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ESTABLISHING A DATASET FOR A 'REFERENCE SITE'

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1. INTRODUCTION

The ICRP published their framework for radiation protection of the environment in *Publication 108* (ICRP 2008). This describes the use of Reference Animals and Plants (RAPs) as the basis for the framework. Publication 108 presented dose coefficient values for the selected RAPs and also reviewed data on the effects of ionising radiation to suggest Derived Consideration Reference Levels (defined as *a band of dose rate within which there is likely to be some chance of deleterious effects of ionising radiation occurring to individuals of that type of RAP*) for each RAP.

In summer 2010 the ICRP released a further report on their protection framework for consultation. This report presented transfer parameter values (organism-media concentration ratios) for adult life-stages of the Reference Animals and Plants (<http://www.icrp.org/page.asp?id=65>). However, there were many radionuclide-RAP combinations for which there were no data. The report also raised the possibility of identifying a series of sites where samples of each Reference Animal and Plant, and their different life-stages, could be collected and analysed. It was suggested that the resultant data would constitute a set of 'reference values' analogous to approaches used by the ICRP for human radiological protection.

Here we describe the sampling of Reference Animals and Plants from a single terrestrial site and summarise analytical results available to date.

2. TERRESTRIAL REFERENCE ANIMALS AND PLANTS AND CURRENT DATA AVAILABILITY

A RAP is defined by the ICRP as: '*a hypothetical entity, with the assumed basic biological characteristics of a particular type of animal or plant, as described to the generality of the taxonomic level of Family, with defined anatomical, physiological, and life-history properties, that can be used for the purposes of relating exposure to dose, and dose to effects, for that type of living organism*' (ICRP 2008). The ICRP described eight terrestrial, four marine and four freshwater RAPs. The RAPs for terrestrial ecosystems are presented in Table 1.

The pending ICRP report presenting transfer parameters for RAPs (<http://www.icrp.org/page.asp?id=65>) presents equilibrium whole organism concentration ratios (CR), where for terrestrial ecosystems:

$$CR = \frac{\text{Activity concentration in whole organisms (Bq kg}^{-1} \text{ fresh weight)}}{\text{Activity concentration in soil (Bq kg}^{-1} \text{ dry weight)}}$$

The report used the same database as that used to prepare a forthcoming IAEA handbook presenting radionuclide transfer values for wildlife (Howard et al. 2011). When summarised for the RAPs at the defined Family level summary, CR values could be derived for relatively few elements. For example, Reference Deer, Duck and Rat all had data for 10 or fewer elements to enable CR values to be estimated while there were no data available for Reference Bee. It was to address this lack of data that the ICRP suggested a series of ('reference') sites should be identified from which samples of each Reference Animal and Plant, and their different life-stages, could be systematically collected and analysed.

Table 1. Terrestrial Reference Animals and Plants as defined in ICRP (2008) and species sampled during this study.

RAP	Family	Species sampled
Wild Grass	<i>Poaceae</i>	<i>Molinia caerulea</i> (Purple moor grass)
Pine Tree	<i>Pinaceae</i>	<i>Picea sitchensis</i> (Sitka spruce)
Bee	<i>Apidea</i>	<i>Apis</i> spp., <i>Bombus</i> spp., <i>Nomada</i> spp.
Earthworm	<i>Lumbricidae</i>	<i>Lumbricidae</i>
Rat	<i>Muridae</i>	<i>Apodemus sylvaticus</i> (Wood mouse)
Deer	<i>Cervidae</i>	<i>Capreolus capreolus</i> (Roe deer)
Duck	<i>Anatidae</i>	Not sampled
Frog	<i>Ranidae</i>	Not sampled

3. METHODOLOGY

3.1 Study site and sampling

Our study site was located in a managed forest predominantly planted with coniferous species in north-west England. The majority of samples were obtained from a *circa* 0.4 km² area during the summer of 2010. The area was largely a mixture of: (i) established coniferous plantation with an understory of predominantly grass and sedge species; (ii) and on wetter areas *Molinia caerulea* clumps with some shrubs and small broad leaf trees.

It was possible to sample all terrestrial RAP species with the exception of Reference Frog and Duck. The species sampled for each RAP are listed in Table 1. The three *Picea sitchensis* sampled were approximately 15 year old self seeded trees. Samples of trunk, branches, needles and cones (one tree only) were retained. Bees were collected using pan traps (Westphal et al. 2008) which were either coloured white or painted yellow or blue using fluorescent paints. Where possible the pots were located close to flowers of similar colours. *Apodemus sylvaticus* were trapped using Longworth traps baited with oats, carrot and insect pupae and containing hay as a bedding material. Earthworms were collected by digging, before processing they were kept in aerated containers containing damp tissue paper to allow the gut to be evacuated.

Three *Capreolus capreolus* were shot in February 2011 from different locations in the larger area of forestry within approximately 2 km of the main sampling area.

Replicate soil samples (top 10 cm) were collected for all sample types. In the case of *C. capreolus* soils were collected in the general vicinity of where they had been shot and where there was evidence of deer activity (obvious signs of grazing, faeces and/or tracks). The geometric mean percentage loss on ignition for 43 soils was 32 % (ranging from 7 to 87 %). The range in soil pH values (determined in distilled water) was 3.9-8.1 (geometric mean = 5.5; n = 25).

3.2 Sample preparation and analyses

Mice samples were first weighed and then skinned, the gut removed and then dissected to obtain samples of muscle, liver, bone and gonad; the remaining organs were bulked. To obtain estimates of muscle and bone weights the remaining carcass was placed in a beetle (*Dermestes maculatus*) colony to clean the bone of all soft tissue. The majority of organs were removed and retained from the deer (including foetuses from two animals) the carcass was subsequently divided down the spine and the muscle and bone mass estimated for one half.

Soil, worm, plant and bee samples were either dried at 20°C or freeze-dried prior to stable element analysis; mammal samples were analysed fresh. When preparing samples care was taken to minimise contact with metallic instruments.

The analyses of specific samples were designed to best use the available materials and budget (i.e. not all samples were analysed for all determinants).

3.2.1 Gamma analyses

Soil and vegetation samples for gamma analyses were dried at 60°C prior to homogenisation and weighing into suitable sized containers (c. 130-700 ml). Samples were counted using hyper-pure Ge-detectors typically for 4 days. Resultant spectra were analysed using Canberra Apex-Gamma software and results decay corrected to the day of sampling. *A. sylvaticus* were analysed as bulked wholebody carcasses (complete with gut contents and pelts) prior to dissection.

3.2.2 ICP-MS analyses

Concentrations of 60 elements (see Table 2) were determined in samples using inductively coupled plasma-mass spectrometry (ICP-MS).

Samples were homogenised prior to analysis using ceramic blades or by crushing. Sub-samples of wood were created using a metal rasp. Triplicates of *M. caerulea* and *P. sitchensis* were analysed.

Aliquots of each sample plus certified reference materials were digested in a mixture of nitric acid and hydrochloric acid using quartz high pressure closed vessels and microwave heating (Perkin Elmer Multiwave). Quantification was by ICP-MS with collision cell (Agilent 7500ce). Reagent blanks and a reagent blank spiked with a known amount of each analyte were analysed with the test samples for recovery estimate purposes.

3.2.3 Carbon and nitrogen analyses

Samples were analysed on an Elementar Vario-EL elemental analyser which uses oxidative combustion and thermal conductivity detection to determine the C and N concentrations in samples.

4. RESULTS AND DISCUSSION

Currently results are available for all analyses other than the ICP-MS determinants for soil and *C. capreolus* samples (these should be available for presentation in June 2011). Consequently CR values cannot be calculated and presented here. However, it is evident that the work will result in data for

radionuclide-RAP combinations for which there were none previously. Table 2 summarises the elements which were detectable in each sample type. Note the table records the presence of a given element in any sample type for a given RAP, however, it may not be possible to calculate whole organism CR values in all instances if an element was detectable in one organ type only.

Table 2. Summary of sample types in which elements were measured above detection limits.

Element	RAP	Element	RAP	Element	RAP
Li	EW,PT,WG	Sr	All	Gd	All
Be	EW	Y	EW,PT,WG	Tb	EW,PT,WG
B	All ¹	Zr	All	Dy	EW,PT,WG
Na	All	Nb	EW,PT,WG,R	Ho	EW,PT,WG,R
Mg	All	Mo	All	Er	EW,PT,WG
Al	All	Ru	R	Tm	EW,PT,WG
K	All	Pd	EW,PT,WG,R	Yb	EW,PT,WG
Ca	All	Ag	B,EW,PT,WG	Lu	EW,PT
Ti	All	Cd	All	Hf	B,EW,PT,WG
V	EW	Sn	All	Ta	nd ²
Cr	EW,PT,WG,R	Sb	EW,PT,WG,R	W	EW,PT,WG,R
Mn	All	Te	EW,PT	Re	nd
Fe	All	Cs	All	Ir	nd
Co	All	Ba	All	Pt	EW
Ni	All	La	EW,PT,WG,R	Au	nd
Cu	All	Ce	EW,PT,WG,R	Hg	EW,PT,R
Zn	All	Pr	EW,PT,WG,R	Tl	B,EW,PT,WG
As	All	Nd	All	Pb	EW,PT,WG,R
Se	All	Sm	EW,PT,WG	Th	EW,PT,WG,R
Rb	All	Eu	All	U	EW,PT,WG

Sample type code: B – Reference Bee; EW – Reference Earthworm; PT – Reference Pine Tree; WG – Reference Wild Grass; R – Reference Rat. ¹All – element detectable in all sample types; ²nd – element undetectable in all sample types

Caesium-137 was detectable in a number of biota sample types. Activity concentrations in the whole body of *A. sylvaticus* ranged from <2 - 26 Bq ¹³⁷Cs kg⁻¹ fresh weight (fw), *P. sitchensis* trunk (4 - 9) x 10⁻² Bq ¹³⁷Cs kg⁻¹ fw, *C. Capreolus* muscle 0.7 – 9 Bq ¹³⁷Cs kg⁻¹ fw and *M. caerulea* 7 – 15 Bq ¹³⁷Cs kg⁻¹ fw. With the exception of ⁴⁰K no other gamma-emitter was routinely detected in biota samples. Caesium-137 was detectable in all soil samples ranging from 11 – 290 Bq ¹³⁷Cs kg⁻¹ dry weight.

The forthcoming IAEA handbook on transfer parameters for wildlife (see Howard et al. 2011) presents a specific activity approach for ¹⁴C rather than relying on CR values relating whole organisms and soil activity concentrations. There are, however, few compilations of carbon concentrations in wildlife. The data presented in Table 3, therefore, make a useful contribution in this respect.

Table 3. Arithmetic mean (\pm SD) carbon concentrations (g kg^{-1} fw).

Sample	Mean	SD	n
Bee	175	9.5	3
Earthworm	75	12.7	4
<i>M. caerulea</i>	156	2.1	3
<i>P. sitchensis</i> trunk	222	38.5	3
<i>P. sitchensis</i> needles	236	7.6	3
<i>P. sitchensis</i> cones	205		1
<i>P. sitchensis</i> seeds	216		1
<i>A. sylvaticus</i> muscle	127	5.0	3
<i>A. sylvaticus</i> bone	65	15.7	3
<i>A. sylvaticus</i> liver	132	11.5	3
<i>A. sylvaticus</i> testes	142	18.8	3
<i>A. sylvaticus</i> other soft tissues	114	5.0	3
<i>C. capreolus</i> liver	173	11.3	3
<i>C. capreolus</i> bone	109	3.8	3
<i>C. capreolus</i> muscle	144	19.7	3

Table 2 demonstrates that once all of results from the ICP-MS analyses are available then the work will have been beneficial in terms of providing CR values for RAP-radionuclide combinations for which there are current few or none. Even where concentrations were below detection limits the results may still be of value in setting benchmark CR values for the RAPs in the absence of any other data. The site used here could, with appropriate permissions and revisiting at the correct times of year, provide data for species of Reference Frog (including the different life-stages) and possibly the Reference Duck. However, the CR values which can be derived from this work will be site specific. How such 'reference site' data will be utilised within the ICRPs framework requires further elaboration (and we understand that the ICRP are in the process of doing this).

To provide wildlife CR values for a large number of elements from the same samples ICP analyses and neutron activation are increasingly being used. This is a cost effective way of addressing the need for data. However, there needs to be some consideration of the appropriateness of applying stable element data to estimate radionuclide transfer for different types of assessment. This is probably most especially true of the terrestrial environment where source terms of radionuclides and stable elements may differ.

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