roca & Vallejo, 1995; Karaoglou *et al.*, 1996). However, despite them, there is a lack of experimental data concerning plants of specific interest, such as tobacco.

This study focuses on: a) the root uptake of ^{134}Cs and its subsequent distribution in the different parts of three varieties of tobacco plants, b) the proportion of ^{134}Cs deposited on the foliar surface, as opposed to the amount deposited on the soil, c) the distribution of radioactive deposition on different parts of the plant (shoot, lower leaves, upper leaves and flowers) and d) the vertical migration of ^{134}Cs into soils. Moreover, a simulation of "smoking" was performed and the content of ^{134}Cs in the smoke of the burning leaves, polluted by ^{134}Cs, was determined, in order to evaluate possible implications to human health (Duffy *et al.*, 1999).

MATERIAL AND METHODS

The following three varieties of Greek tobacco plants were used in this study (with their biotypes given in parenthesis): McNair (McN), Kaba Koulac Classic of Makedonia (KP 14/a) and Katerinis (S53). In addition to their ability to survive in unfavourable environmental conditions, the selection of tobacco varieties was based on the fact that these types are widely cultivated in Greece and around the world. The plants were cultivated in pots, using the same type of soil (Sandy Clay Loam) under the same climatic conditions described below in the laboratory, during the period of plant growth, and later in the field during radiopollution, under spring weather.

PLANT GROWTH

Tobacco seeds were planted in peat substrate that was watered regularly and remained under controlled conditions for a month (temperature: 28° C, humidity: 65%, light: 14 h and darkness: 10 h per day). Thereafter a first transplantation of the very young tobacco plants was made, using peat pallets (Jiffy-7) under the above mentioned conditions. At the end of this transplantation, we had at least fifteen plants per variety. The transplantation phase lasted approximately one month, on a daily basis, so that the proper development of the plants was not affected. Finally, the young plants of the three tobacco varieties were transplanted into 20 cm × 25 cm pots, with soil from an open-field cultivated tobacco area. A Sandy Clay Loam (SCL) soil type, with pH = 7.6, 31% $CaCO_3$ and 25 ppm of K, was used for cultivation of all plants and for all experiments. After a few days in the laboratory, the plants were moved to the field, under climatic conditions similar to the Mediterranean spring.

^{134}CS CONTAMINATION

Forty-five days after the placement of the plants to the field, radioactive pollution was simulated under: a) open-field conditions for soil contamination and root uptake of Caesium, and b) greenhouse conditions for Caesium interception by aerial parts of the plants. The radio-pollution was performed at an intermediate vegetation stage of the tobacco plants, with the first leaves well developed and a plant height of approx. 30 cm. Caesium-134 Chloride was applied in aqueous solution. From the fifteen plants of each variety, five were randomly selected for direct (or foliar) pollution, five for root uptake of ^{134}Cs and five plants remained as control.

Regarding root uptake, ^{134}Cs in the form of aqueous solution (activity: 3 kBq ml^{-1}) was deposited (50 ml per pot) with a syringe on the soil surfaces of the first group of fifteen tobacco plants (five plants per variety). Two months after contamination, the content of ^{134}Cs in the tobacco plants (which had reached the mature state) was measured and compared as a function of plant parts (shoot, lower leaves, upper leaves, flowers) and variety (McN, KP 14/a, S 53).

Regarding foliar radiopollution, a provisional greenhouse was constructed (2 m long, 1.5 m wide and 1.3 m tall) using a transparent plastic membrane. The pollutant (^{134}Cs in aqueous solution, 1.2 kBq ml^{-1}) was deposited on the plants using a micro-spraying device (aerographer). The total sprayed volume was 100 ml. Under the experimental conditions used, ^{134}Cs deposition was considered to be dry, due to the high indoor temperature of the greenhouse and the subsequent fast vaporization of the micro-spray. The tobacco plants of this second group (fifteen in total, five for each variety) were planted in parallel rows, randomly. The distance between plants was similar to that in an open field for commercial tobacco production. To ensure the best possible uniform contamination of all plants, the ^{134}Cs micro-sprayer (aerographer) was placed horizontally in four different positions, at the highest part of the greenhouse wall. The quantity of ^{134}Cs used was calculated in such a way to achieve a deposition similar to the one occurred in Greece after the Chernobyl accident (approx. 40 kBq m^{-2}, Izrael *et al.*, 1996). The duration of the spraying was approximately fifteen minutes in each position, while the total duration of pollutant deposition into the greenhouse and onto the plants was estimated not to be longer than 1 hour.

SIMULATION OF SMOKING

A small quantity (3.5 g) of leaves (equal to approximately five cigarettes) was burned in a special device, developed to simulate the process of smoking. The smoke was absorbed on a micro-porous filter (class P3 of European and DIN 3181 standards, mod. 230-235). At the end of the simulation the content of ^{134}Cs in both the ash of burned leaves and in the absorbed smoke was measured. For this investigation, only leaves from the tobacco variety that was the most radiopolluted, were used. The simulation of smoking was repeated three times.

MEASUREMENT OF ^{134}CS

Every sample (plant part and soil) taken for measurement of ^{134}Cs was homogenized, dried for about 24 hours at 70-80^{o} C and weighed in the laboratory. They were then placed in separate pre-weighed plastic tubes of 5 cm length and 7.5 cm diameter and analysed for ^{134}Cs content using a gamma-spectrometry-detector Ge(Li) (Canberra), that was shielded by a 10 cm thick lead, with efficiency 18% and resolution 1.9 keV (FWHM) at 1 332 keV. The detector was calibrated using IAEA reference-sources at the same geometry mentioned above.

STATISTICAL ANALYSIS

Data of the radionuclide absorption were analysed using 2-way ANOVA with factors: the variety and the part of the plant. Means for the variety and for the part of the plant were separated with Tukey HSD test (a = 0.05).

RESULTS AND DISCUSSION

The most significant results of this study are being presented in groups, in the following four sections: a) root uptake of ^{134}Cs by the three varieties of tobacco plants, b) foliar uptake or interception of ^{134}Cs by the plants, c) vertical migration of ^{134}Cs in the soil of pots, and d) content of ^{134}Cs in the smoke of burned radio-polluted tobacco leaves (simulation of smoking).

ROOT UPTAKE

Figure 1 shows the concentration of ^{134}Cs in the different plant parts (lower leaves, upper leaves, flowers and stems), following root uptake by the three varieties of tobacco plants (McNair, Kaba Koulac and Katerinis).

Figure 1. — Mean ^{134}Cs content in different plant parts of three tobacco plant varieties following root uptake.

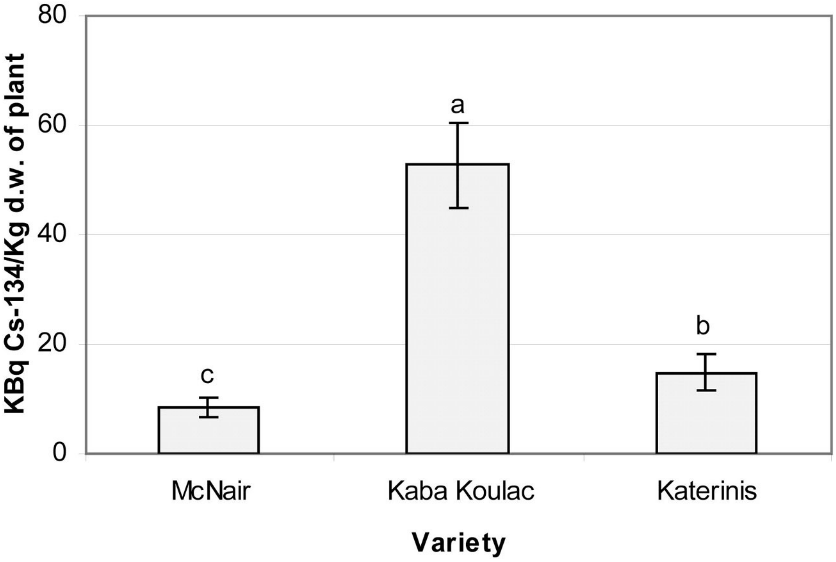

Figure 2. — Mean ^{134}Cs content in the three tobacco plant varieties (+/– standard error). Bars followed by different letter differ significantly (Tukey HSD test, a = 0.05).

Statistical analysis showed that a significant difference exists between the root uptake of ^{134}Cs by the different tobacco plant varieties (Tab. I). The mean radionuclide content (+/– standard error) is presented in figure 2. For the three varieties (McNair, Kaba Koulac and Katerinis), the calculated radionuclide transfer factor TF was found to be 0.055, 0.346 and 0.097 respectively with

$$\mathrm{TF} = \frac{\text{activity concentration of nuclide per kg of dry plant - mass}}{\text{activity concentration of nuclide per kg of dry soil}}$$

TABLE I

Effects of variety and part of the plant on the ^{134}Cs uptake (2-way ANOVA)

Source	Nparm	DF	F Ratio	Prob > F
Variety	2	2	325.089 7	< .000 1
part of plant	3	3	187.636 2	< .000 1
Variety * part of plant	6	6	36.506 6	< .000 1

In other words, the ^{134}Cs TF for the Kaba Koulac variety seems to be 6 times higher than that of the McNair and 3.5 times higher than that of the Katerinis variety.

Statistical analysis also showed that a significant difference exists between the distribution of ^{134}Cs in the lower leaves, upper leaves, flowers and stems of the tobacco plants (Tab. I). Their mean radionuclide concentration (+/– standard error) is presented in figure 3. Taking into account all plant varieties, the bulk of the activity (more than 50%) was measured in the lower leaves and approximately 20% of the activity was detected in the stems.

Figure 3. — Mean ^{134}Cs content in the different parts of the plant (+/– standard error). Bars followed by different letter differ significantly (Tukey HSD test, a = 0.05).

FOLIAR UPTAKE

The first observation made for all plants of the three tobacco varieties is that the proportion of ^{134}Cs intercepted by their leaves was approx. 65% (K 5%), while only 35% (K 5%) of the ^{134}Cs was directly deposited on the soil.

From the results in table II it was concluded that the direct contamination of the tobacco plants depends on their morphological differences or leaf inclination. Although the McNair variety had the widest and most horizontally orientated leaves, it had the smallest measured radio-pollution concentration. On the other hand, the Katerinis variety, with the most pointed leaves, had the largest pollutant concentration (12 times as high as the McNair variety), probably due the roughness of leaves. Finally, the Kaba Koulac variety was measured to have an average radiopollution compared to the other two varieties.

TABLE II

^{134}Cs contamination of leaves as a function of the tobacco-plant variety, inclination of leaves and mean surface of leaves

Variety of tobacco plant	Leaf inclination(degree)	Leaf surface (mean, cm^2)	Radioactivity (kBq m^{-2})
McNair (McN)	5^{o} (K2^{o})	85.96 (K9.62)	14.82 (K8.12)
Kaba Koulac (KP 14/a)	15^{o} (K5^{o})	50.87 (K4.25)	108.11 (K19.67)
Katerinis (S53)	15^{o} (K5^{o})	66.66 (K6.32)	181.68 (K24.15)

Figure 4 shows the direct contamination of ^{134}Cs for the lower leaves, upper leaves, flowers and shoots of the three tobacco plant varieties. We observed that a significant difference exists in the amount of ^{134}Cs intercepted by each part of the three plant varieties. This difference seems to be due to the non-homogeneous distribution of leaves in the green-

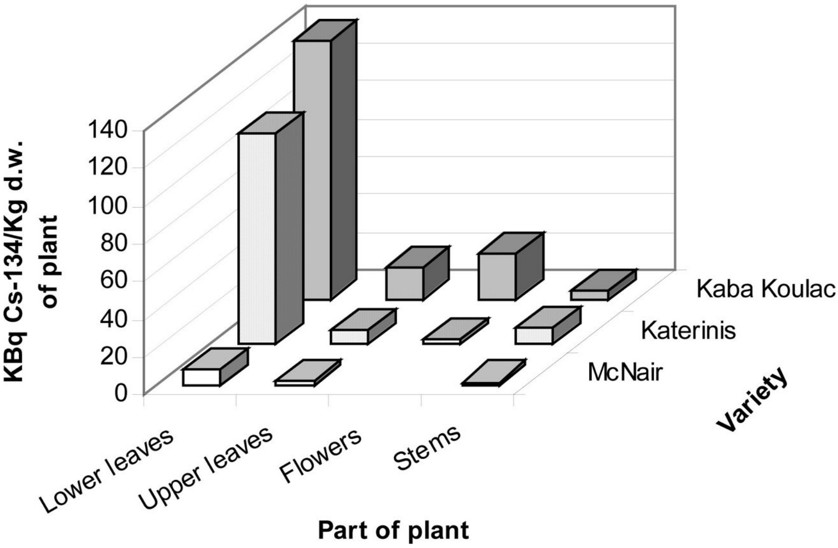

Figure 4. — Mean ^{134}Cs content in different plant parts of three tobacco plant varieties following direct radiopollution.

house space. Moreover, we observed that the main quantity of ^{134}Cs was intercepted by the lower leaves of the plants at an approximate ratio of 4:1 compared to the quantity intercepted by the upper leaves.

MIGRATION OF ^{134}CS IN THE SOIL

A further important finding of this study is that the main quantity of ^{134}Cs used for contamination of the soil-surface of pots (wet deposition) was located in the upper 5 cm layer of soil, while no significant quantity of ^{134}Cs was detected in the deeper soil-layers during the two-month period of tobacco cultivation. The average activity of the upper layer of soil was approx. 150 kBq kg^{-1} (15 kBq kg^{-1}).

Concerning the greenhouse experiments, it was observed that the soil contamination resulting from the dry deposition of ^{134}Cs was contained in the first upper centimeter of soil, while no remarkable quantity of ^{134}Cs was detected moving deeper in the pot soil.

CONTENT OF ^{134}CS IN THE SMOKE

Table III shows the content of ^{134}Cs in both the ashes of the burned radio-polluted leaves and the smoke absorption-filter. The results showed only traces of ^{134}Cs into the microporous absorption-filter, from the burning of the most radio-polluted leaves of the Kaba Koulac variety. In fact, the amount of radionuclide detected in the filter was less than 1% of the

TABLE III

Content of ^{134}Cs in the ash of burned radio-polluted leaves and in the absorption-filter (following a smoking-simulation)

Tobacco plant variety	Content of(mean) ^{134}Cs		
	Leaves (Bq/sample)	Filter (Bq/sample)	Ash (Bq/sample)
Kaba Koulac (KP14/a)	977.91 (K99.02)	6.66 (K4.31)	971.27 (K85.67)

amount of ^{134}Cs measured in the ashes of the leaves. This large percentage of pollutant measured in the ash is probably due to the fact that its particle size, during the "smoking" phase remained big enough (approx. 1 μ) to avoid transfer into the smoke-flux.

CONCLUSION

In this study, a significant plant variety dependent difference was observed in calculated Transfer Factor (TF) and the distribution of ^{134}Cs in different plant parts, regarding the root uptake of ^{134}Cs. A similar varietal-difference in the ^{134}Cs content of leaves was observed for foliar uptake (direct deposition), due to morphological peculiarities of the three tested tobacco varieties. Moreover, the lower leaves of tobacco plants absorbed the greatest quantity of ^{134}Cs, which could be further related to the production process of cigarettes and to their quality. Finally, the amount of ^{134}Cs detected in the smoke, following the burning of leaves, was measured to be less than 1% of the total amount in the ashes of tobacco. This means that the quantity of radionuclides inhaled by the smokers is relatively low; however, a relevant risk for human health is to be taken into account.

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