

## Field effects studies in the Chernobyl Exclusion Zone: Lessons to be learnt

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### ABSTRACT

In the initial aftermath of the 1986 Chernobyl accident there were detrimental effects recorded on wildlife, including, mass mortality of pine trees close to the reactor, reduced pine seed production, reductions in soil invertebrate abundance and diversity and likely death of small mammals.

More than 30 years after the Chernobyl accident there is no consensus on the longer-term impact of the chronic exposure to radiation on wildlife in what is now referred to as the Chernobyl Exclusion Zone. Reconciling this lack of consensus is one of the main challenges for radioecology. With the inclusion of environmental protection in, for instance, the recommendations of the International Commission on Radiological Protection (ICRP), we need to be able to incorporate knowledge of the potential effects of radiation on wildlife within the regulatory process (e.g. as a basis on which to define benchmark dose rates).

In this paper, we use examples of reported effects on different wildlife groups inhabiting the Chernobyl Exclusion Zone (CEZ) as a framework to discuss potential reasons for the lack of consensus, consider important factors influencing dose rates organisms receive and make some recommendations on good practice.

### 1. Introduction

The 1986 Chernobyl accident led to the largest release of radioactivity to the terrestrial environment in the approximately 60 years of nuclear power production. In the weeks following the accident, the human population and farm animals were evacuated from an area of approximately 3500 km<sup>2</sup> around the reactor; this area was subsequently increased to 4760 km<sup>2</sup>. Approximately 2600 km<sup>2</sup> of this abandoned area is in the Ukraine and has become known as the Chernobyl Exclusion Zone (CEZ); the remainder is in Belarus. The area is highly heterogeneously contaminated by a number of radionuclides including <sup>137</sup>Cs, <sup>90</sup>Sr, <sup>241</sup>Am and Pu- isotopes (Kashparov et al., 2018); many shorter-lived radionuclides released by the accident have now decayed. However, dose rates in the CEZ remain sufficiently high (e.g. Beresford et al., 2019) that, based upon our existing understanding, we would anticipate radiation induced effects on many types of wildlife which may potentially impact on populations (ICRP, 2008).

In the aftermath of the accident, wildlife in some areas of the CEZ were exposed to extremely high dose rates with consequent significant detrimental effects being observed in a range of organism types (e.g. Gersk'kin et al., 2008). There is no challenge to such observations; the effects were clear and in line with our established understanding of the effects of radiation.

However, whilst the CEZ has offered the opportunity to conduct studies into the effects of chronic radiation exposure on wildlife there is considerable scientific debate with regard to reported studies

conducted in the area (e.g. Smith, 2008; Wickliffe and Baker, 2011; Mousseau and Møller, 2012; Beresford et al., 2012). This lack of consensus relates to studies conducted one or more decades after the accident. Because of the topic, radiation effects on wildlife, this lack of scientific consensus can have a high public profile and represents one of radioecology's key challenges.

### 2. Scientific conflict – some examples

Many of the reported studies conducted over the last 20 years report radiation induced effects at comparatively low dose rates (e.g. see Møller & Mousseau, 2011). An example of this is a study of the abundance of butterflies, bumblebees, grasshoppers, dragonflies and spiders conducted in 2008 (Møller and Mousseau, 2009). The authors report a negative relationship between abundance for each of the five taxa and radiation. The negative relationship extended into the range 0.01–0.1 μGy h<sup>-1</sup> which is considerably below any 'no-effect' benchmark used in regulatory assessments of the potential impact of radiation on wildlife (Howard et al., 2010) and at the lower end of the range anticipated for the natural background exposure of wildlife in, for instance, the United Kingdom (Beresford et al., 2008a).

For some organism types, conclusions as to if there is any impact of radiation or not, contrast between studies. For instance, Deryabina et al. (2015) report no effect of radiation on the abundance of a range of medium-large mammals (2008–2010) in the Belarusian portion of the CEZ. The abundance of ungulate species being similar to that of

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untampered nature reserves in Belarus; numbers of wolves were higher in the CEZ than in Belarussian nature reserves (Deryabina et al., 2015). Based on a study using motion activated wildlife trap cameras in 2014, Webster et al. (2016) found no evidence of an influence of radiation on the distribution of medium-large carnivores or wild boar (*Sus scrofa*). Similarly, in earlier studies (1994–1995), Baker et al. (1996) found no impact of radiation on the diversity and abundance of small mammal species. Conversely, based on a snow tract study conducted in 2009, Møller and Mousseau (2013) report a negative effect of radiation on the abundance of mammals and also an impact on predator-prey relationships.

Similar contrasting results have been reported for the influence of radiation on soil biological activity. Both Bonzom et al. (2016) and Mousseau et al. (2014) studied leaf litter decomposition over a range of contamination gradients within the CEZ. Whilst Bonzom et al. report no detrimental effect of radiation on organic matter decay (2010–2011), Mousseau et al. report a ‘severely depressed’ decomposition at more contaminated sites (2007–2008) with a consequent increase in forest floor thickness.

In this paper, we explore potential reasons that contribute to the lack of consensus in published studies on the effect of radiation on wildlife in the CEZ and put these into context with recommendations made by an international workshop, which are considered in this issue (Barnett and Welch, 2016).

### 3. Estimating radiation exposure in the field

Radiation exposure in field effect studies has often been poorly determined (Beresford et al., 2012; Garnier-Laplace et al., 2013, 2015; Beaugelin-Seiller et al., 2019). Studies that report exposure (or dose rates) as opposed to soil activity concentrations largely only use

handheld dose rate meters. Where the measurement height is given (this information is often lacking), then it is typically at, or close to (5 cm), ground level (e.g. Møller and Mousseau, 2013; Lehmann et al., 2016). Some studies report the dose meter readings in units, which are not applicable to wildlife (i.e. sievert (Sv)). While part of the International System of Units (<http://www.bipm.org/en/about-us/>), the Sv is the unit of effective dose that accounts for the biological effect of radiation on humans and as such human derived values of radiation weighting factors are used in its calculation (ICRP, 2007). That said, when considering external dose from <sup>137</sup>Cs, the main contributor in the CEZ, the relative values in Sv and gray (Gy) are likely to only differ by about 15% (Wood and Copplestone, 2011).

Using handheld dose meters is likely generally acceptable as a marker of differences in contamination levels between study sites. However, their use to ascribe dose rates to measurable radiation effects has limitations as they only provide an indication of the external dose rate and neglect the contribution from radionuclides internal to the organism’s body nor do they account for differences in external dose as a consequence of occupancy (e.g. does an animal live underground, in tree, fly etc.). Furthermore, the <sup>137</sup>Cs:<sup>90</sup>Sr ratio is not consistent across the CEZ (Kashparov et al., 2003) and this will consequently impact on a relationship between results of hand held dose rate meters and the actual absorbed dose organisms receive.

The estimation of dose in the CEZ needs to account for the highly heterogeneous nature of contamination in the CEZ (Kashparov et al., 2018; Beresford et al., 2008b). There also needs to be consideration of home range size of the organisms of interest (Smith et al., 2015) and how life stage may affect exposure (e.g. Tagami et al., 2018).

As Fig. 1 shows, on a large scale the Cs-137 deposition throughout the CEZ varies spatially because of the weather patterns that occurred

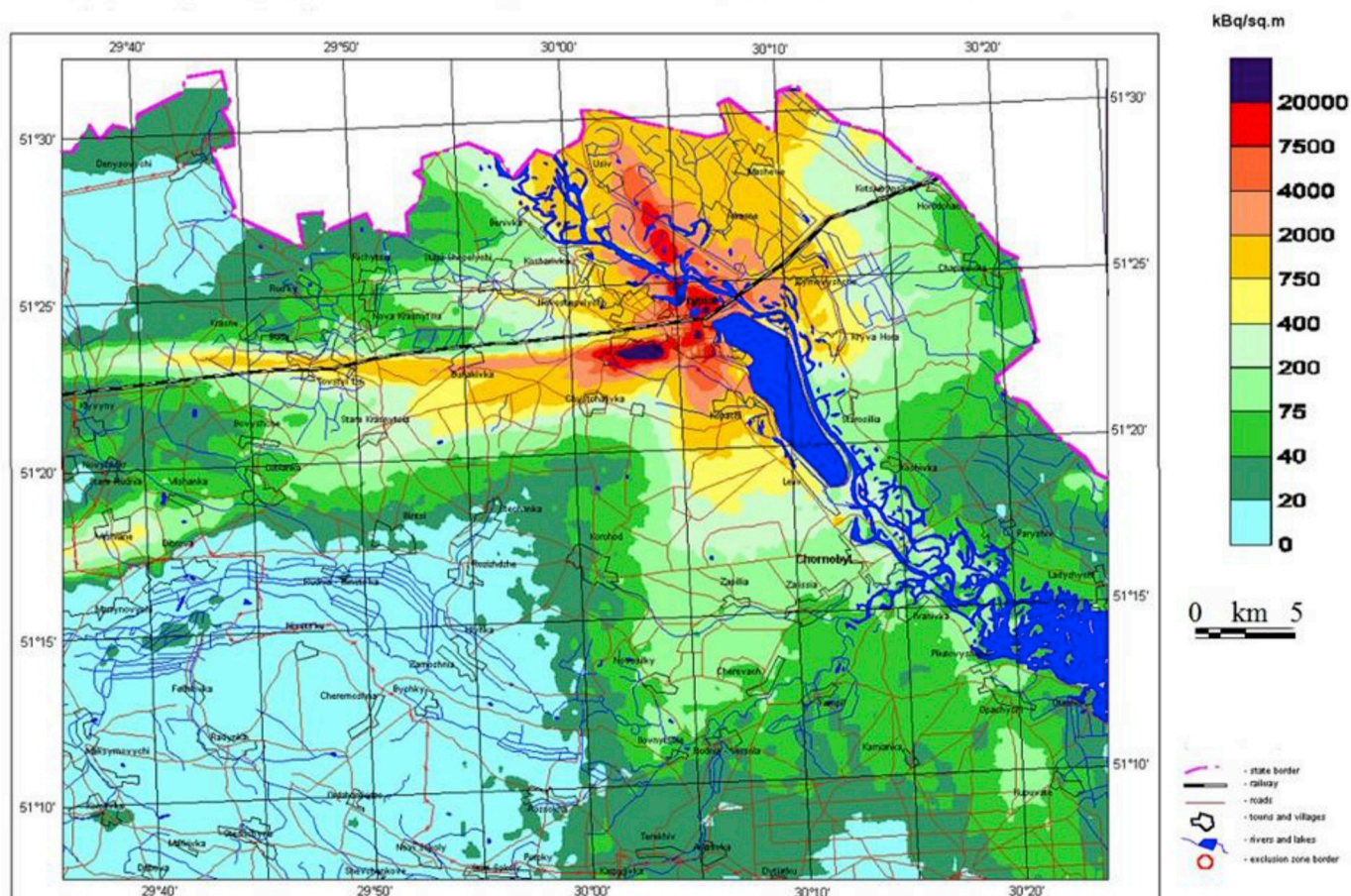


Fig. 1. Spatial pattern of <sup>90</sup>Sr contamination (kBq m<sup>-2</sup>) estimated for 1997 (UIAR, 1998).

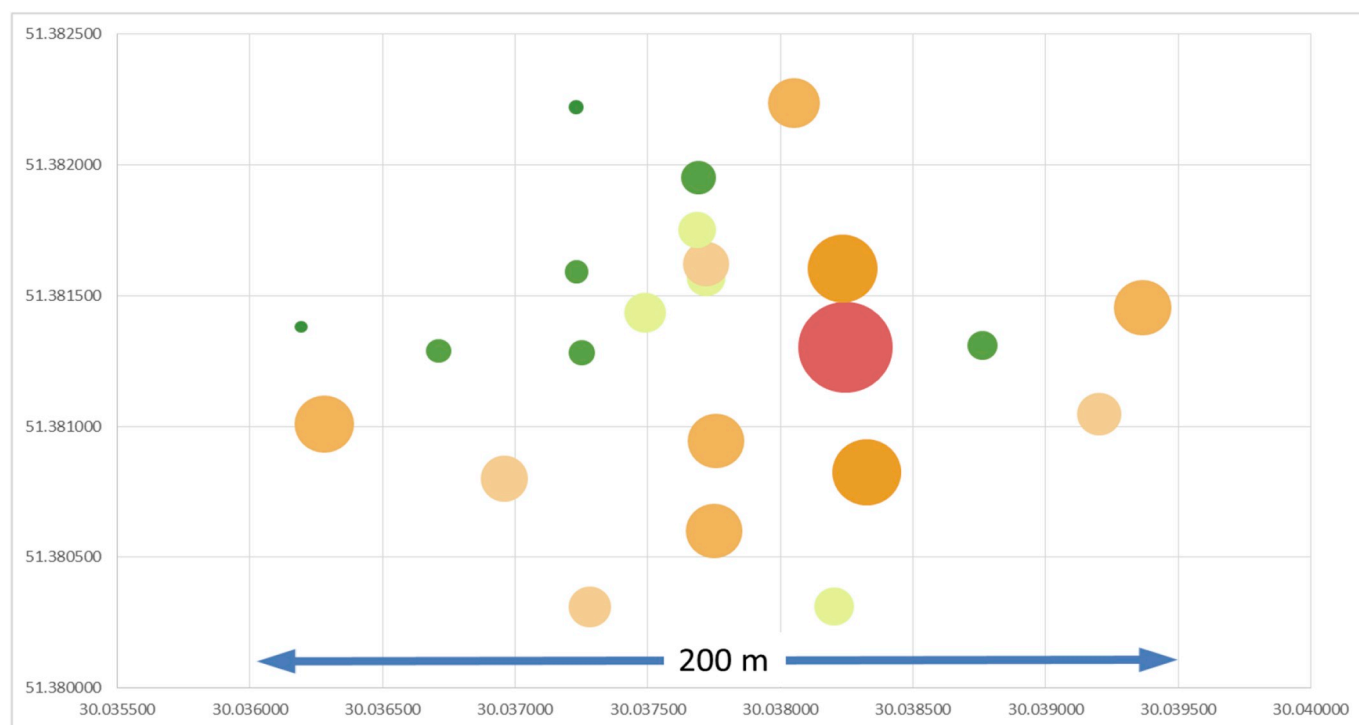


Fig. 2. Spatial variation in soil activity concentration ( $\text{kBq kg}^{-1}$  dry mass) within a Chernobyl exclusion zone field site used to estimate exposure of small mammals using TLDs (Beresford et al., 2008b; Gaschak et al., 2018). Size of circle signifies  $^{137}\text{Cs}$  activity concentration, which ranges from 28 to  $210 \text{ kBq kg}^{-1}$ .

over the 10 days the main releases from the accident occurred in 1986 (UIAR, 1998). Soil activity concentrations, and hence dose rates, can be highly variable on a localised scale as shown in Fig. 2, which shows how the levels vary over a  $200 \text{ m} \times 200 \text{ m}$  field site (Beresford et al., 2008b; Gaschak et al., 2018). Despite these issues, there are papers reporting effects in the literature which have taken just 2–3 measurements at different (random) positions over e.g. 100 m transects and then relate these readings to observations reportedly indicative of radiation induced effects. Such an approach is insufficient to even determine where a ‘site’ sits on the radiation gradient with any degree of confidence (e.g. an external (or ambient) dose rate estimated on the basis of individual sampling points in Fig. 2 could vary by an order of magnitude). Furthermore, to our knowledge, no papers in the literature report the variation observed in the external dose rates (i.e. we have never seen a dose relationship reported for studies in the Chernobyl zone with uncertainty on the x-axis) or take this into consideration in the statistical evaluation (see section 5).

Home ranges can be defined as the areas occupied by individuals for the majority of their time (Minta, 1992; USEPA, 1993; Waser, 1987). However, few published studies consider the importance of home range size. Home ranges vary by organism, for example, small mammals such as the bank vole -  $(4\text{--}7) \times 10^{-4} \text{ km}^2$  (Lindblom, 2008), roe deer -  $0.6\text{--}10 \text{ km}^2$  (Maillard et al., 2002; Mysterud, 1999; Zejda and Bauerova, 1985; Guillet et al., 1996), wolves -  $100\text{--}230 \text{ km}^2$  (Maillard et al., 2002; Tannenbaum et al., 2013). Home range sizes can also vary depending upon the type of ecosystem in which the organism resides. For example, small mammals have larger home ranges in habitats with poor food availability per unit area compared with habitats with good food availability (Akbar and Gorman, 1993). Furthermore, some organisms might only spend a limited fraction of their time in the contaminated areas of interest.

### 3.1. Improving understanding of dose rate and uncertainty in estimation

The use of handheld dosimeters does not give a direct measure of dose received by study organisms as discussed above. However, for a

study of small mammals within the CEZ conducted in 2005, Beresford et al. (2008b) reported that dose rates recorded by a hand-held dosimeter (5 cm above the soil surface) were generally similar to dose rates estimated from thermoluminescent dosimeters (TLDs) attached to the study animals. Similarly, Chesser et al. (2000) report that external dose rates estimated from TLDs attached to voles (*Microtus oeconomus*) were in close agreement with estimates using a hand-held dose meter at ground level (study conducted in mid-1990's). However, Chesser et al. suggested no relationship between external and internal dose rates. The external dose was demonstrated to be dominated by  $^{137}\text{Cs}$  (contributing  $\geq 99\%$ ) at all three study sites used by Beresford et al. (2008b). Chesser et al. also report that the external dose rate to small mammals was higher than the internal dose rate suggesting the total internal dose rate from  $^{90}\text{Sr}$  and  $^{134,137}\text{Cs}$  to voles at a Red Forest site was about 43% of the external dose rate (or approximately 30% of the total dose) in the mid-1990's.

Beresford et al., 2019 report the sampling of a range of species from a site at the western edge of the Red Forest conducted in 2014. Using measured soil and biota concentrations dose rates were predicted for organisms at this site using the ERICA Tool (Brown et al., 2016). The contributions of internal dose to the total dose rate ranged from approximately 6% (bee species) to over 90% (*Pinus sylvestris*, Scots pine) (Fig. 3).

The CEZ has a range of radionuclides present and to conduct an accurate dose assessment there is a need to consider all of these; where actual radionuclides are considered, as opposed to the more commonly reported ambient dose rates, there tends to be a focus on radiocaesium and  $^{90}\text{Sr}$  (e.g. Chesser et al., 2000; Deryabina et al., 2015). Fig. 4 compares the contributions of  $^{137}\text{Cs}$ ,  $^{90}\text{Sr}$ ,  $^{241}\text{Am}$  and Pu-isotopes to the estimated whole-organism internal dose rate of species sampled by Beresford et al., 2019. For most organisms,  $^{90}\text{Sr}$  is the major contributor to internal dose. However, for some organisms  $^{137}\text{Cs}$  contributes a similar or larger proportion of the internal dose. The contribution of  $^{241}\text{Am}$  and Pu-isotopes is typically  $< 10\%$  of the total dose rate (assuming a radiation weighting factor for  $\alpha$ -emissions of 10; the ERICA Tool default value (Brown et al., 2008)). However, in the case of the

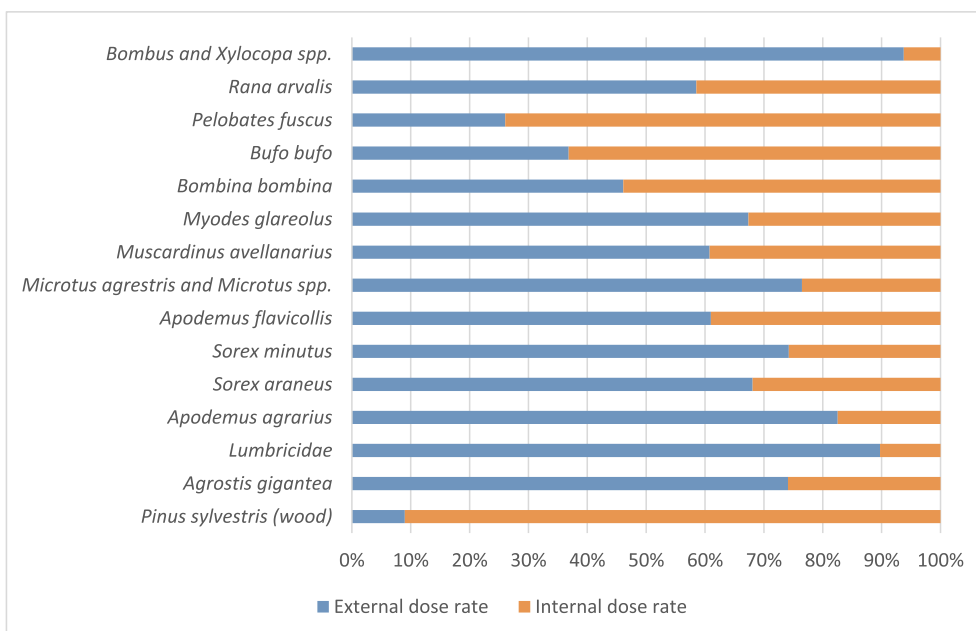


Fig. 3. Contributions of internal and external exposure estimated for different organisms at a site towards the western edge of the Red Forest in 2014 (from Beresford et al., 2019).

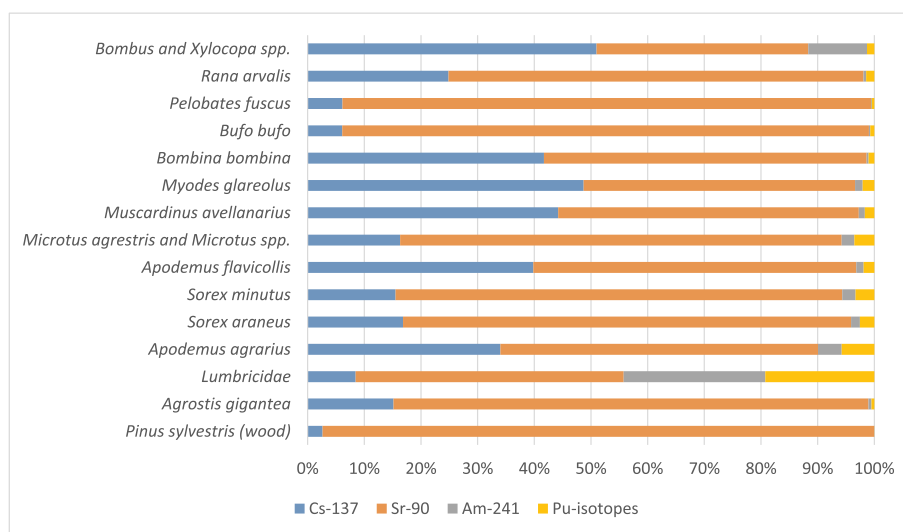


Fig. 4. Contribution of different radionuclides to the internal dose rate estimated for different organisms at a site towards the western edge of the Red Forest in 2014 (from Beresford et al., 2019).

earthworm (Lumbricidae) these  $\alpha$ -emitting radionuclides are estimated to contribute > 40% of the total internal dose rate; there is the possibility that this is due to residual soil in the gastrointestinal tract after depuration which was used in the study. Beresford et al. (2016),<sup>1</sup> report activity concentrations of  $^{90}\text{Sr}$ ,  $^{137}\text{Cs}$  and Pu-isotopes in songbird, bat and mammal species at a site in the CEZ from sampling conducted 2005–2008. These data have been used here to estimate dose rates using the ERICA Tool (with the default  $\alpha$ -radiation weighting factor). For bat species  $^{90}\text{Sr}$  was estimated to be the largest contributor to internal dose rate (97%) with  $^{137}\text{Cs}$  contributing about 3%; the

contribution of Pu-isotopes was < 0.01%. For small ground dwelling mammals  $^{137}\text{Cs}$  and  $^{90}\text{Sr}$  were estimated to contribute about 23% and 77% respectively with a contribution of approximately 0.04% from Pu-isotopes. Strontium-90 was estimated to contribute about 81%,  $^{137}\text{Cs}$  19% and Pu-isotopes 0.11% for birds.

In interpreting field effects studies, authors should endeavour to make the most robust dose assessment possible (see also discussion in Beaugelin-Seiller et al., 2019). To achieve this, in addition to the adequate estimation of soil activity concentrations (see above), the ideal approach would be to determine whole-organism activity concentrations and also estimate external dose rates directly for study organisms. Within the CEZ it has been possible to determine  $^{90}\text{Sr}$  and  $^{137}\text{Cs}$  activity concentrations in live-organisms (Bondarkov et al., 2011) if it is not possible or desirable to sacrifice individuals. The data discussed above show, for vertebrate species and plants at least, that these two radionuclides are likely to dominate the internal dose for many organism

<sup>1</sup> In conducting these analyses it was realised that Tables 2 and 3 in Beresford et al. (2016) have an error in their legends – the units are  $\text{Bq kg}^{-1}$  and not  $\text{kBq kg}^{-1}$  as stated, an Erratum has been published (Beresford & Wood, this issue); Gaschak et al. (2018) presents the full dataset from this study.



a) Early 1990's in the Red Forest showing no living pine trees but deciduous trees continuing to grow (photograph by Nikolay Kuchma)



b) Red Forest in spring 2016 showing establishment of deciduous trees (courtesy: <http://tree.ceh.ac.uk/>)

**Plate 1.** The Red Forest from the early 1990's through to September 2016.

types. Hence, it is likely that a reasonable estimate of internal dose would be obtained from  $^{137}\text{Cs}$  and  $^{90}\text{Sr}$  determinations if it is not possible to destructively sample organisms and analyse organisms for actinide elements. There are a number of models now available, which can be used to estimate dose rates to wildlife (e.g. see Beresford et al. (2010) which discusses the application of a number of such models to data from the CEZ). By preference, the input to these models should be measured activity concentrations in biota. Where this is not possible site-specific transfer parameters should be used to estimate organism activity concentrations rather than relying on default model values. Transfer parameter values are available for a number of terrestrial species within the CEZ, which could be used to improve future assessments, though these tend to be biased towards vertebrate species (e.g. Barnett et al., 2009; Beresford et al., 2008b, 2016, 2019; Gaschak et al., 2003, 2010; 2018; Ryabokon et al., 2005). However, there is some indication of a reduction in the transfer of  $^{90}\text{Sr}$  with increasing soil

contamination possibly as a consequence of  $^{90}\text{Sr}$  being predominantly in the form of fuel particles at more contaminated sites (see discussion in Beresford et al., 2016).

For many organism types it is likely that estimates of individual external dose could be obtained by the use of dosimeters attached to them (e.g. TLDs) (see review by Aramrun et al. (2018)). Organisms as small as bees could be fitted with TLDs, though the use of dosimeters would require sufficient rates of recapture (to remove and subsequently analyse the dosimeters) and achieving this may be resource intensive. Beresford et al. (2008b) showed that with sufficient soil sampling to represent the home range of the study species reasonable estimates of external dose could be achieved via the use of a model to estimate external dose; though determining the actual areas utilised may be difficult for some species. If using one of the available wildlife assessment models to estimate external dose rates in the CEZ then users need to be aware that some of the models include a skin/fur/feather



Plate 2. View to the Chernobyl number 4 reactor from over the Red Forest (courtesy: N. Entwistle, University of Salford; <https://www.ceh.ac.uk/redfire>).

shielding factor (e.g. the ERICA Tool (Brown et al., 2008)) whilst others do not (see Vives i Batlle et al. (2007, 2011) for discussion). The selection of model would therefore have some impact on the external dose rate given the relatively high activity concentrations of  $^{90}\text{Sr}$  in soils in the CEZ. However, it is likely that  $^{137}\text{Cs}$  would still dominate the estimated external dose rate (though the external dose rate estimate for  $^{90}\text{Sr}$  could be 5 or 6 orders of magnitude different depending on the model used (Vives i Batlle et al., 2007).

With an adequate number of measurements, the common application of using handheld meters to estimate dose rate should give an approximation of the contamination gradient between sampling sites. However, they do not give a robust estimate of total dose received by an organism and this limitation should be acknowledged in future publications. That said on the basis of evidence from the CEZ handheld dose rate meters may give a reasonable approximation of external dose rate (Chesser et al., 2000; Beresford et al., 2008b). However, external dose rate may not be correlated with internal dose rate, though external dose may dominate the total dose rate in some instances.

The need for as robust as possible dose assessments is well demonstrated by Garnier-Laplace et al. (2015) who conducted a dose reconstruction of bird census data for Fukushima contaminated areas of Japan. The original work by Møller et al. (2012, 2015a,b) reported ambient dose rates measured by hand-held dosimeter. Garnier-Laplace et al. estimated dose rates typically about a factor of three higher than the ambient dose rates reported in the initial publications with the result that the trends in census data were somewhat reconciled against existing knowledge of radiation effects.

#### 4. Site and exposure history

In some studies, the reported relationships between radiation exposure and the effect measure of interest, appear to be driven by points of undue influence (i.e. sites with especially high radiation levels). For instance, Bonzom et al. (2016) suggest that two data points in the study of Mousseau et al. (2014) appear to drive the negative relationship between mass loss from leaf litter and radiation level. A good example of a potential effect of points of undue influence can be seen in Fig. 2 of Møller and Mousseau (2013) where at least three data points are visible at dose rates of around  $250 \mu\text{Sv h}^{-1}$  and these appear to have a strong influence on the significant relationship reproduced in this figure. These key data points were most likely from the Red Forest given the

field sites marked on Fig. 1 of the Møller & Mousseau paper; the Red Forest is also the most likely place to find dose rates of the magnitude cited ( $> 200 \mu\text{Sv h}^{-1}$ ).

Coniferous trees in the Red Forest were killed in 1986 and the area subsequently regenerated with deciduous species and understorey vegetation. Plate 1 shows photographs of the ecosystem at different times to illustrate how the site has changed from coniferous forest (primarily *Pinus sylvestris* L.) to birch (*Betula pendula* Roth.), black alder (*Alnus glutinosa* L.) and other understorey vegetation. Gaschak (2016) notes the poor habitat quality of the Red Forest compared to other areas of the CEZ. At this site in particular, with the death of coniferous species, in part because of their greater radiosensitivity, the ecosystem has changed because of radiation exposure both directly and indirectly. For example, indirect effects have occurred on the light and nutrient levels because of the loss of canopy cover or changes in the leaf litter composition from acidic pine needles to deciduous leaf litter. These changed environmental conditions allowed more radioresistant deciduous species to develop. With a change from coniferous to deciduous woodland cover, there was also disturbance in the ecological communities present, with changes in the diversity of species associated with the different woodland types (Geras'kin et al., 2008). Consequently, the major disturbance caused by the accident and the subsequent ecological changes in species composition and diversity, have had a major influence on how the site has changed over time. These complex changes in site history need to be considered as potential confounding factors when trying to understand the current relationships between organisms inhabiting contaminated sites with differing radiation levels.

As shown in Plate 2, the Red Forest is also near the former nuclear power plant infrastructure where there is still a relatively high degree of human activity. Disturbance by humans may have some influence on observations of some wildlife species in the Red Forest.

Over the  $> 25$  years we have been working in the CEZ we have observed that the area is generally 'rewilding' with former agricultural land becoming scrub and woodland and urban areas being reclaimed by nature. The changes in the Red Forest are more extreme, but, accounting for (or even acknowledging) site evolution/history in published studies is generally lacking.

The CEZ experienced very much higher dose rates in the past. In 1986 dose rates were sufficient to result in, for instance: suppression of tree growth (over at least 11,900 ha), reduced seed numbers in herbaceous plants, reduction in herbaceous plant diversity, order of

magnitude reductions in soil invertebrate densities (3–7 km from the Chernobyl plant) and reductions in small rodent like mammal numbers (Geras'kin et al., 2008). The higher doses in the past have potential consequences (defined here as 'memory effect') for current observations (i.e. are relationships between biological/ecological parameters and radiation the effect of current or past doses?). Memory effect reflects the impact of factors such as adaptation, high mutation load, death and immigration. For example, Geras'kin et al. (2008) showed that the severity of radiation effects on species/ecosystems in the CEZ were strongly dependent on the dose received in the early period after the accident.

## 5. Lies, damned lies, and statistics?

Field sampling strategies and the statistical analysis of data from studies conducted within the CEZ have been previously commented upon (Beresford et al., 2012; Garnier-Laplace et al., 2013). The design of the field study and the approach used to analyse data are key in ensuring that the results and findings are reliable and repeatable. Recognising this importance, in this section we discuss some general statistical issues relevant to studies conducted in the CEZ; we do this in a non-judgemental manner and do not make reference to any particular published studies from the CEZ and also issue guidance on what is good practice.

### 5.1. What is statistically significant and how should we interpret it?

The nature of scientific enquiry typically starts with a scientific question of interest, followed by examination of evidence (which we will return to) and then an overall statement of the weight of evidence with regard to that scientific question. Statistics and statistical models are an important component of scientific enquiry. In the radiation context, a typical question of interest might be expressed as "are mammal abundances (or any other biologically relevant response) negatively correlated with levels of radioactive contamination in the CEZ?" It is important to define the scientific question as clearly and as unambiguously as possible. The wording of our question here suggests an hypothesis *a-priori* about how radiation affects abundance (it postulates a negative correlation), a more neutral form might be to ask "how does radioactive contamination affect abundance across the CEZ?"

To answer such questions requires clear definition of a relevant measure (e.g. reproductive status) of biota and biodiversity (e.g. species richness, or abundance) and a measure of radioactive contamination or dose (whether nuclide specific or more general) in the region, the extent of the region needs to be very carefully defined since some species of interest might have large home ranges or there may be temporal issues to take into account. It is important therefore to state your question and define the variables you are going to measure and what their characteristics are in line with many international guidelines (e.g. ICRU, 2006).

The next step is to observe and measure, by selecting an appropriate sampling design and implementing it within the region of interest. The general goal in any experiment or survey is to account for natural variability by, where possible, controlling extraneous factors, and where not possible to measure those extraneous factors as covariates. For field work, the design step will typically involve carrying out a survey for our question of interest, involving potentially: a) capturing and measuring animals (using a power analysis would determine the optimal sample size to enable a hypothesis to be tested to the desired level of significance); and b) mapping the radiation environment (through soil sampling, in-situ monitoring etc.). As it is not possible to measure every animal nor sample/monitor everywhere, we generally study a sample of the animal population. The sample typically comprises a small percentage of the population and by using statistical sampling methods (random, systematic or stratified) we hope to avoid

any biases (conscious or unconscious). To measure the radioactivity in the study area, we might generate maps (spatially continuous) based on a monitoring campaign giving either point measures at specific locations (geostatistical data) or from areal data from in-situ monitoring or aerial survey. To answer our question of interest we then need to link these two data sources, often through regression equations but bearing in mind that we may be further challenged as many species of interest are not static and will roam through the region.

In many scientific fields, it is possible to design experiments, controlling factors that determine some (hopefully a large percentage) of the variation in the response. Experiments help us answer questions or test hypotheses, and we can design them to minimise bias, to keep errors in comparison small. This means we can keep confounding factors under control, allowing us to make stronger inferences. Our other alternatives are observational studies (or natural experiments). In the Chernobyl case, we are not able to fully design an experiment, and we can only observe (in a natural sense), we cannot control all the environmental or ecological factors, rather we measure them and consider them as covariates to be incorporated in our formal statistical analysis. However, factors such as, the type of observation, the size of the sample and measurement locations, all need to be considered before we begin to think about any formal statistical analysis.

### 5.2. Why do we need statistical analysis?

We are familiar with the view "if you need to use statistics, then you have not designed your experiment very well". This ignores the fundamental facts of life; we need statistics because we need to recognise and handle variation. Variation, arises because of the fundamental uncertainty in our measurements, in our sampling and from simply recognising that if we were able to repeat the sampling and monitoring we would get different results. And because we want to make inferences and predictions (i.e. not simply about the sample of animals we have observed but about the population of animals (most of whom we have not observed)).

### 5.3. Reporting results of statistical analysis

The reporting of the results of a statistical analysis may be: a confidence interval (a plausible range of values for an unknown quantity with a given statistical confidence, typically 95%); a p-value from a hypothesis test, which informs about whether a finding is statistically significant, (e.g. the p-value is less than 0.05 and so result is statistically significant); or an equation from a model such as a regression equation with parameter estimates (e.g. the intercept and slope of a fitted line).

Statistical significance is concerned with the ability to discriminate between treatments given the background variation. However, statistical significance does not equate always to 'practical importance'. For example, we might observe a small, but still statistically significant difference between the average number of fledglings that survive in two nests in different radiation zones, but such a small difference may not be relevant to species survival. Furthermore, statistically significant relationships may explain little observed variation (i.e. they have a poor  $R^2$  value).

There is one other component of our statistical analysis which we need to consider, namely power. Power is the probability that we correctly conclude that the null hypothesis should be rejected (i.e. we are more likely to see an effect if there is one there to be detected), where the null would say there is no difference/no effect/no trend. We want a high power, where power is a function amongst other things of sample size; a high power makes it less likely we will say there is not an effect when there may be one. The consequences of low power include overestimates of effect size and low reproducibility of results (Button et al., 2013). It is uncommon to see power calculations quoted in the methods sections of published papers.

#### 5.4. Other relevant and common statistical issues

##### 5.4.1. Generalised linear models and random effects

As we consider our regression model, we need to be conscious of the assumptions being made. We often assume that the response variable is normally distributed, but this may not be the case, e.g. if the response is a count (or has a Poisson distribution) such as the number of wolves observed, then we need to use generalised linear models.

The basic regression models also typically assume that observations are independent, but, we often have dependence which must be dealt with. Dealing with dependence can be a challenge; one of the most common situations in many studies that induces dependence is that of repeated measures (i.e. the same individual is observed over time, perhaps not an issue in most field studies in the CEZ). Another common situation might be spatial (e.g. individual animals all living in the same forest or nest), the responses of different individuals in the same unit, share common sources of variation and are therefore not statistically independent. Statistical models with dependent observations are commonplace, and one of the commonly used forms of models to address this is known as a *mixed* model. A mixed model introduces one or more random effects, which are used to capture the common source(s) of variation. Random effects also encompass variation among individuals (for example, when multiple responses are measured per individual), genotypes, species and regions or time periods (Bolker et al., 2009).

##### 5.4.2. Correlation, association and causation

The existence of a statistically significant correlation (or association) between two variables (e.g. one increases as the other increases) does not necessarily imply causation (i.e. the increase in one variable causes the increase in the other); there may be other unobserved variables which are involved in the 'observed' relationship. Correlation also only assesses linear relationships; there may be real non-linear relationships, which the standard correlation coefficient does not capture.

##### 5.4.3. Goodness of fit and unusual observations

An important aspect of statistical model building is to challenge the results, by checking the validity of assumptions and the robustness of the findings. If the inferences are strongly influenced by only a few unusual observations, then beware. The existence of outliers (i.e. unusual observations) can often be best detected visually in a scatterplot or boxplot, and there are simple numerical rules that flag such observations, but there then needs to be a scientific decision about what should be done with the outliers. Simple deletion is never the right answer. In many situations, it is worth examining the robustness of any identified relationship by fitting a model with and without such observations.

## 6. Conclusions

The Chernobyl accident offered a unique opportunity to study the effect of ionising radiation on ecosystems. Effects on a range of wildlife in the first years after the accident were recorded by scientists from the former Soviet Union countries and these were broadly in agreement with what would have been anticipated from existing understanding of the effects of radiation on wildlife.

Dose rates remain sufficiently high in some areas of the CEZ that we would anticipate some effects of radiation on wildlife. However, a number have studies have reported significant and serious effects and related these to dose rates that would appear to be too low to implausibly low. In some instances, results between different workers are conflicting. In others reported effects are verified by photographic evidence.

Potential reasons for some of the apparent conflict in results and interpretation of studies conducted in the CEZ are discussed above. We should also accept that studies conducted within the CEZ have a degree

of compromise associated with them – facilities/equipment may be limited, access hours restricted and scientists may only be able to be there for relatively short periods of time. However, when writing-up such studies, authors should be clear about the limitations of their work.

Correct interpretation of statistical results play a key role in ensuring that conclusions are sound and that uncertainty surrounding them is represented properly (Wasserstein and Lazar, 2016). The reliability and reproducibility of scientific findings are topics, which are being increasingly debated in many scientific domains, and part of these debates are framed round the use and miss-use of statistics. A number of international organisations have issued statements concerning good statistical practice, for example: “*Researchers should disclose the number of hypotheses explored during the study, all data collection decisions, all statistical analyses conducted and all p-values computed. Valid scientific conclusions based on p-values and related statistics cannot be drawn without at least knowing how many and which analyses were conducted, and how those analyses (including p-values) were selected for reporting.*” (Wasserstein and Lazar, 2016). It is also important that power of the studies be reported.

If the underpinning data from the field studies were made openly available a significant step would be made to addressing the disagreement on the magnitude of effects due to exposure to ionising radiation observed in the CEZ (and Fukushima areas) by enabling data re-evaluation by others. Recently, datasets from the CEZ have been made available for: radionuclide activity concentrations in and transfer to a range of terrestrial vertebrates (Gaschak et al., 2018); radionuclide deposition in the CEZ and particle studies (Kashparov et al., 2017, 2018); a ‘reference site’ in the CEZ (Beresford et al., 2018). All of these could be used to improve future dose assessments.

However, improved dose assessments are only likely to change reported dose rates by up to approximately an order of magnitude (Beaugelin-Seiller et al., 2019; Beresford et al., 2019). This would not necessarily resolve the controversy between observed field effects in the CEZ and the currently recommended benchmarks below which there should be negligible effect on wildlife (ICRP, 2008; Andersson et al., 2009). To better understand the potential impact of radiation in the CEZ we need to better acknowledge the potential for ‘memory effect’ as we have discussed here and design studies which investigate it. Future studies should ensure that they have appropriate controls (this has been lacking in some reported studies); the long-term study of control sites/organisms would also give information on temporal variation which could be useful in interpreting results from studies in the CEZ.

UNSCEAR (2015) discuss “*attributability of health effects*” and “*inference of risk*” in the context of human health and ionising radiation. In summary, they recommend that observations of potential radiation induced effects should be presented together with the underlying assumptions and analysis of uncertainties. In our opinion, similar recommendations are applicable to wildlife effect studies.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.jenvrad.2019.01.005>.



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