



Soil nematode assemblages as bioindicators of radiation impact in the Chernobyl Exclusion Zone



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HIGHLIGHTS

- First characterisation of nematode assemblages in the Chernobyl Exclusion Zone
- An accurate estimation of total dose rate absorbed by nematodes was conducted.
- The total dose rate to nematodes mainly depends on external alpha and beta radiation.
- Nematodes have low sensitivity to chronic exposure to radioactive contamination.

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ABSTRACT

In radioecology, the need to understand the long-term ecological effects of radioactive contamination has been emphasised. This requires that the health of field populations is evaluated and linked to an accurate estimate of received radiological dose. The aim of the present study was to assess the effects of current radioactive contamination on nematode assemblages at sites affected by the fallout from the Chernobyl accident. First, we estimated the total dose rates (TDRs) absorbed by nematodes, from measured current soil activity concentrations, Dose Conversion Coefficients (DCCs, calculated using EDEN software) and soil-to-biota concentration ratios (from the ERICA tool database). The impact of current TDRs on nematode assemblages was then evaluated. Nematodes were collected in spring 2011 from 18 forest sites in the Chernobyl Exclusion Zone (CEZ) with external gamma dose rates, measured using radiophotoluminescent dosimeters, varying from 0.2 to 22 $\mu\text{Gy h}^{-1}$. These values were one order of magnitude below the TDRs. A majority of bacterial-, plant-, and fungal-feeding nematodes and very few of the disturbance sensitive families were identified. No statistically significant association was observed between TDR values and nematode total abundance or the Shannon diversity index (H'). The Nematode Channel Ratio (which defines the relative abundance of bacterial- versus fungal-feeding nematodes) decreased significantly with increasing TDR, suggesting that radioactive contamination may influence nematode assemblages either directly or indirectly by modifying their food resources. A greater Maturity Index (MI), usually characterising better soil quality, was associated with higher pH and TDR values. These results suggest that in the CEZ, nematode assemblages from the forest sites were slightly impacted by chronic exposure at a predicted TDR of 200 $\mu\text{Gy h}^{-1}$. This may be imputable to a dominant proportion of pollutant resistant nematodes in all sites. This might result from a selection at the expense of sensitive species after the accident.

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1. Introduction

The Chernobyl nuclear accident occurred on the 26th of April 1986 and led to the release of 9.35×10^3 to 1.24×10^4 PBq into the environment (Yablokov et al., 2009).

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Acute exposure which occurred immediately after this accident led to important damage to wildlife. A sharp decrease in the density of forest soil invertebrates and major effects on their reproductive processes were observed in the close vicinity of the Chernobyl nuclear power plant (Krivolutsky and Pokarzhevsky, 1992; Krivolutsky et al., 1992). One year later, the resident population of invertebrates slowly recovered due to the migration of insects from surrounding areas. Within 2.5 years of the accident, the density of mesofauna in the Chernobyl Exclusion Zone (CEZ) was almost completely restored, although the diversity was lower than before the accident (Krivolutsky and Pokarzhevsky, 1992; Gerask'in et al., 2008).

Twenty-seven years later, the CEZ still exhibits a residual background radiation level significantly affected by the radioactive fallout, and mostly associated with persistent ^{137}Cs , ^{90}Sr and plutonium and americium isotopes. At present, the ambient external gamma dose rates in the zone vary from background ca. $0.1 \mu\text{Gy h}^{-1}$ to ca. $10 \times 10^3 \mu\text{Gy h}^{-1}$ (Garnier-Laplace et al., 2013).

Unfortunately, the long-term ecological consequences of wildlife exposed to ionising radiation at a chronic low dose rate due to the Chernobyl disaster are still unclear, particularly regarding the effects on abundance and species biodiversity of flora and fauna (Hinton et al., 2007; Gerask'in et al., 2008). For example, 17 years after the accident a study by Murphy et al. (2011) revealed that there was no relationship between radiation dose rates, which varied from 0.1 to $30 \mu\text{Gy h}^{-1}$, and the abundance or diversity of aquatic invertebrate communities in Chernobyl-contaminated lakes. Similar findings were observed previously in Hanford sites (U.S.), where radioactive contamination did not appear to be associated with changes in the structure of aquatic communities of invertebrates following chronic exposure ranging from background to more than $22,500 \mu\text{Gy h}^{-1}$ (Emery and McShane, 1980).

In contrast, Jackson et al. (2005) observed a decrease in soil invertebrate diversity (but not abundance) in the CEZ sites with external gamma dose rates ranging from 0.1 to $140 \mu\text{Gy h}^{-1}$. Subsequently, Møller and Mousseau (2009) demonstrated reduced abundance of aboveground invertebrates (bumblebees, butterflies, grasshoppers, dragonflies and spiders) in the CEZ at external gamma dose rates which were one order of magnitude below those published by Jackson et al. (2005).

Several authors justified this lack of consistency on the observed long-term impact of radiation on wildlife by the lack of accurate dose rate estimates, which may have introduced a bias in the comparison of the results (Smith, 2008; Beresford and Coppelstone, 2011; Wickliffe and Baker, 2011; Garnier-Laplace et al., 2013).

Indeed, total dose rate (TDR) absorbed by wildlife is composed of the external dose rate in addition to the internal dose rate due to the incorporation of radionuclides through many pathways according to their mode of life (e.g., ingestion, inhalation, dermal absorption). However, most studies only reported external dose rates and did not consider the contribution of all radiation types (including alpha, beta and gamma emitters). These observations highlight, particularly in the context of the recent Fukushima nuclear power plant accident (Japan), the need to improve the estimate of radiological dose exposure in order to properly evaluate the consequences of radioactive contamination in exposed wildlife.

In the present study, we assessed the ecological impact of long-term exposure to radioactive contamination in the CEZ using nematodes as biological indicators.

Nematodes are abundant and ubiquitous soil invertebrates. As they complete their life cycle in soil with a low rate of dispersal, they are considered to be continuously and homogeneously exposed to soil radioactive contamination. Nematodes are therefore a suitable biological model to obtain an accurate estimate of radiation dose. Furthermore, nematode abundance and diversity are influenced by any change in the quality of their soil habitat or food resource and are often used in the assessment of soil disturbances caused by exposure to pollutants

(Ekschmitt and Korthals, 2006, 2009; Yeates et al., 2003; Zhao and Neher, 2013).

Previous studies described an increase in mortality in nematode populations and an increase in DNA damage in *Caenorhabditis elegans* after acute (several Gy or kGy and short-term exposure) gamma irradiation (Buchan et al., 2012; Graig et al., 2012; McNamara et al., 2003). Unfortunately, the results from these studies cannot be directly extrapolated to field conditions as it was demonstrated previously that acute and chronic exposures to ionising radiation do not lead to the same biological effects (Pereira et al., 2011). Moreover, the sensitivity of organisms may differ from laboratory to field conditions and from single species to complex ecological systems (Garnier-Laplace et al., 2013). Thus, fieldwork to specifically address the toxicity of chronic exposure to irradiation is essential. This study constitutes the first use of nematode assemblages as bioindicators of field radioactive contamination.

We first obtained an accurate estimate of the absorbed dose rate to the nematodes at a range of sites with varying levels of radioactive contamination. This was performed to provide an appropriate description of the exposure, taking into account exposure pathways (internal/external exposure), and all radionuclides and radiation types (alpha, beta and gamma emitters). Secondly, we evaluated the impact of the predicted TDRs on nematode assemblages (abundance, diversity, indices of ecological function). The contribution of the predicted dose rate was examined in relation to those of other abiotic variables (pH, soil humidity, soil texture, organic carbon content) in order to consider the influence of confounding factors.

2. Materials and methods

2.1. Study sites

The sampling campaign was conducted in the Chernobyl Exclusion Zone (CEZ) in Northern Ukraine (temperate continental climate). The exclusion zone corresponds to a $90 \text{ km WE} \times 45 \text{ km SN}$ enclosed and abandoned area, as humans deserted the area around the power plant during the weeks following the accident. It is a plain region with alternating meadows and forests. According to our analysis and sampling capacities, a total of 18 deciduous forest sites (populated mainly by alder, birch, aspen and oak) were chosen for nematode sampling and analysis. The sites were selected on the basis of similar ground vegetation and were dominated by graminaceous species. These sites differed significantly in their level of radioactive contamination. The ambient external dose rate at each site was first assessed using a portable digital survey metre (FH40G, Thermo Scientific). Sites 1 to 18 were selected in order to test a wide range of external radiation levels (dose rates increased from site 1 to site 18). Each sampling site was localised by GPS coordinates (Fig. 1).

2.2. Soil sampling

The sampling campaign was from April 25th to May 8th, 2011. During the campaign, the mean temperature was $12.1 \pm 3.7 \text{ }^\circ\text{C}$ and the sum of precipitation was 72 mm. Soil samples were collected from the top 0–10 cm layer. The surface of the sample plot was approximately 100 m^2 ($10 \text{ m} \times 10 \text{ m}$). After removing the litter layer, ten soil cores ($2.5 \text{ cm diameter} \times 10 \text{ cm in depth}$) per plot were randomly collected and pooled to constitute a composite sample. Visible roots and leaves were removed from the samples. Sub-samples of composite soils were used to analyse soil characteristics (humidity, pH, total organic carbon (C_{org}), particle size distribution (PSD)) and nematode assemblages.

2.3. Extraction and identification of soil nematodes

Nematodes were extracted from 150 to 200 g of fresh soil sub-samples using the Seinhorst elutriation method (Seinhorst, 1962).

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