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Ra-226 IN CATTAILS (*Typha latifolia*) AND BONE OF MUSKRAT (*Ondatra zibethica*) FROM A WATERSHED WITH U TAILINGS NEAR ELLIOT LAKE, CANADA, WITH CALCULATION OF CONCENTRATION RATIO

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ABSTRACT

Radionuclide levels were measured in bone and diet items of adult muskrat from the Serpent River drainage basin which contains U tailings at Elliot Lake, and from control sites in Ontario. Muskrats from waters with mean total ^{226}Ra levels in the period 1984-1987 of 111.0 mBq.L^{-1} or more, near U tailings at Elliot Lake, Ontario, Canada, had a mean bone ^{226}Ra level of 468.0 mBq.g^{-1} (dry weight, n = 30), those from nearby waters with less than 111.0 mBq.L^{-1} had a mean bone level (61.5 mBq.g^{-1}, n = 11 similar to that of animals taken in uncontaminated local control areas 20 km from the tailings (92.8 mBq.g^{-1}, n = 16); animals from a control area at Sudbury, 130 km distant, had the lowest mean burden (11.5 mBq.g^{-1}, n = 24). Levels were unrelated to age or sex of the animals.

Ra-226 levels in the main plant eaten by muskrat (cattail, *Typha latifolia*) varied by plant part and place of collection. Roots sampled in the contaminated area had the highest mean ^{226}Ra level (1,135.0 mBq.g^{-1}), stems and leaves had 284.2 and 275.9 mBq.g^{-1} respectively (n = 6 in all cases). Radium-226 levels in cattails generally declined with increasing distance of sample location from Elliot Lake. Cattails from the local control locations had a mean level of ^{226}Ra of 20.0 mBq.g^{-1}; distant control samples had 15.1 mBq.g^{-1} (weighted average).

Concentration ratios between dietary items (plant parts) and bone of muskrat taken at Elliot Lake-high sites were calculated (fresh weight basis) to range from 1.1 to 8.5. These were similar to values calculated for other species and in general agreement with values calculated for humans. Transfer parameters were relatively low from diet to muskrat bone and it seems unlikely that adverse effects accrue to animals living on the Elliot Lake U tailings.

Key words: Radiobiology; Muskrats; Cattails; Environment.

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The muskrat is judged to be a useful indicator (biomonitor) of environmental radionuclide contamination as correlation between bone and ambient (water) levels of ^{226}Ra is high. Estimated yearly intakes of radionuclides by people eating muskrat were calculated to be below current allowable levels set by Canadian regulatory authorities.

INTRODUCTION

As part of a study of radionuclide movement through an ecosystem containing U mining and milling operations, ^{226}Ra was measured in bone tissue of muskrat (Ondatra zibethica), and its dietary plants from uranium mining and waste management area at Elliot Lake, Ontario, Canada, and in bone of animals from uncontaminated waters of a distant control area. The work was done to provide information on ^{226}Ra levels and concentration factors of use in modelling radionuclide movement, to assess muskrat as a bioindicator of radionuclide contamination, and to calculate radionuclide intake by humans consuming the animals.

Mining and milling of U ore at Elliot Lake, Ontario (Figure 1), have produced extensive waste deposits over the last thirty years. These deposits, both abandoned and active, drain into the Serpent River which enters Lake Huron at Serpent Harbour. Egress of radionuclides from the deposits needs to be monitored in order that tailings control and rehabilitation procedures may be assessed. Up to 85% of the radioactive material present in the U ore is present in the final waste which is considered a potential source of environmental radioactive contamination if not managed and contained properly. Information on the release of radionuclides from the tailings, and their subsequent movements through the ecosystem, needed to assess risk to man, is scanty.

Of particular concern is ^{226}Ra as only 1% of the ^{226}Ra present in the ore is removed during processing and the resulting slurry waste contains up to 100 times the permissible level of radionuclide (ICRP 1978). Furthermore, ^{226}Ra has high specific activity and long half life, and its chemical analogy to the essential element Ca makes it seek bone as a deposition site in the body. Bound in calcium hydroxyapatite bone crystals, ^{226}Ra may cause tissue damage, possibly resulting in osteosarcoma, as it emits alpha and beta radiation (van Dilla et al. 1958; Mays et al. 1975; Schlenker et al. 1982; Raabe et al. 1983).

Ra-226 enters the watershed via surface and ground water flow regimes as a result of leaching from active and inactive tailings and through accidental tailing overflows or decants (required to discharge excess water from tailings ponds) and discharge of mill water (McKee et al. 1985). Contamination from natural sources is low compared to that resulting from human activity (Mahon 1982; Hesslein and Slavicek 1984; Baweja et al. 1987).

Effective tailings management and rehabilitation requires understanding and knowledge of radionuclide movement through the ecosystem of which the tailing deposits form a part. Modelling is commonly used to assist this understanding and to predict behaviour of the contaminated ecosystem. In turn, modelling requires knowledge of transfer parameters between biologically linked components of the system (e.g., substrate and plant tissue; plant and herbivore etc.). The concentration ratio (CR) is a transfer parameter defined as the ratio between steady-state concentrations in connected compartments of a model (ICRP 1978) such as an animal's diet (vegetation) and the animal's tissue (e.g., bone); it is useful in modelling equilibrium conditions. The CR, while not an indicator of the effects of radionuclide concentration on an organism, is a tool by which differences in accumulation by organisms may be assessed.

Radionuclide movement from substrate to vegetation to animal (tissue) has been measured among several components in the contaminated ecosystem. Food and tissues of voles (*Microtus pennsylvanicus*), deer (*Odocoileus virginianus*), and moose (*Alces alces*), all terrestrial mammals from the vicinity or drainage of the U tailings at Elliot Lake, Ontario, have been studied under wild and laboratory conditions to provide data useful in this regard (Burns et al. 1987; Cloutier et al. 1983, 1985a, b; and 1986; MacLaren 1978, 1987). Information on mammals associated with the water bodies of the area is restricted; MacLaren (1978) gives levels of ^{226}Ra in muscle tissue of four beaver (Castor canadensis) from the vicinity of Quirke Lake; Clulow et al. (1988X) report on tissue and diet levels of ^{226}Ra in 194 beaver from Elliot Lake and control areas. Beaver from Northern Saskatchewan have been reported to carry measurable burdens of ^{226}Ra (Beak 1986). Wren et al. (1987) report measurable levels in bone of otter (Lutra canadensis) in the Elliot Lake region. ^{226}Ra levels in faeces of hares, Lepus americanus, from the area have been reported (Clulow et al. 1986) as have levels in cutworms (Agrotis ipsilon) eaten by gulls (Larus argentatus) visiting the tailings (Clulow et al. 1988). Arthur and Markham (1982) reported that coyote (Canis latrans) taken from contaminated areas have measurable levels of radionuclides in their tissues.

Mammals have been examined as candidate biomonitors of radionuclide contamination (Cloutier et al. 1984). Biomonitors should be abundant, easily obtained, limited in their movements, widespread in their distribution, and accurate reflectors of differences in environmental radionuclide concentrations.

Muskrat, Ondatra zibethica along with beaver, Castor canadensis, are the principle aquatic mammals of the Elliot Lake area. Studies indicate that the muskrat may be a good indicator of metal concentrations in aquatic environments (Everett and Anthony 1977; Erickson and Lindzey 1983). Muskrat were investigated because they are abundant in and around tailings areas and adjacent watershed systems, they are easily acquired, as they are trapped for their fur; and are relatively long-lived animals. Proulx and Gilbert (1983) reported the maximum lifespan of muskrat in an Ontario marsh to be 5 y. Muskrat have widespread distribution (Errington 1961; Perry 1982).

Knowledge of the natural history of the animals is essential both for the initial design of an investigation of this type and for subsequent interpretation of results. Food habits and general mobility of the animals are of particular importance.

Muskrat diet varies with habitat, season, and food availability (Butler 1940). Perry (1982), in his review of the muskrat, describes the animal as chiefly herbivorous, eating shoots, roots, bulbs, tubers (rhizomes), stems and leaves of various hydrophytes. Cattail (Typha latifolia) is the most important plant species utilized in Canadian populations which use it both for food and lodge construction. Other food items include bullrush (Scirpus validus), pondweed (Potamogeton sp.), water horsetail (Equisetum fluviatile), and reed grass (Phragmites maximum). Muskrat frequently dig into pond or lake bottoms to acquire such food items. Young muskrat, only a few weeks old, tend to feed more on bank vegetation than adults. In winter, food items are limited to underground plant parts or whatever can be reached through swimming under the ice; large food caches are uncommon. Errington (1941) observed that in times of severe food shortage parts of the lodge or bedding may be consumed. In addition, animal matter (crayfish, molluscs, frogs, fish and turtles) may constitute a portion of the diet. These items are eaten as dietary supplements during times of food shortages or in periods of prey abundance (Errington et al. 1963). Lantz (1910; cited in Perry 1982) in his study on muskrat, reported 63 g or more of moist food in some muskrat stomachs. Stearns and Goodwin (1941) determined dry stomach contents in Delaware muskrat to range from 0.1 to 17.7 g with an average of 2.4 g. No differences were found between stomach contents of males or females in percentages of moisture, ether extract, crude protein, crude fiber, ash and nitrogen - free extract. Analyzing the roots and tops of ten plants considered representative of vegetation where muskrat were trapped, the workers determined that crude fiber measurements on the other hand were higher in plant tops than roots. Butler (1940) found a consumption of 20.5 ± 3.2 cattail plants per animal each day whereas McEwen et al. (1974) measured mean dry matter intake to be 80 $g.kg^{-1}.d^{-1}$.

Muskrat are mostly sedentary but sometimes undertake extensive travels. Dispersal is most common after ice breakup when movements associated with exploratory drifting (of juveniles) and establishment of breeding territories (by adults) occur. Social and environmental pressures, such as high density or depletion of food resources, may also result in muskrat dispersal (Errington 1951; Perry 1982). Translocation up to 34 km has been observed (Errington 1951). Movement was noted in 30 to 40% of marked populations and male dispersion is greater than that of females (Aldous 1946; Erickson 1963). In comparison, regular muskrat movement tends to be within a 15 m radius of a dwelling for more than 50% of the time with few movements being greater than 150 m. Most foraging tends to be within 10 to 15 m of their lodge (Wragg 1955; MacArthur 1978).

The objectives of the study reported here were to measure levels of ^{226}Ra in the bone and food items of muskrat in the region of Elliot Lake (uranium mining district) and an uncontaminated control area near Sudbury, Ontario, and to calculate therefrom concentration ratios useful in modelling ^{226}Ra movements in the ecosystem, to assess the animal as a biomonitor of radionuclide contamination, and to calculate intakes by human consumers.

METHODS AND MATERIALS

SAMPLING LOCATIONS

Investigations took place in the Sudbury-North Bay and Timagami sections of the Great Lakes - St. Lawrence Forest Region of Canada (Rowe 1959) described in detail elsewhere (Clulow et al. 1988X). The area has rugged outcrops of Canadian Shield bedrock among wet flats and lowlands variably covered with mixed hardwoods and conifers.

Specimens were collected from the study area: 'Elliot Lake', containing the Town of Elliot Lake, U tailings deposits, and several lakes and waterways. Substantial variation in ^{226}Ra concentration in these water bodies has been reported (see below). Control specimens were taken from 'Control-local', the area of Tweedle, Sagard and Poulin Townships, located about 40 km NW of Elliot Lake, upwind and in a different watershed, chosen because of accessibility, location, and lack of U industry operations. Occurrences high in Th-series radionuclides are known in the area (W. Meyer, Resident Geologist, Ministry of Northern Development and Mines, Government of Ontario, Sudbury, personal communication). Another local control area, chosen for ease of access about 15 km south of Elliot Lake, was the source of a few muskrat in 1985 only. Additional samples were obtained from 'Control-distant', an area more than 160 km E of Elliot Lake, centred on Sudbury but extending north to Lakes Wanapitei, Boot, Rathbun, Portage, and Matagamasi, chosen for its remoteness from Elliot Lake and its ease of access. Several U occurrences have been reported in the area, but none have been developed (W. Meyer, personal communication).

Water quality records of the Ministry of the Environment of the Province of Ontario (MOE) (MOE 1981, 1982, 1987) indicate substantial variation in ^{226}Ra concentration from place to place and over time at MOE sample stations in the vicinity of Elliot Lake. Water sampling stations within the study area at Elliot Lake were assigned to one of two classes based on the average radionuclide levels of water sampled at each station. The average total ^{226}Ra concentration at each station was calculated by summing all total ^{226}Ra water values for the years 1984 to 1987 and dividing by the number of samples. Prior to 1984, only filtered ^{226}Ra values were available;

these values were not used in this study. Site classes within the study area were defined as follows:

Elliot Lake-high, with ^{226}Ra levels $\geq$ 111 mBq.L^{-1} (included are all tailings deposits and portions of the Serpent River below an effluent discharge site at the Quirke Mine waste management area).

Elliot Lake-low, with ^{226}Ra levels <111 mBq.L^{-1} (included are Dunlop and Elliot Lakes).

Muskrat were assigned to a MOE water sampling station, and a corresponding site class, depending on the location of capture. Animals collected close to a MOE water sampling station were assigned to that station and subsequently included in the appropriate site class (-high or -low) corresponding to its average total ^{226}Ra water value. When animals were taken at a location influenced by water passing two MOE sample stations, an average water quality value of the intermediate location was calculated using all sample data available from both MOE stations. Specimens acquired in areas lacking a sampling station were assigned to the value of the closest MOE station downstream in the water course.

ANIMAL SAMPLES

Muskrat were trapped in the Elliot Lake, Control-local and Control-distant areas from May to mid-June in 1985 and 1986. A scientific collecting permit was issued by the Ministry of Natural Resources for trapping out of season. Traps were placed near muskrat houses, shoreline runways, or in stands of cattails, a favoured food item of the animal. Apples were used as bait (Aldous 1946) in NATIONAL™ traps, size 1, and animals were killed with ether upon capture. In the Elliot Lake area, trapping locations were selected where suitable muskrat habitat corresponded with known water levels of ^{226}Ra. The number of traps varied at each location and from day to day throughout the sampling periods. Traps were visited at least once a day and twice a day when possible. Registered trappers provided muskrat carcasses from locations in the Elliot Lake, Control-local and Control-distant areas during the fall 1986 harvest.

Location and date of capture for all animals were recorded. All carcasses were individually labelled, bagged and deep frozen (-20°C) within 8 hours of capture for animals trapped by the authors, or within 3 days of capture by registered trappers.

PLANT SAMPLES

Cattails (*Typha latifolia*) were abundant on all locations where muskrat were trapped and are a major food of this species (Perry 1982). Three samples were collected in June 1986 at the Elliot Lake, Control-local and Control-distant areas. Sampling within the study area occurred at two locations where ^{226}Ra water values were high ($\geq$148 mBq.L^{-1}). These locations were at the Panel Mine waste management area below tailings Dam 'A' (MOE 1981), and along the Serpent River approximately 0.5 km downstream from the point of entry of effluent from the Quirke waste management area. These samples were combined (since all samples were within the Elliot Lake-high site) and constituted samples representing the Elliot Lake-high site. No cattail samples were collected from locations in Elliot Lake-low sites. Cattail samples were collected from the Control-local area (Sagard township) north of Elliot Lake, and the Control-distant area in Sudbury, Ontario. Plants were dug out so as to obtain as much root (including rhizome) as possible. Once unearthed, cattails were shaken to remove adherent soil and placed in plastic bags for transport to the laboratory.

SAMPLE PREPARATION

Animals: Dissection and Tissue Preparation

Frozen muskrat were thawed, weighed, dissected and sexed. As most animals had been intentionally skinned prior to acquisition, carcasses were weighed minus the skin using an OHAUS^{TM} heavy duty balance (capacity 30 kg) to 1 g. Left and right hind legs, skull and reproductive organs (ovaries, and uterus with any fetuses, in females; testes and penis in males) were removed and preserved. Observation of reproductive organs (upon removal at dissection) permitted definite sex determination of the animals used in this study (Anderson 1965). Bones of the right hind leg (femur, tibia and fibula), were selected for analysis and were prepared as follows: most muscle was removed with surgical scissors then bones were placed in sealed plastic bags and immersed in boiling water for 1 h. This facilitated removal of adherent tissue later peeled and scraped off with a scalpel. Femur, tibia and fibula of one leg comprised each muskrat bone sample.

Age Determination

Muskrat specimens were assigned to three age classes on the basis of tooth development and molar fluting. The first upper right molar from the skull of each specimen was removed and examined using the method described by Olsen (1959) and Pankakoski (1980): Juvenile (J) (7 months of age or less) molars show little or no root development and molar fluting ends deep in the alvelolar socket; Subadult (SA) (8 to 12 months of age) molars have moderate root development and fluting barely emerges from the bone line; Adult (A) ($\geq$1 year old) molars have highly developed roots and molar fluting extrudes well beyond the bone line.

Plants: Tissue Preparation

Cattails were washed in distilled water to remove excess substrate, partitioned into root (includes rhizomes), stem and leaf components, placed in individual paper bags and air-dried overnight.

Ashing, digestion, and ^{226}Ra estimations of bone tissue samples were carried out as described elsewhere (Clulow et al. 1988X). Air dried samples (circa 10 g) of root (includes rhizomes), stem, and leaves of cattail plants from all areas sampled were placed in individual porcelain crucibles and dried at 70 to 80°C to constant weight (to 0.01 g).

QUALITY ASSURANCE

Reliability of analytical methods used in this study for the detection of ^{226}Ra was assessed in the following manner: a) samples of cow shank bone (U.S. Bureau of Standards reference material), spiked with known amounts of ^{226}Ra and ^{133}Ba, were measured with the standard method and showed recovery rates of 98 ± 10%; b) standard ^{226}Ra samples from CANMET were run from time to time through the analytic procedure and results compared to the known values.

STATISTICAL ANALYSES

Ra-226 concentrations in Elliot Lake area muskrat bone samples were analyzed separately for sex, age, and area or site variations with the last two being considered most important. Sex and age variation was tested for significance only to justify pooling data from animals of different ages and sexes; seasonal variation could not be tested as sample sizes were too small. Initially, nonparametric tests were performed on differences among groups. Mann-Whitney U tests (two tailed) were applied when comparing two groups. Significance was indicated ($P < 0.05$) when $U_{calculated} > U_{table}$, (alpha = 0.05) (Sokal and Rohlf, 1987). Kruskal-Wallis tests were employed for comparisons among three or more groups. Significance was indicated ($P < 0.05$) when $K_{calculated} > \chi^2_{table}$ (alpha = 0.05) (Lapin 1979). As no significant differences were found among them ($P \geq 0.05$), sex and age categories were pooled and the data re-analyzed. Animals from both Control areas were pooled. Raw data from site categories (Elliot Lake-high and -low), and control areas (-local and -distant) were tested for normality using a Chi square (χ^2) goodness-of-fit test (Zar 1974). Data were $\log_{10}$-transformed to normalize them, then area and site categories were tested at the 5% level using a one way analysis of variance (ANOVA) on transformed data. Fisher's protected LSD test (PLSD) was then performed on area and site groupings to see if significant differences occurred among Elliot Lake sites and Control areas.

CALCULATIONS AND DATA PRESENTATION

Values are expressed throughout as $mBq.g^{-1}$ dry weight of plant or animal tissue and $mBq.L^{-1}$ for water from MOE sites, except where noted. This form facilitates comparison of these data to those of other workers.

Concentration ratios (f_{1e}) were calculated using the following formula (after ICRP 1978):

$$f_{1e} = \chi_1/\chi_e$$

where, χ_1 = radionuclide concentration in animal tissue ($mBq.g^{-1}$ wet weight)

and, χ_e = radionuclide concentration in water ($mBq.mL^{-3}$) or vegetation ($mBq.g^{-1}$ wet weight).

Confidence intervals (95%) were calculated for mean skeletal ^{226}Ra concentrations and transfer parameters were based on these values. For these calculations radionuclide concentration in bone was expressed as $mBq.g^{-1}$ wet weight, as required by the standard formula above. Radionuclide concentration in diet items needed to be calculated on a wet weight basis at the time of consumption. This presented a problem since moisture content of vegetation varies with time of day and season due to water stress (Wilson et al. 1953; Boyer 1968), and with changes resulting from absorption or loss of water after removal from the living tree. Leaves and branches may lose varying amounts of water by evaporation if left exposed on the bank, or may absorb unknown quantities of water during storage. Tissue moisture is also lost in varying amounts due to differences in handling, and the time and method of sample storage prior to analysis. As a result, values for cattails were calculated as $mBq.g^{-1}$ dry weight and adjusted to standard moisture contents of 74, 81, and 84% for leaves, stems and roots respectively; values were arrived at by measuring samples on collection and after drying to constant weight.

RESULTS

ANIMALS

A total of 81 muskrat specimens were trapped or purchased from registered trappers in the period spring 1985 through late fall 1986. Table 1 summarizes animal specimens procured.

Comparison of ^{226}Ra levels in bone of ten male and nine female animals of the same age class (subadult), from the Elliot Lake-high site indicated the difference to be non-significant (Mann-Whitney, $U_{calculated} = 58 \leq U_{table} = 70, P \geq 0.05$). This result warranted pooling of sex groupings. Similarly, ^{226}Ra levels in muskrat of three age classes (juvenile, subadult and adult), did not differ significantly in eleven muskrat taken in the Elliot Lake-low site (Kruskal-Wallis, $K_{calculated} = 4.66 \leq \chi^2_{table} = 5.99$ $P \geq 0.05$), and 30 animals from Elliot Lake-high sites ($4.52 \leq 5.99$, $P \geq 0.05$); on this result, age classes were pooled.

AREA AND SITE EFFECTS

Bone ^{226}Ra levels of Elliot Lake and Control area groups were tested for normality with χ^2 goodness-of-fit (Zar 1974). Logarithmic transformation of the data was carried out as some site groupings showed a significantly non-normal distribution. For example, when the distribution of raw ^{226}Ra values from 30 muskrat from the Elliot Lake-high were tested, $\chi^2_{calculated} = 39.52 > \chi^2_{table} = 14.07$, $P < 0.05$, indicating significant deviation from a normal distribution. When re-analyzed after $\log_{10}$-transformation, the same data approached normality: $\chi^2_{calculated} = 2.84 < \chi^2_{table} = 12.59$, ≥ 0.05. Similar testing showed that most Elliot Lake and Control groups were normally distributed when data were $\log_{10}$-transformed. The exception was Control-distant data, however, its deviation from normality was modest: $\chi^2_{calculated}$

= 24.83 χ^2_{table} = 14.07, $P < 0.05$, and the data were treated as belonging to a normally distributed population for statistical purposes. Elliot Lake and Control population data ($\log_{10}$-transformed) were compared. Groups (Figure 2) varied significantly by location (ANOVA, $F_{3,76} = 9.916$, $P < 0.05$). Data have been back-transformed for graphical presentation. Fisher's PLSD test when applied at the 5% level of significance, showed values at the Elliot Lake-high site were significantly higher than Elliot Lake-low site and Control area values, Elliot Lake-low and Control-local area values did not differ significantly:

	Control -distant	Elliot Lake -low	Control -local	Elliot Lake -high
n =	24	11	16	30
mean ^{226}Ra conc. in bone (mBq.g^{-1} dry wt., back-transformed)	11.5	<u>61.6</u>	<u>92.8</u>	468.0

(means underlined are not significantly different, $P \geq 0.05$)

VEGETATION

Mean concentrations of ^{226}Ra (mBq.g^{-1} dry wt.) in washed *Typha latifolia* samples from the Elliot Lake area were: 275.9 in leaves, 248.2 in stems, and 1,135.0 in roots (n = 6 in all cases).

WATER

Average total ^{226}Ra water values calculated from the MOE water quality data for individual and intermediate water sample stations in Elliot Lake, Ontario, ranged from 75 to 978 mBq.L^{-1} in Elliot Lake-high areas and 7 to 72.5 mBq.L^{-1} in Elliot Lake-low areas. The Control-distant area near Sudbury had an average ^{226}Ra water level of 9.0 mBq.L^{-1}. Water quality data were not available from the Control-local area.

Ra-226 IN BONE AND WATER

Ra-226 levels in bone of animals from Elliot Lake-high and -low sites and the Control-distant area were considered in relation to the average total ^{226}Ra concentrations in water at their places of origin. Positive regression coefficients (b; from the regression equation Y = a + b x) resulted for muskrat skeletal concentrations when compared with water data (all data were log-transformed) (b = 1.16, Figure 3). The correlation coefficient (R) was high (0.87). ANOVA indicated regression of $\log_{10}$-transformed bone ^{226}Ra concentrations on ^{226}Ra concentrations in water was highly significant (F_{59} = 190.7, P <0.0001, Table 2).

CONCENTRATION RATIOS

Concentration ratios, between vegetation items used in the diet and bone, were calculated. Vegetation values were converted to $mBq.g^{-1}$ wet weight as described previously. Table 3 provides a synopsis of concentration ratios from plant parts to muskrat bone.

The range in concentration ratios can be restricted by calculating a single value for the ^{226}Ra content of the diet. As no information is available on the percentage of the diet consisting of cattails, or the contribution of each plant portion to the diet, it was assumed they eat only cattails and that consumption of leaves, stems, and root material is in the ratio of 1:1:1. The weighted average concentration calculated by taking into account contributions for each component of the cattail is 100.17 $mBq.g^{-1}$ wet weight. Using this calculated dietary value, and the 95% confidence intervals of bone level (Table 3), then the 95% confidence limits for the concentration ratio from diet to bone of muskrat taken in the Elliot Lake-high site is 1.9 to 4.0.

DISCUSSION AND CONCLUSIONS

AREA AND SITE EFFECTS

An objective of this study was to compare ^{226}Ra concentrations in bone tissue of animals from contaminated areas to those in animals from uncontaminated control areas. Although a significant difference in mean bone ^{226}Ra level was observed between Elliot Lake-high sites and Control areas, the Elliot Lake-low sites did not differ from the Control-local area. This indicates radionuclide contamination to be a localized phenomenon: animals living only a few hundred metres from high levels of mining-associated radioactive waste do not carry a significantly higher burden of ^{226}Ra than those from an undisturbed control area close to Elliot Lake.

Findings in this study confirm those of previous animal studies from the Elliot Lake area and other U mining areas. Wren et al. (1987) reported measurable ^{226}Ra levels in otter from the Elliot Lake area but not in animals from a control area. Cloutier et al. (1985a,b, 1986) and Clulow et al. (1986) showed meadow vole taken near the Elliot Lake tailings had significantly higher bone ^{226}Ra levels than did controls and that hare faeces from the same areas varied similarly. Swanson (1985) found significantly higher ^{226}Ra levels in all fish species studied from her contaminated area (Beaverlodge Lake, Saskatchewan) compared to controls. Swanson (1985) also notes similar trends in fish sampled around mines or areas with high natural background levels in Ontario, Canada (Elliot Lake and Bancroft), in Colorado, U.S.A., and in Czechoslovakia. Significant differences in animal bone concentrations between Elliot Lake-high and -low sites suggest that contamination is localized.

SEX AND AGE EFFECTS

The results obtained in this study indicate that ^{226}Ra concentrations in bones of animals taken near tailings do not vary by sex or age. These findings differ from those in other reports. Cloutier (1984) and Cloutier et al. (1983, 1985a,b, 1986) found ^{226}Ra skeletal concentrations in meadow voles from Elliot Lake did not differ between sexes but age effects were seen in different seasons; adults had the highest concentration in the spring and subadults were higher in the fall. Muth and Glöbel (1983) also reported an age dependence for ^{226}Ra concentration in their study on human bone. Two maxima of skeletal ^{226}Ra concentrations were determined coinciding with periods of high skeletal growth from birth to 1 y old and between 10 and 16 y of age. Bruenger et al. (1983 demonstrated a decrease in skeletal ^{226}Ra concentrations with age in beagles, and Hantke (1959), and Muth et al. (1960) reported age dependence for ^{226}Ra in the bone of chickens. On the other hand, Montalbano et al. (1983) studying duck muscle tissue found that mean concentrations of ^{226}Ra were not different between sex categories and among age classes. Swanson (1983, 1985) reported no significant relationship between age or sex and radionuclide concentration in all fish species studied in Northern Saskatchewan.

Radionuclide uptake and retention by mammals may be influenced by biological variables other than environmental exposure. Such variables include season of animal collection, sex, age, species and diet (Wren 1986). These factors may modify bioaccumulation of metals independently, for example, age effects on ^{226}Ra uptake in beagles were demonstrated by Lloyd et al. (1976, 1983) when all parameters were controlled under laboratory conditions. Compounding effects may also occur. An example is the season and age interactions affecting ^{226}Ra uptake under natural conditions demonstrated by Cloutier (1984) in the meadow vole at Elliot Lake. Wren (1986) provides additional examples of the influence of such variables on metal uptake.

Differences in ^{226}Ra accumulation between sexes of Elliot Lake area muskrat were analyzed and showed no significant differences. This lack of variation in skeletal ^{226}Ra concentrations was not surprising. As mentioned in the studies above, other animals did not demonstrate different levels between sexes and since feeding habits are not known to differ between sexes in the muskrat, sex differences were not anticipated.

Conflicting results were apparent in regard to age class and tissue ^{226}Ra concentration. Most mammal studies mentioned previously showed that age had an effect on ^{226}Ra accumulation. During periods of rapid bone growth ^{226}Ra is taken up in preference to Ca and built into the hydroxyapatite crystals of the bone (Muth and Glöbel 1983). However, ^{226}Ra is more easily eliminated than Ca when bone is not in a major growth stage and diffusion and recrystallization are the prominent processes occurring (Muth and Glöbel 1983). Thus one would expect differences in ^{226}Ra accumulation with differences in age, a phenomenon which was not observed in this study. Finer division of age classes with more sampling from birth to one year old might reveal a bone ^{226}Ra to age relationship.

SPECIES COMPARISONS

Muskrat had significantly higher levels of ^{226}Ra in their bones than did beaver from corresponding Elliot Lake sites and the Control-local area (Clulow et al. 1988X). Beaver from the Control-distant area, however, had higher skeletal ^{226}Ra levels than muskrat.

Wren (1986) suggested differences between species is primarily attributable to diet. This appears to be supported by this study. The cattail, the most important hydrophyte in the muskrat diet, had higher ^{226}Ra tissue levels than aspen and white birch (important food items in beaver diet (Hill 1982; Proulx and Gilbert 1983). When obtaining food the muskrat digs in the substrate on lake and pond bottoms, especially during winter when food supplies are limited (Perry 1983). Such feeding activities, among sediments high in ^{226}Ra, increase the probability that particulates adhering to plant parts will be ingested. Beaver, in contrast, consume above-ground parts of terrestrial plants (woody material, and leaves) with lower levels of ^{226}Ra. This food, stored in caches for winter consumption, may decrease foraging for underground roots and tubers high in ^{226}Ra. Submersion in contaminated water for an extended period (during winter) might result in tree branches absorbing ^{226}Ra; a study of radionuclide levels in cached food would clarify this.

Differences in ^{226}Ra accumulation by species have been reported: Swanson 1983 demonstrated differences among fish studied and related this difference to feeding habits and diet. Bottom-feeding fish (lake chub, spottail shiner, nine-spine stickleback, and trout perch) generally had higher radionuclide levels than large predator fish analyzed (lake trout, whitefish, and white sucker). These fish in turn were higher than a planktivorous species (cisco) analyzed. Among the large predator fish analyzed ^{226}Ra bone concentrations varied between species and Swanson also attributed the difference in ^{226}Ra accumulation to their diet (lake trout feeding mainly on cisco < whitefish feeding on chironomids, lake chub, stickleback, and benthic algae < white sucker feeding on benthic invertebrates). Lloyd et al. (1983) also demonstrated a difference in ^{226}Ra retention among two varieties of the same species: St. Bernard dogs retained a greater fraction of injected ^{226}Ra than beagles of the same age.

VEGETATION

Data obtained for cattails in this study confirm vegetation data from Kalin (1988). Kalin reports mean ^{226}Ra values of 11.1, 22.2 and 2701.0 for cattail stems, leaves and roots, respectively, which are in general agreement with data from this study.

CONCENTRATION RATIOS

Concentration ratios calculated from cattail parts to bone of muskrat from Elliot Lake-high sites ranged from 1.1 to 8.5 (Table 3). These figures are comparable to those reported for the beaver, another aquatic herbivorous mammal sampled from the area, which ranged from 0.6 to 5.4 between various plant items and bone; from a mixed diet to bone the value was 1.3 (Clulow et al. 1988X). A study of wild meadow voles trapped on Elliot Lake tailings (Cloutier et al. 1986), contains data on ^{226}Ra levels in vegetation used as vole food (211 mBq.g^{-1}, dry weight), and bone (1,506 and 703 mBq.g^{-1} dry weight in animals trapped in summer and fall, respectively), yielding CRs of 7.1 and 3.3 with corresponding transfer coefficients of 4.6×10^{-2} and 2.8×10^{-2}. Burns et al. (1987), working with young animals in the laboratory, reported 59 mBq.g^{-1} (dry weight) in bone of voles fed a diet spiked with ^{226}Ra (350 mBq.g^{-1} dry weight) indicating a CR of 0.16 and a corresponding transfer coefficient of 1.4×10^{-3} during the period of rapid bone growth studied. Concentration ratio between vegetation items and bone of moose in the Serpent River drainage had been calculated as 3.03 (MacLaren 1987); this calculation is based on small sample sizes and pooling of Serpent River drainage data with data from an uncontaminated area. The CR between aspen leaves and bone of local ruffed grouse (Bonasa umbellus) was 1.04; values between other plants eaten and bone were less than one (Clulow et al. 1988XX). Muth and Glöbel (1983) calculated a concentration ratio of 23.7 for humans which is higher than values reported here.

BONE AND WATER LEVELS OF ^{226}Ra

Muskrat showed a positive regression between skeletal ^{226}Ra values and average total water ^{226}Ra values associated with the areas of procurement. The correlation coefficient was high (0.87) and ANOVA indicated that regression was significant. This suggests that muskrat reflect environmental (water) ^{226}Ra gradients present in the Elliot Lake area. The variation indicated by the correlation coefficient values obtained in this study for muskrat may be a reflection of animal movement and length of habitation in the area of procurement. Muskrat movement is relatively restricted compared to the beaver. Muskrat movement during spring breakup or in times of social and/or environmental stresses has been reported up to 34 km. This species, however, is relatively sedentary with few movements being greater than 150 m (MacArthur 1978; Perry 1982).

On the basis of the concentration ratios calculated for muskrat with respect to vegetation, and regression analysis performed on muskrat ^{226}Ra levels with respect to environmental (water) ^{226}Ra levels, it is concluded that muskrat reflect radionuclide levels in the environment to a degree that makes the animal of interest as a bioindicator of this contamination.

IMPLICATIONS

Accumulation of ^{226}Ra in bone implies a higher rate of retention than elimination. Upon initial exposure to ^{226}Ra, concentration might be expected to be higher in trabecular bone which remodels more rapidly than cortical bone. A gradual increase in ^{226}Ra concentration will occur in bones with larger cortical portions as the remodelling process continues (Lloyd et al. 1976). In addition, ^{226}Ra may be more tightly locked into the bone when incorporated into the cortical portion, which is relatively inaccessible to the circulation (Mays et al. 1975). High skeletal burdens of ^{226}Ra can have major effects. Lloyd et al. (1976) found beagles injected with high concentrations of ^{226}Ra (3.7 x 105 Bq.kg^{-1}) had greater ^{226}Ra retention in trabecular bones (lumbar vertebrae and sternum) than in cortical bones (radius, ulna, fibula, and tibia). They attributed this difference in retention levels to observed radiation damage of the bone remodelling process at high skeletal concentrations of ^{226}Ra. Raabe et al. (1983) also studied skeletal burdens in beagles. They noted the occurrence of primary bone cancer, osteosarcoma, after chronic irradiation by skeletal deposited ^{226}Ra.

Schoeters (1982) working on mice injected with ^{226}Ra noted radiation damage to haemopoietic marrow stem cells from skeletal burdens of ^{226}Ra. He also mentioned that the cells considered to be the targets for bone tumor development, osteoprogenitor cells, may also incur radiation damage. Haemopoiesis and bone formation seem to be closely related and originate from the same region (Schoeters 1982). On the other hand, studies on chronically irradiated wild meadow voles (receiving approximately 6 x 10^3 times background levels) by Ross (1984, 1985) revealed no increase in the incidence of chromosome aberrations in bone marrow cells. Ross also reported a study on red-back voles (_Clethrionomys gapperi_) living at exposures of approximately 10 times background level which demonstrated similar results. In the present study no major effects, such as those mentioned above for high skeletal burdens, are believed to have occurred in muskrat as skeletal burdens of ^{226}Ra were relatively low in both cases.

TRANSFER TO OTHER SPECIES

Predation on muskrat by mink (_Mustela vison_) and raccoon (_Procyon lotor_) is well recorded (Perry 1982). Muskrat are occasionally taken by avian predators such as bald eagles (_Halioeetus leucocephalus_), great horned owls (_Bubo virginianus_) and ferruginous hawks (_Buteo regalis_) (Willner et al. 1980). Utilization of muskrat as food items by predators varies, and can be quite high depending on availability of alternate prey items. However, meat and internal organs from muskrat carcasses, which are presumed to be lower in ^{226}Ra burden, are eaten more readily than bone which is less easily assimilated by predators and scavengers. The spread of ^{226}Ra to predators through the consumption of muskrat or beaver is probably quite slow.

The recommended International Commission on Radiological Protection (ICRP) limit for intake of ^{226}Ra by radiation workers is 2 x 10^5 Bq.y^{-1} (ICRP 1979a,b, 1980). Limits for other individuals are much lower: from 2 x 10^4

$Bq.y^{-1}$ to 4×10^3 $Bq.y^{-1}$ in the case of prolonged exposure. To approach the lowest limit, human consumers would have to eat more than 10 kg of raw muskrat bone in one year, or about 27 g daily. It is unlikely that intake from soups and broth is as much as 1% of this. If loss of radionuclide occurs during preparation of muskrat for the table then the amounts required to approach the ICRP limits would be even greater. Halford (1983) reported a loss in ^{137}Cs. ^{134}Cs, ^{60}Co, ^{140}La and ^{110}Ag when fresh waterfowl were cooked (dry roast). The amount of ^{137}Cs and ^{34}Cs in the carcasses declined by about 27% with cooking. Trappers and hunters of the Elliot Lake area report that muskrat (along with beaver, grouse, rabbit, and hare) are roasted or stewed with onions and tomatoes, before eating. The stewing process, with its mildly acidic environment due to tomatoes, may serve to elute some ^{226}Ra and Ca from the bones in the carcass. The level of such liberation is unknown. As consumption of muskrat by the people of the area, especially members of the Serpent River Band living near the river mouth, is not as prevalent as in years past (Chief Commanda, personal communication), levels of consumption likely to supply ^{226}Ra in amounts required to exceed the ICRP limits seem unlikely.

CONCLUSIONS

Muskrat sampled at Elliot Lake area had generally elevated levels of skeletal ^{226}Ra with the highest levels in animals from contaminated waters near tailings; those from less contaminated waters did not differ significantly from samples taken in a control area near Elliot Lake; both these groups had higher levels than samples from a distant control area (Sudbury, Ontario). Radionuclide level in muskrat bone was related neither to sex nor to age; this latter finding differs from observations on some other animal species.

Concentration ratios between dietary items (plant parts) and bone of muskrat taken at Elliot Lake-high sites were calculated to range from 1.1 to 8.5. These values were in general agreement with values calculated for other mammal and bird species sampled in the area. As transfer parameters appear relatively low from diet to muskrat bone tissue, it seems unlikely that adverse effects will accrue to muskrats near Elliot Lake U tailings. Since skeletal tissue is less easily assimilated by predators than soft tissue, which in turn may have lower levels of ^{226}Ra, spread of the radionuclide through trophic levels is probably slow and hazard to predators seems unlikely.

The muskrat is judged to be a useful indicator of environmental (water) levels of ^{226}Ra on the basis of the close correlation of bone and environmental levels of the radionuclide.

Human consumption would have to be in excess of 10 kg (raw) muskrat bone each year to approach the ICRP limits; this seems unlikely.

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Table 1. Number of muskrat specimens studied from Elliot Lake and Control areas.

	collection periods			
	Spring 1985	Spring 1986	Fall 1986	Total
areas and sites				
Control-distant area	5	10	9	24
Control-local area	4	4	8	16
Elliot Lake-low sites	-	-	11	11
Elliot Lake-high sites	8	16	6	30
Totals:	17	30	34	81

Table 2. Analysis of variance table for regression of $\log_{10}$-transformed ^{226}**Ra** levels ($mBq.g^{-1}$ dry weight) in the bone of muskrat from Elliot Lake and Control-local against $\log_{10}$-transformed ^{226}**Ra** ($mBq.l^{-1}$)levels in water from the same locations.

Source	DF	SS	MS	F-Test
Regression	1	34.12	34.12	190.7
Residual	59	10.56	0.179	$P < 0.0001$
Total	60	44.68		

DF = degrees of freedom, SS = sum of squares, MS = mean square

Table 3. Concentration ratios between cattail parts and bone of muskrat at the Elliot Lake-high site.

compartment	part	[^{226}**Ra**] (mBq.g^{-1} dry wt.)	moisture content %	[^{226}**Ra**] (mBq.g^{-1} wet wt.)	concentration ratios* (plant to bone)
muskrat bone		-	-	194.09 - 399.94*	-
cattail	leaf	275.9	74	71.7	2.71 - 5.58
	stem	248.2	81	47.2	4.11 - 8.47
	root	1,135.0	84	181.6	1.07 - 2.20

* 95% confidence limits

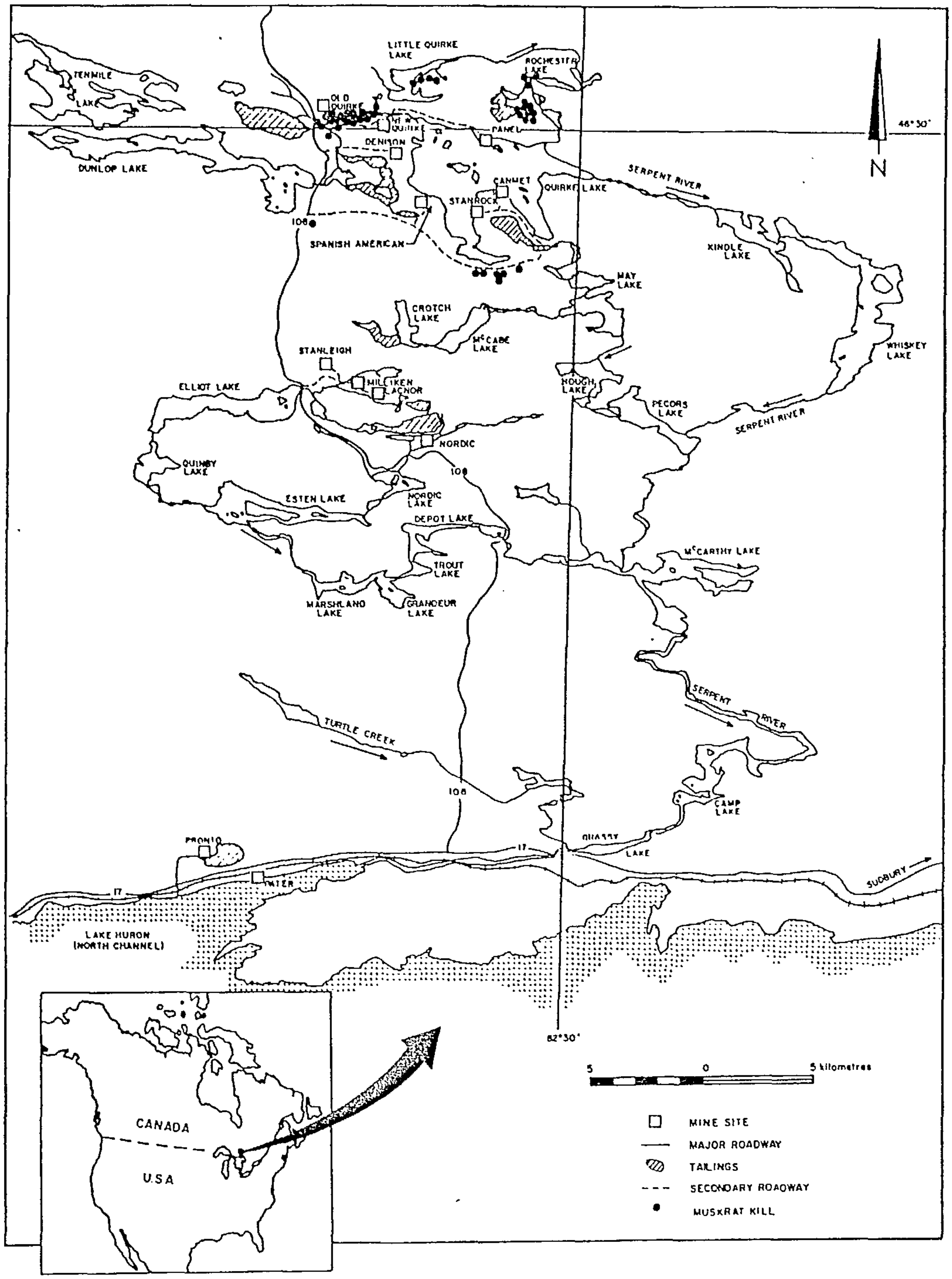

Fig. 1 - Collection sites of muskrat specimens in the vicinity of Elliot Lake, Ontario.

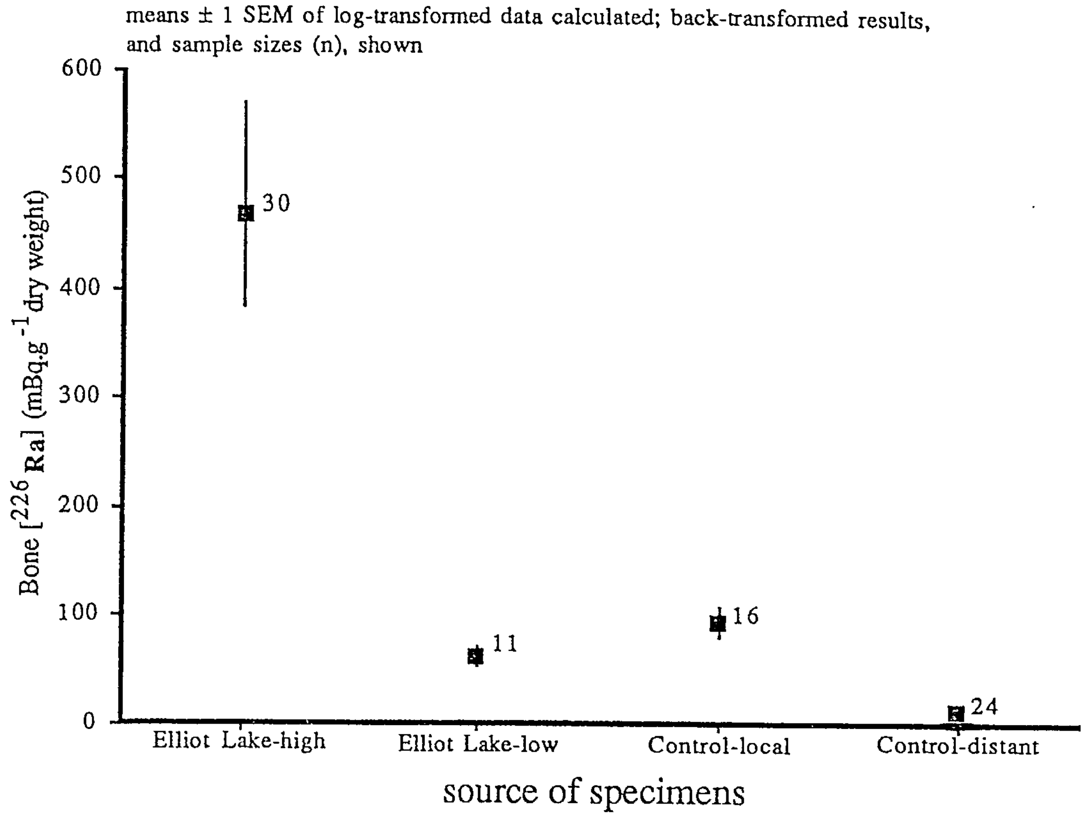

Fig. 2 - Levels of ^{226}Ra (mBq.g^{-1} dry weight) in muskrat bone from Elliot Lake and Control locations.

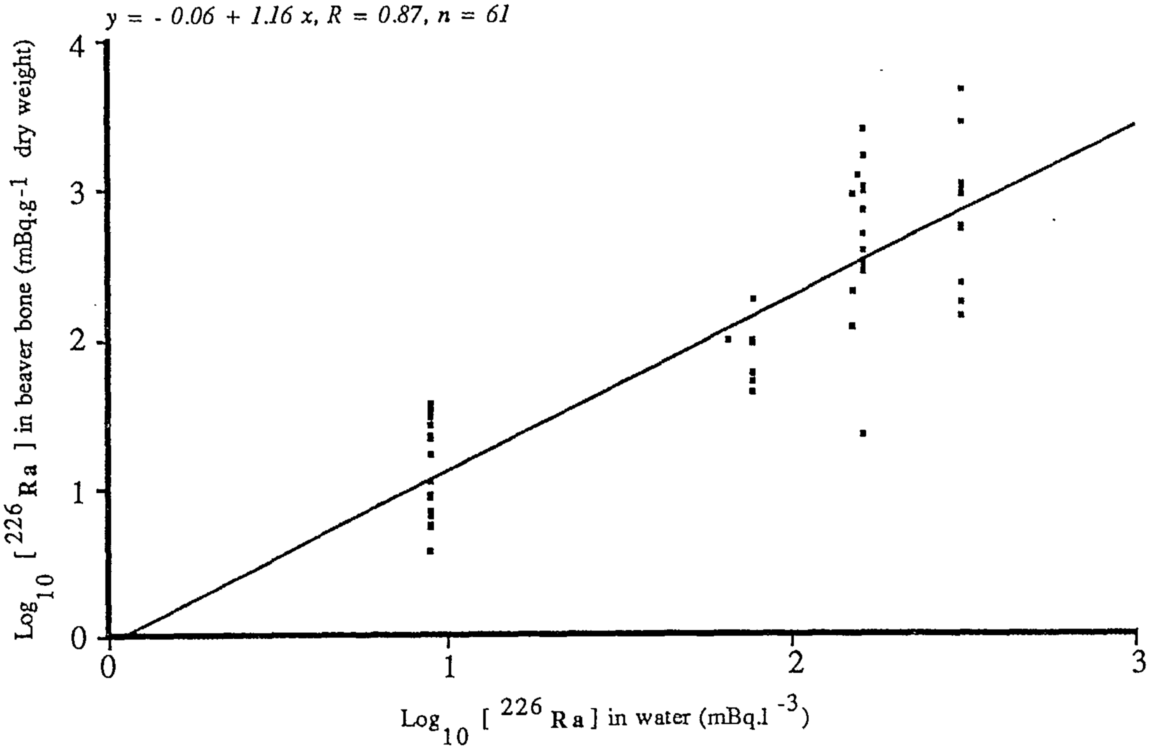

Fig. 3 - Levels of ^{226}Ra (log$_{10}$ mBq.g^{-1} dry weight) in muskrat bone from Elliot Lake and Control-local locations in relation to log$_{10}$-transformed ^{226}Ra levels (mBq.L^{-1}) in water sampled at the same locations.