(7) contained, as measured by gasliquid chromatography and ultraviolet, infrared (6) and mass spectrometry, a mixture of 30 percent trans-(C4)-trans-(C6)-alloocimene and 70 percent trans-(C₄)-cis-(C₆)-alloocimene (8).

The identification of dipentene was made on the basis of identical retention times (3.7 minutes) and mass spectra for the smoke isolates and for an authentic sample (9). The major ion fragment had a ratio of mass to charge of 68 which is typical of dipentene and distinguishes it from several of the isomeric terpenes (10).

JAMES D. MOLD

THOMAS B. WALKER

JAMES B. WILLIAMS

Research Department, Liggett and Myers Tobacco Company, Durham, North Carolina

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Radium-226, Radium-228, Lead-210, and Fluorine in

Persons With Osteogenic Sarcoma

Abstract. Concentrations of the naturally occurring alpha-emitting radioelements, radium-226, radium-228, and lead-210, and of stable lead and fluorine were determined in bone specimens from 32 individuals having a verified osteogenic sarcoma. Comparison of these results with those for the average person showed no significant differences in either the absorbed dose (rad) from the accumulated radioisotopes or in the concentrations of the elements studied.

As part of a program to determine the toxicity to humans of internally deposited radioelements, we have studied the uptake and retention of Ra²²⁶, Ra²²⁸, Th²²⁸, and Pb²¹⁰ arising from the environment (1, 2, 3). Since large amounts of Ra²²⁶ or other bone-seeking radioelements have been shown to produce a high incidence of osteogenic sarcoma (4), it is conceivable that the normal natural environmental radiation may have a direct effect on the spontaneous incidence of this disease.

Natural levels of radium, however, are more than a factor of 10³ lower than the levels known to be toxic and the range of the variations in dose is small. In addition, exposure is lifelong, and the dose is distributed more uniformly throughout the skeleton than that at the known toxic levels. In spite of these differences in deposition and dose, it has been presumed by some that the variation in natural radiation dose will result in a corresponding variation in the incidence of osteogenic sarcoma.

Naturally occurring radioisotopes which contribute significant radiation doses to bone include K40, Ra226, Ra228, and Pb^{210} . The metabolism of K^{40} is 26 JUNE 1964

such that there is little individual variation within a given age group (5). We have, therefore, compared the Ra²²⁶, Ra²²⁸, and Pb²¹⁰ content of the bone from normal individuals with that in proven cases of osteogenic sarcoma. In addition, we have determined the concentrations of two stable bone-seeking elements, lead and fluorine.

The concentrations of these elements depend on the environmental concentrations. Radium-226 is acquired through both food and water, but since most of the world's population consumes water with no significant concentration of radium, food is the primary source. In these individuals the Ra²²⁶ concentration is low and the values span a relatively small range (6). In some local areas, however, the drinking water comes from wells which have a high Ra²²⁶ concentration, and individuals consuming this water may have a body burden ten or more times greater than those whose only source of radium is food (1). The Ra²²⁸ content of the body is similarly controlled, but because of the short half-life of the Ra²²⁸ (5.8 years) (7) the concentration in the body varies with age. Thorium-228, a decay product of Ra²²⁸, is not appreciably absorbed from either food or water and hence its presence in bone is due to the decay of its parent, Ra²²⁸. Fluoride, like radium, enters the body primarily through food and drinking water, whereas Pb²¹⁰ and lead are acquired from both food and air (3, 8).

For this study undiseased bone was obtained from the area adjacent to the tumor. When received, the samples were cleaned of soft tissue and bone marrow, dried at 110°C, and stored in sealed polyethylene bags at a temperature of -27° C until analyzed. The Ra²²⁶, Ra²²⁸, Pb²¹⁰, stable lead and fluorine were determined as already described (9).

The samples chosen to represent bone from subjects with osteogenic sarcoma were specimens taken from bone removed in the treatment of this disease at the Mayo Clinic over the period of about a year. These subjects, with one exception, had been exposed only to natural environmental radium. The one exception, who had received the equivalent of an intravenous injection of about 400 μ g of radium, has been excluded from this study and is discussed elsewhere (10).

An osteogenic sarcoma is defined as a malignant tumor, the malignant cells of which produce osteoid tissue even if in only small foci. These sarcomas may be classified as osteoblastic, chondroblastic, or fibroblastic depending on whether there is a predominance of osteoid, chondroid, or fibromatoid differentiation. Osteogenic sarcomas were graded from 1 to 4, the larger values corresponding to the greater degree of cellular dedifferentiation. The distribution of the grades of malignancy (1 to 4) in this series of 32 cases was 0, 22, 44, and 34 percent, respectively, which is very similar to the 1.6, 17.2, 54.4, and 26.7 percent, respectively, found in a series of 430 cases (11). There is a corresponding similarity in histologic types: osteoblastic, chondroblastic, and fibroblastic types were, respectively, 63, 12, and 25 percent in this series and 50, 27, and 23 percent in the larger series. The ratio of 19 males to 13 females, and the anatomical distribution of skeletal involvement are also typical for any group of cases of osteogenic sarcoma. The group of subjects selected for this study, therefore, appears to be a representative sample.

The measurements of the Ra²²⁶, Ra²²⁸, Pb²¹⁰, total Pb and F in bone from these individuals having verified osteogenic sarcomas are summarized in Table 1.

Table 1.	Concentrations	of Ra ²²⁶ ,	Ra 228,	Pb210,	Pb a	and	F in	bone	ash	of	individuals	with	an	osteo-
genic sa	rcoma.													

Radi	Other elements (μ g/g ash)			
Ra ²²⁶	Ra ²²⁸	Pb ²¹⁰	Pb	F
$0.008 \pm .002$	$0.003 \pm .002$		4.0	199
$.011 \pm .002$	$.004 \pm .002$	$0.019 \pm .009$	30.5	191
$.011 \pm .003$	$.005 \pm .003$	$.076\pm.006$	9.5	244
$.012 \pm .002$	$.006 \pm .004$		16.4	247
$.012 \pm .003$	$.007\pm.003$	$.075\pm.003$	4.2	140
$.013 \pm .001$	$.004 \pm .001$	$.061 \pm .007$	17.7	190
$.013 \pm .002$	$.006\pm.002$	$.077\pm.007$	9.9	192
$.013 \pm .001$	$.002 \pm .001$	$.054\pm.006$	22.0	620
$.016 \pm .002$	$.006\pm.002$	$.043 \pm .003$	16.9	123
$.016 \pm .003$	$.005\pm.002$		12.6	83
$.017 \pm .002$	$.003 \pm .001$	$.054\pm.006$	24.0	110
$.018 \pm .014^{*}$	$.006\pm.003$		3.3	
$.020 \pm .001$	$.004\pm.002$	$.097 \pm .008$	13.8	390
$.020 \pm .003$	$.006\pm.002$	$.042 \pm .005$	7.0	83
$.002 \pm .003$	$.005\pm.002$		10.8	930
$.023 \pm .003$	$.005\pm.002$	$.105 \pm .006$	15.0	456
$.024 \pm .003$	$.007\pm.002$		6.5	1110
$.027 \pm .002*$	$.007\pm.001$	$.054\pm.008$	7.6	465
$.031 \pm .003$	$.007\pm.003$	$.099 \pm .008$	16.3	1190
$.033 \pm .002$	$.010\pm.002$	$.064 \pm .008$	14.7	81
$.033 \pm .006*$	$.009 \pm .009$		18.1	210
$.033 \pm .006$	$.003\pm.002$		10.1	533
$.035 \pm .006$	$.008 \pm .001$	$.092\pm.011$	20.6	251
$.037\pm.003$	$.013\pm.006$		7.5	134
$.038 \pm .002$	$.008\pm.002$	$.088\pm.011$	24.7	135
$.038 \pm .003$	$.003\pm.002$	$.072\pm.007$	24.5	944
$.042 \pm .002*$	$.004 \pm .002$	$.110 \pm .009$	25.9	998
$.044 \pm .004*$	$.016 \pm .003$		25.8	608
$.048\pm.003$	$.018 \pm .002$		26.2	1080
$.050\pm.002$	$.010 \pm .001$	$.182\pm.010$	38.6	1150
$.058\pm.003$	$.013 \pm .004$	$.066 \pm .007$	7.3	386
$.075\pm.003$	$.019\pm.002$	$.063 \pm .008$	22.6	223
		Mean \pm S.D.		
0.028 ± 0.003	0.007 ± 0.001	0.080 ± 0.007	16.1 ± 1.5	411 ± 63

* Noncortical bone.

The means and the standard deviation of the means, 0.028 ± 0.003 pc of Ra²²⁶ and 0.007 ± 0.001 pc of Ra²²⁸ per gram of ash, are significantly higher (P <.001) than the 0.0124 \pm 0.005 pc of Ra²²⁶ and 0.003 \pm 0.002 pc of Ra²²⁸ per gram of ash found in individuals consuming water of low radium content (1, 2). In contrast, concentrations of the mean Pb²¹⁰, stable lead, and fluorine of 0.080 pc, 16 µg, and 411 µg per gram of ash, respectively, are not significantly different from those in individuals not having osteogenic sarcomas (3, 12).

For individuals with a lifelong (or at least 30 years before measurement) exposure at continuous and nearly constant rates of intake, it was found that the concentration of Ra^{226} in human bone from both the food and drinking water could be expressed as follows (1):

$$R = 0.0124 + 0.02 C_w \tag{1}$$

where R is the expected radioactivity (in picocuries per gram of ash) and C_{w} is the concentration of Ra^{226} (in pc/liter) in the individual's drinking water. Since most surface water supplies contain 0.1 pc of Ra^{226} per liter or less (13), individuals consuming these waters obtain most of their body radium from food. Variations in the radium content of food do not appear to be more than about 20 percent (14) and have been neglected in Eq. 1. In the Midwest, many individuals consume water with 4 to 37 pc of Ra²²⁶ per liter (15) and thus obtain most of their body burden from the drinking water.

The somewhat high value of the mean Ra^{226} concentration found in this survey may be shown to correlate very well with the probable Ra^{226} intake of

Table 2. Comparison of absorbed dose to the skeleton from internal emitters in average individuals and in those with an osteogenic sarcoma.

Ensitter	Ave indiv	rage vidual	Osteogenic sarcoma patient			
Eliniter -	pc/g ash	mrad/ yr	pc/g ash	mrad/ yr		
Ra 226	0.012	0.9	0.027	2.1		
Ra ²²⁸	0.003	0.7	0.007	1.6		
Pb210*	0.105	4.1	0.080	3.1		
K40		10.		10.		
Totals	5	15.7		16.8		

* Values in cortical bone.

these subjects and, thus, is not due to metabolic effects such as a reduced dietary discrimination factor or a selective deposition in the skeleton. In 19 of the cases of osteogenic sarcoma, the residential history was sufficiently stable and water samples were available so that comparison could be made with the uptake previously observed for other individuals. The expected concentration of Ra²²⁶ in bone ash, R, was computed from Eq. 1 and compared with that observed. Multiple regression analysis and application of the appropriate form of the Student's t test indicates that the radium concentration in the bone from the individuals with an osteogenic sarcoma is not significantly different (P < .01) from that expected.

Evaluation of the total radiation dose requires information about type of housing and medical x-ray exposure history which is not now available. If, however, we compare only the internal radiation dose in these patients with that of the average person, an indication of the significance of internal emitters may be inferred. The absorbed dose for Ra^{226} , Ra^{228} , and Pb^{210} was calculated on the assumption that ash equaled 37 percent of the wet skeleton. The dose rates for 1.0 pc of parent nuclide per gram of ash are as follows:

$Ra^{226} + 30\%$	daughters	
	thru Po ²¹⁴	76 mrad/yr
$Ra^{228} + 100\%$	daughters	233 mrad/yr
$Pb^{_{210}} + 100\%$	daughters	39 mrad/yr

As shown in Table 2, the Ra²²⁶ and Ra²²⁸ concentrations are higher in osteogenic sarcoma bone than in bone from the average person. However, the lower Pb²¹⁰ concentrations compensate to reduce the difference in dose to insignificance particularly in view of the larger K^{40} contribution (16). The increase in the Ra²²⁶ dose rate is 1.2 mrad/yr which is only 7 percent of the total dose from the internal emitters. If one also includes the average radiation dose rate to the skeleton from cosmic and terrestrial sources of about 80 mrad/yr (17), then the difference of 2.1 mrad/yr in radium ($Ra^{226} + Ra^{228}$) dose rate amounts to only about 2 percent of the total. The relative biological effectiveness (RBE) of the various radiations, however, has not been considered.

Unfortunately, good values for the relative biological effectiveness of the radiations in the continuous, low-level human exposure are not available, and the dose equivalent (18) (RBE dose) cannot be estimated with any degree of certainty. Since the RBE for alpha par-

ticle irradiation appears to be greater than one, the effect of the irradiation from Ra²²⁶, Ra²²⁸, and Pb²¹⁰ may be greater than that from the other internal emitters. However, the same RBE factor must be used for the normal individual as for the one having an osteogenic sarcoma so that the difference in dose is still relatively small.

In addition, no great significance can be attached to the slightly higher dose rates calculated for the osteogenic sarcoma cases because the hospital from which the osteogenic sarcoma samples were obtained is located within the geographical area having high concentrations of radium in municipal waters. Consequently, one expects a higher concentration of Ra²²⁶ and Ra²²⁸ in the skeletons of these individuals than in those from other areas.

In conclusion, this study has demonstrated that for an individual with an osteogenic sarcoma, the internal dose rate, radium metabolism and possibly the metabolism of Pb²¹⁰, stable lead, and fluorine, do not differ significantly from those of the average human being. Thus, in future studies, the metabolism of these substances by individuals with osteogenic sarcomas may be assumed to be identical to those of the unaffected population, so that measurements the former group are unnecessary.

While it is conceivable that the variation in environmental radiation may have a direct effect on the spontaneous incidence of osteogenic sarcoma in man, this study is not suitable for this purpose. Estimation of the osteogenic sarcoma incidence requires knowledge of the population at risk for each of the dose levels. Although these data are available for a limited geographical area, they are not available for the region in which these individuals lived. This estimate must await completion of an epidemiological study such as is in progress in the Midwest.

> H. F. LUCAS, JR. R. B. HOLTZMAN

Radiological Physics Division, Argonne National Laboratory, Argonne, Illinois D. C. DAHLIN

Section of Surgical Pathology, Mayo Clinic, Rochester, Minnesota

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26 JUNE 1964

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Sterols in Recent Aquatic Sediments

Abstract. The presence of sterols in recent fresh-water sediments was confirmed and, for the first time, sterols were found to be widely distributed throughout recent marine sediments. Quantities ranged from 60 to 300 parts of cholesterol carbon per million parts of organic carbon in the sediments. No correlation of sterol content with environment or depth of burial in the sediment was apparent.

The suggestion that sterols might be a source material for some of the compounds in petroleum was first advanced by Walden (1) and numerous references have been made to this possibility during the intervening years; the literature on the subject has been reviewed by Bergmann (2). Unequivocal demonstration that such is the case should include evidence not only that sterol degradation products are present in crude oils, but also that sterols themselves are present in those recent sediments which are thought to represent future source beds of petroleum. The first evidence for the presence of sterol degradation products in crude oil was the recent isolation of Diels' hydrocarbon from Ponca City crude oil by Mair and Martinéz-Picó (3). However, convincing evidence for the occurrence of sterols in recent sediments is lacking since statements in the literature on this point are contradictory.

Although sterols have generally been found in recent terrestrial and freshwater sediments by Turfitt (4) and others (2), Trask and Wu (5) were able to detect them in only one recent marine sediment. Fox and Oppenheimer (6) specifically stated that sterols could not be found in recent marine sediments; this is especially surprising in light of the work of Turfitt (7), who showed that lack of aeration and high water content were factors which inhibit the growth of organisms responsible for the decomposition of sterols in soils. Since the evidence at hand suggests no

Table 1. Concentration of sterols in recent sediments expressed as parts of cholesterol carbon per million parts of organic carbon of each sediment.

Loca-	Depth within sedimer	nt (cm)
tion	0- 15- 105- 110- 115-	- 120-	125-
	15 30 120 125 130	135	140
	Tamarack Bog		
3	128		
	Okefenokee Swamp: savan	nah	
3	60 86		
	Okefenokee Swamp: cypr	ess	
4	118* 60†		
	Bellefontaine Marsh		
3	261 237		
	Mississippi Sound		
1	174		102
4	160		
	Gulf of Mexico		
1	107 91 157	144	
2	147		
3A	304	209	
4	250		
	Santa Barbara Basin		
1	172 104		
•	San Nicolas Basin		
2	99		

^{*} Duplicate determinations gave values of 113 and 122. † Duplicate determinations gave values of 53 and 66.