Ecological effects of ionizing radiation on earthworms

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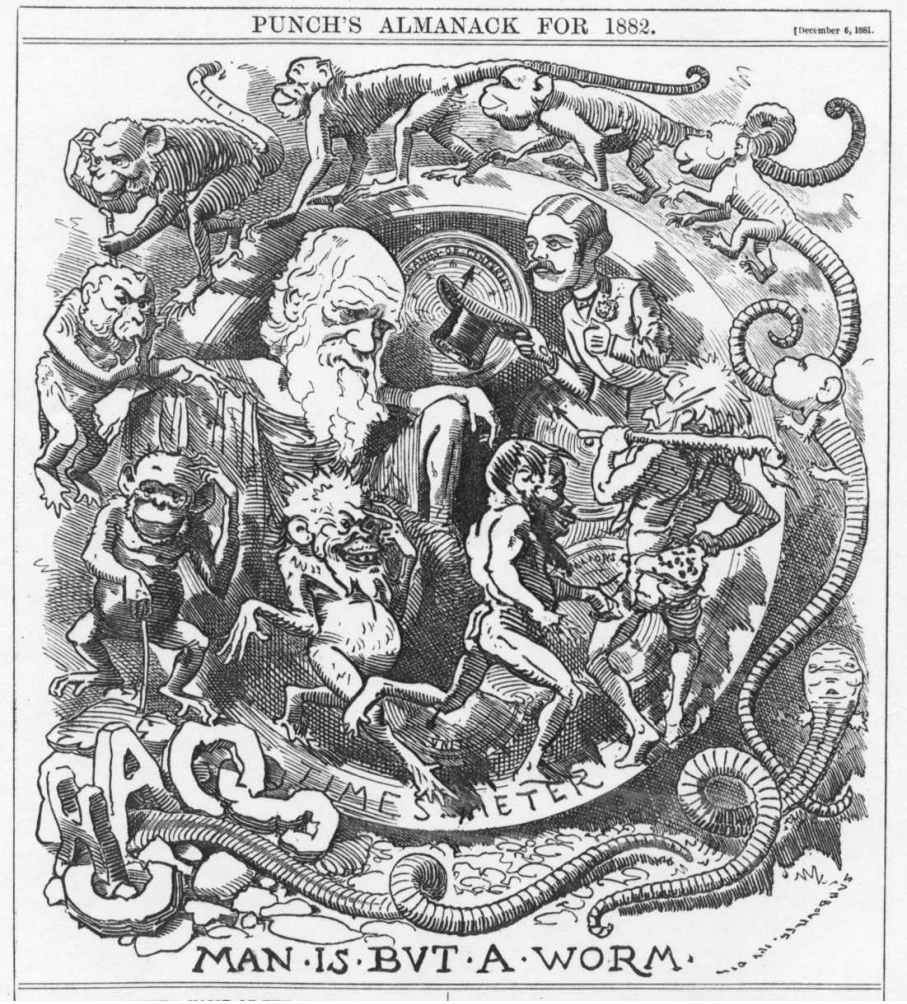
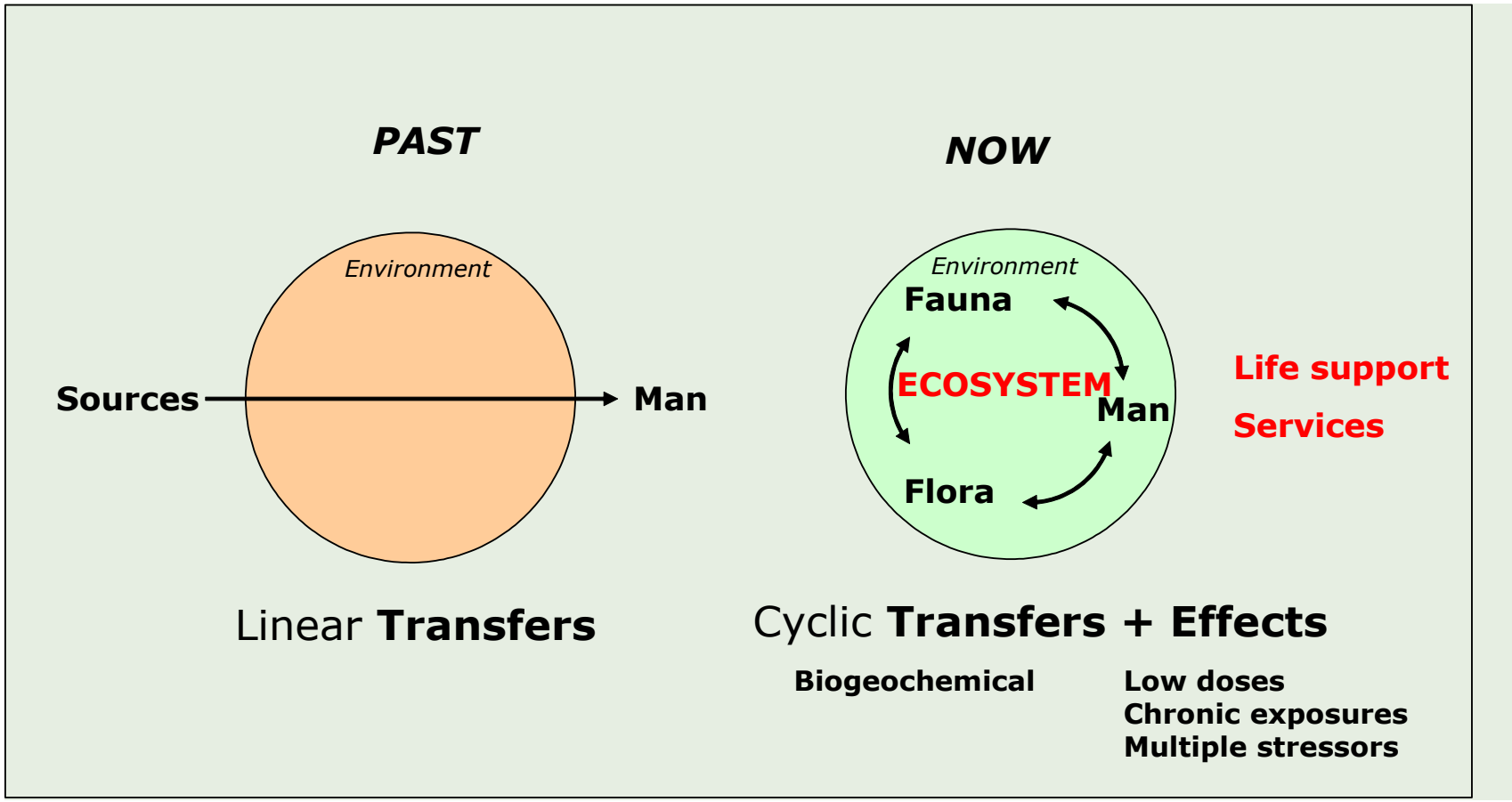
CERAD
CENTRE FOR ENVIRONMENTAL RADIOACTIVITY

Overview

- Earthworms as a reference organism in radioecology
- Field Studies
- Laboratory Studies



Radiation effects on Non-Human Species



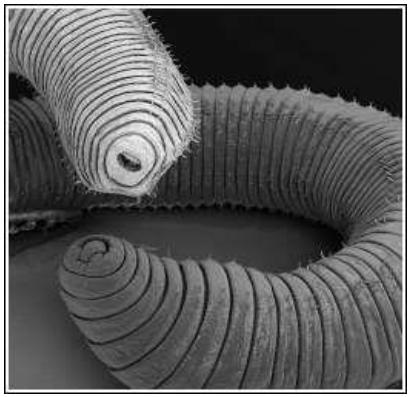
Brechignac, et al. 2012

ICRP « Reference Animals and Plants » - RAPs

- Recognized test species in chemical toxicity studies; standardised tests (OECD)
- Potentially high exposure to radionuclides
- Important role in soil ecosystems

- Deer
- Rat
- Bee
- Earthworm
- Pine tree
- Grass

- Flat fish
- Crab
- Macroalga

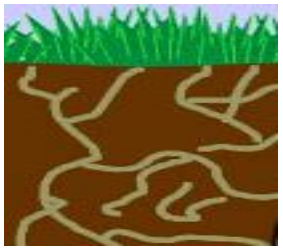


Important for the soil fertility

Eat dead organic material

•**Increases the bioavailability of nutrients for other organisms**

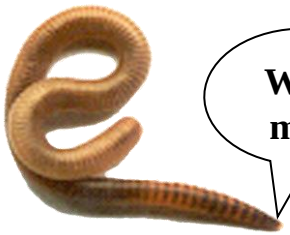
They make burrows in the soil



•**Increase the aeration and water drainage**

•**Mixing organic and inorganic components of the soil**

Important role in the food web



Field Studies

Chernobyl: Effects in Soil Invertebrates

- Populations of soil invertebrates reduced 30-fold, reproduction strongly impacted

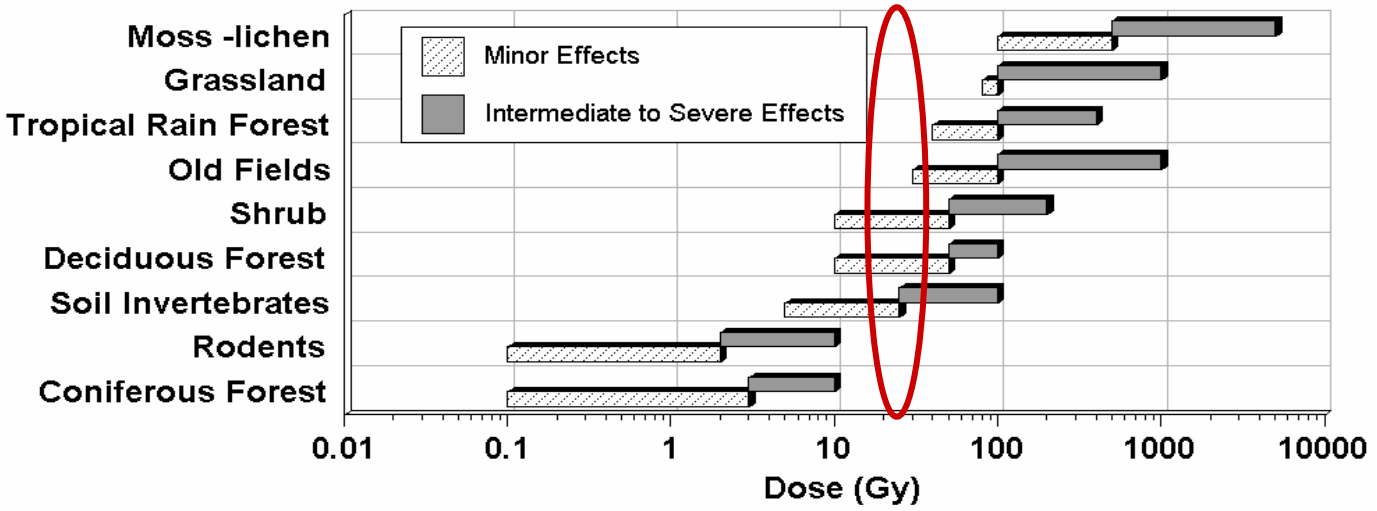
Paper

RADIATION-INDUCED EFFECTS ON PLANTS AND ANIMALS: FINDINGS OF THE UNITED NATIONS CHERNOBYL FORUM

Thomas G. Hinton,* Rudolph Alexakhin,† Mikhail Balonov,‡ Norman Gentner,§ Jolyn Hendry,‡ Boris Prister,** Per Strand,†† and Dennis Woodhead‡‡

Chernobyl: Effects in Soil Invertebrates

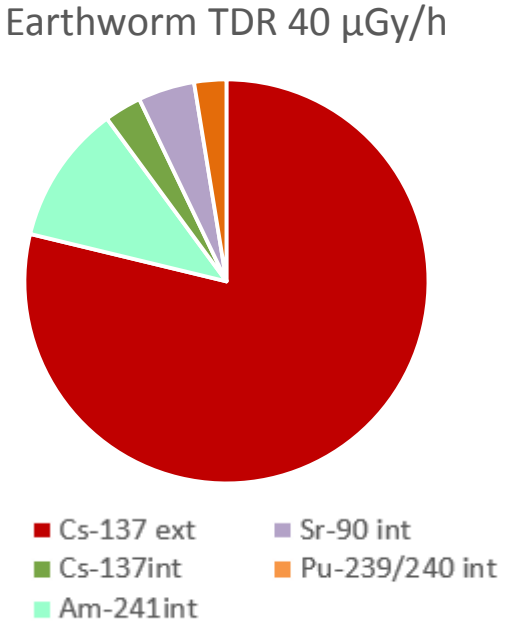
- 30 Gy altered community structure (species diversity) for 2.5 years



Field Studies: Chernobyl

- Field Doses Chernobyl (ERICA)

	Soil Activity Bq/kg	Internal (µGy/hr)	External (µGy/hr)
Cs-137	1,000,000	10	300
Sr-90	500,000	17	7.5E-05
Pu-238+239	10,000	10	1.7E-03
Am-241	10,000	60	1.6E-02



- Potential for higher gut and skin doses?

- Refined dosimety to gut ongoing including particle and microdosimetry for Chernobyl sampled worms
- Ongoing studies on genetic diversity and epigenetic markers

Vodny Site, Komi Republic

- Between 1931 and 1956 the Vodny area in the Komi Republic, Russia, was the main site of Soviet radium production.

Apart from the radium production contamination, largely undisturbed and low human activity for many decades.

Transportation of radium concentrate in wooden barrels, 1940 (Evseeva et al 2000)

Field Study: Contaminated vs Reference sites

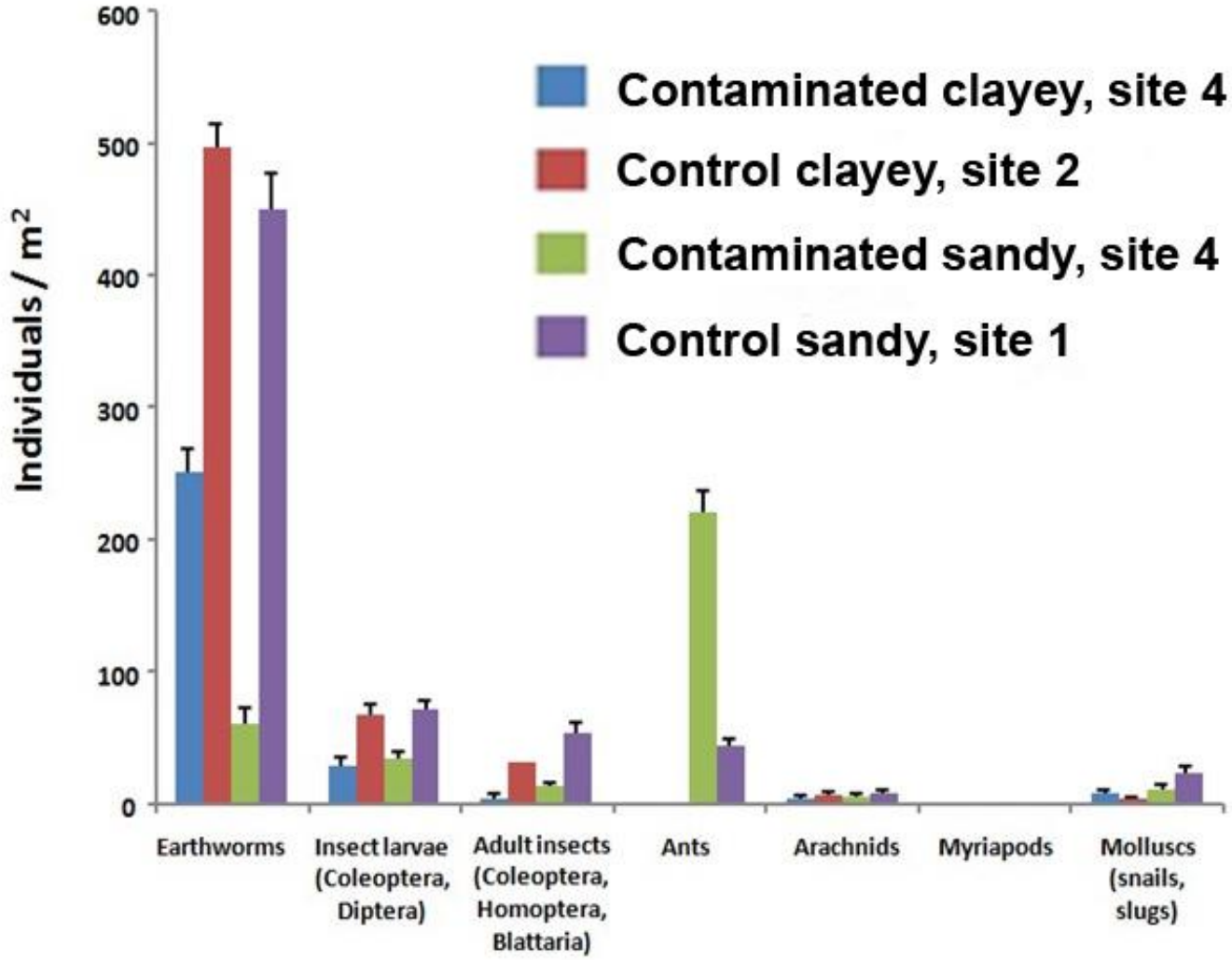
- Species diversity:
  - macrofauna taxonomic identification (4 sites, 30 plots per site)
  - Plants/insects (Russian partners

Radionuclide concentration in soil

Specific activities of radionuclides in soil, Bq / kg	Site 1 (Reference)	Site 2 (Reference)	Site 3	Site 4
U	2.6 ± 0.7	7.5 ± 1.5	21 ± 2	38 ± 17
^{230}Th	16 ± 7	18.5 ± 2.2	1000 ± 51	3240 ± 1199
^{226}Ra	38 ± 12	30.5 ± 10.7	6260 ± 355	10923 ± 3115
^{210}Po	56 ± 16	134 ± 20	4008 ± 57	7666 ± 2191
^{210}Pb	78 ± 21	37 ± 5.5	931 ± 430	1835 ± 896
Dose rate of γ-radiation in the air at 1 m from the soil surface, mGy / h	0.08–0.12	0.09–0.12	0.14–5.4	2.5–10.5

Mean **external** soil oranisms µGy/hr: Clayey 17, Sandy soil 14; control 0.05-0.1
Mean **Internal** soil organisms: µGy/hr: Clayey 80, Sandy soil 60; control 0.5-1
Gut dose could be up to 1 mGy/hr

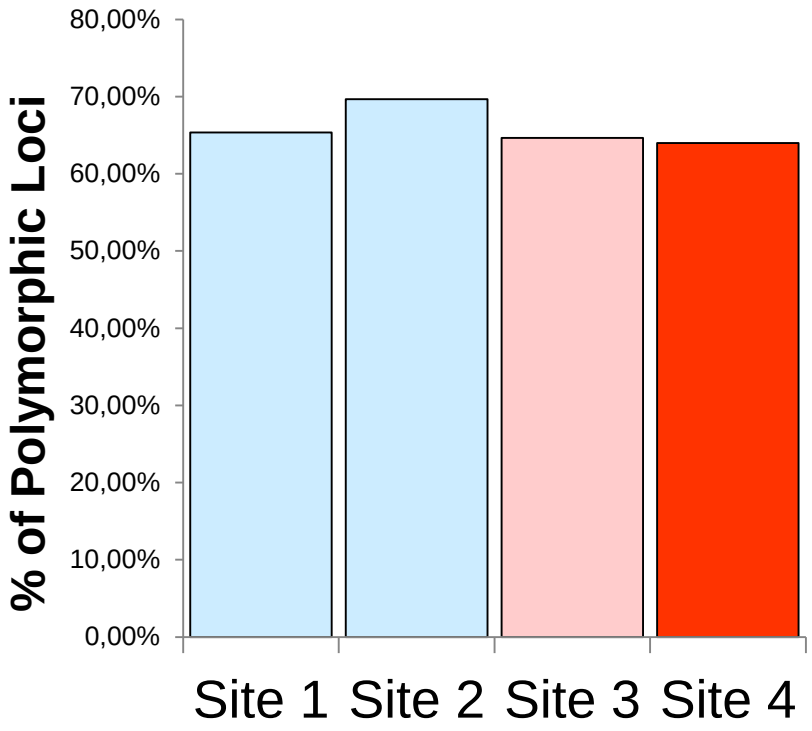
Soil invertebrate density

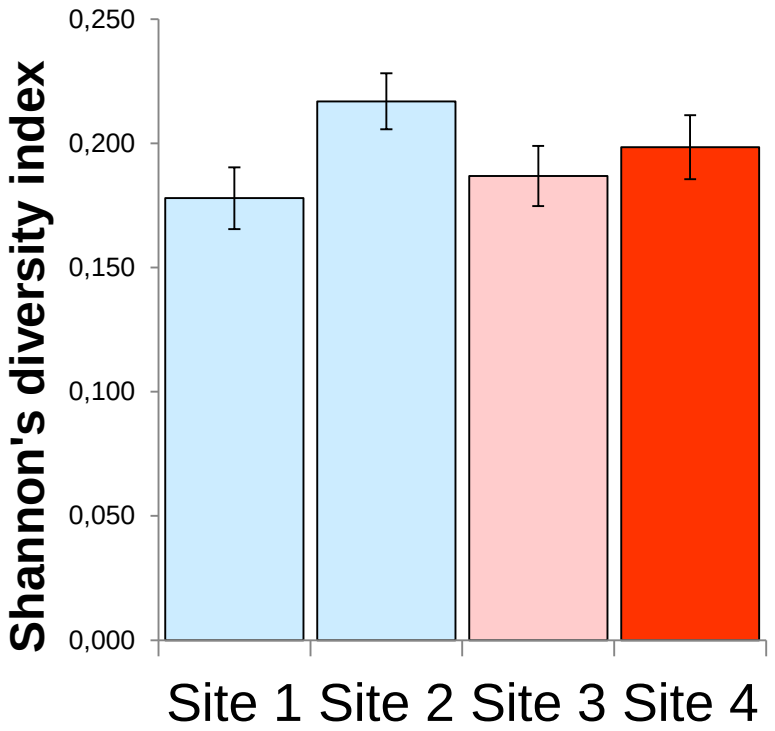


Field Doses Komi max 1-200 µGy/hr

Lapied, et al, in prep

Worm Genetic diversity – no difference





Genetic diversity of earthworms, sampled from contaminated and reference sites. Percent of polymorphic loci, and Shanon's diversity index were calculated from binary AFLP data.

Adaptation – DNA damage repair

Analysis of DNA-damage by Comet assay

Site 1

Site 4

A. caliginosa

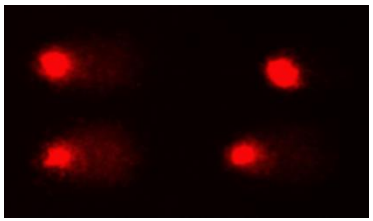
4 Gy
(0.7 Gy/min)

1 min
5 min
30 min
60 min
240 min

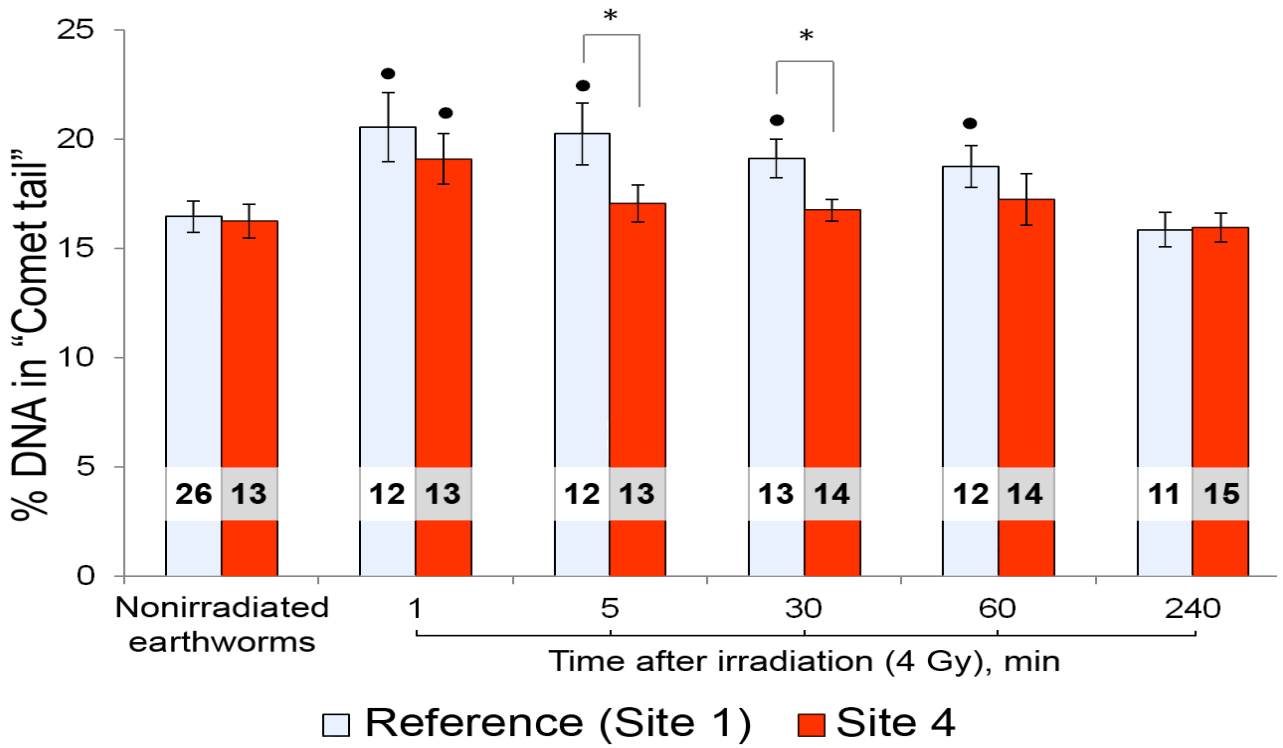
Time after irradiation

Freezing in liquid nitrogen

Alkali comet assay



DNA Repair dynamics



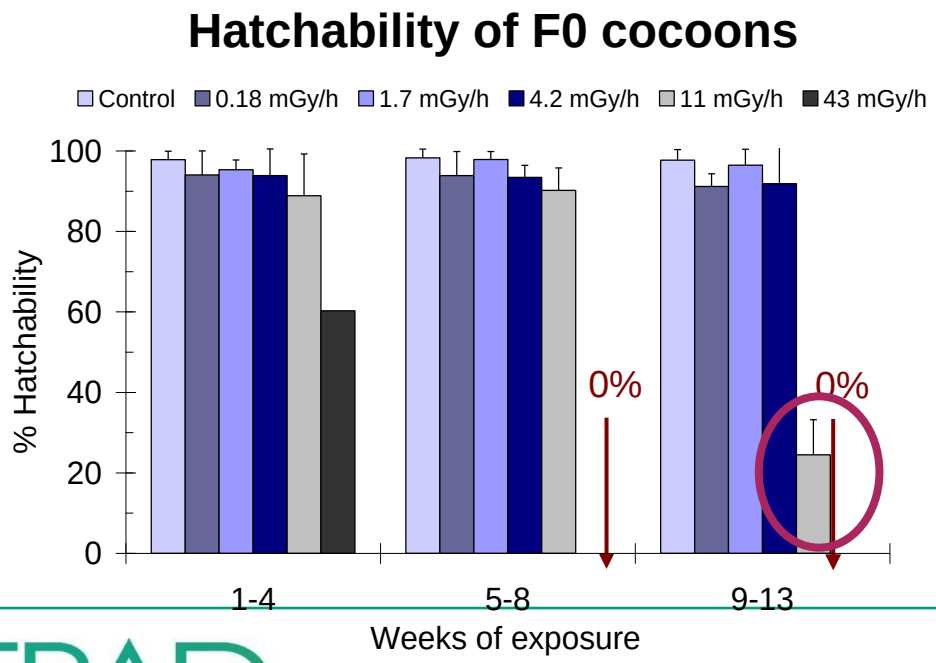
Repair dynamic of DNA damages induced by acute additional irradiation at 4 Gy in *A. caliginosa* from sites 1 and 4. * – difference between worms from the control and contaminated sites is significant at $p < 0.05$. ● – difference from the spontaneous level of DNA-damages is significant at $p < 0.05$ (Student t-test).

Laboratory Studies

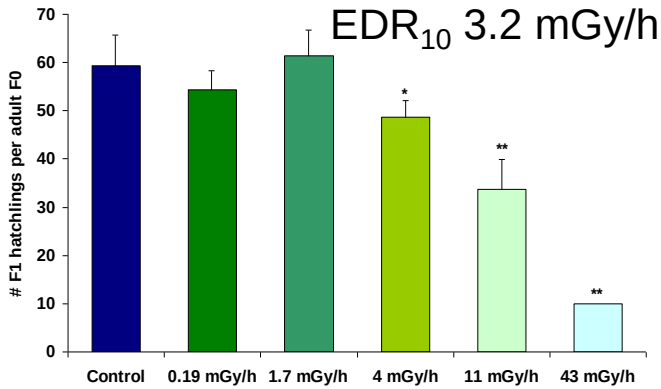
FIGARO – Low dose irradiation source, CERAD

Earthworm Reproduction Study

- Species Eisenia fetida
- Significant effects on cocoon hatchability (11 mGy/hr) – but not before 9-13 weeks
- Correlation with DNA damage (Comet assay) - Hertel-Aas et al 2010) and apoptosis (Lapied, in prep)



- **Reduction in the total number of offspring produced by each F0**

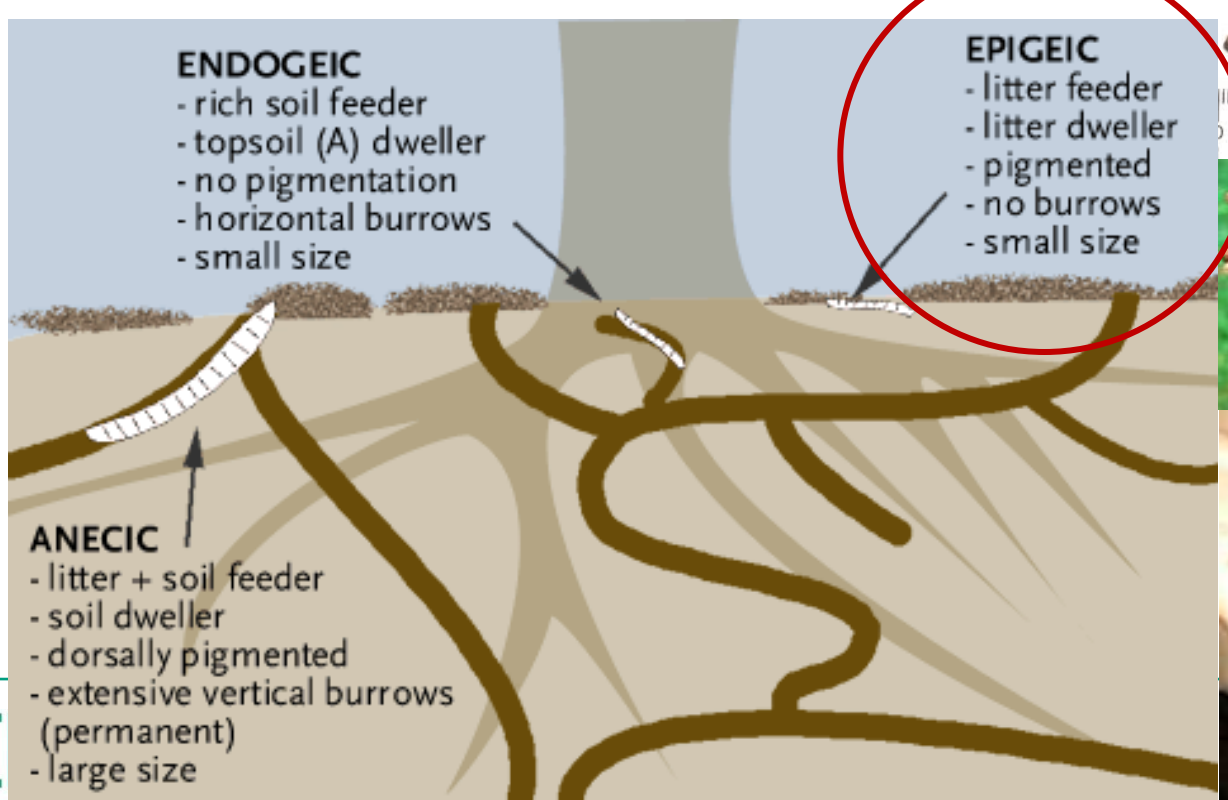


Hertel-Aas *et al*., Radiation Research, 2007

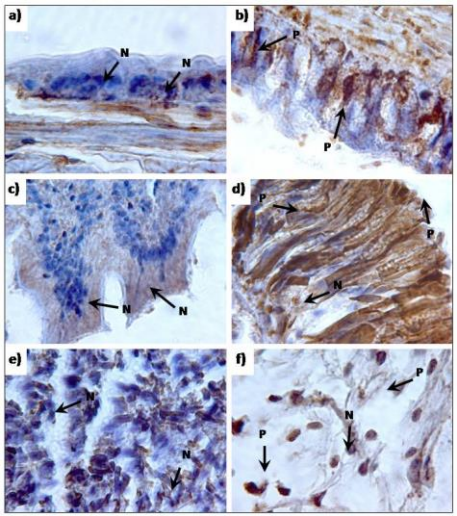
“An earthworm is not just an earthworm”

- The total number of species is estimated to exceed 2000
- Three major ecological groups of earthworm have been identified based on the feeding and burrowing behaviours of the different species.

E. fetida



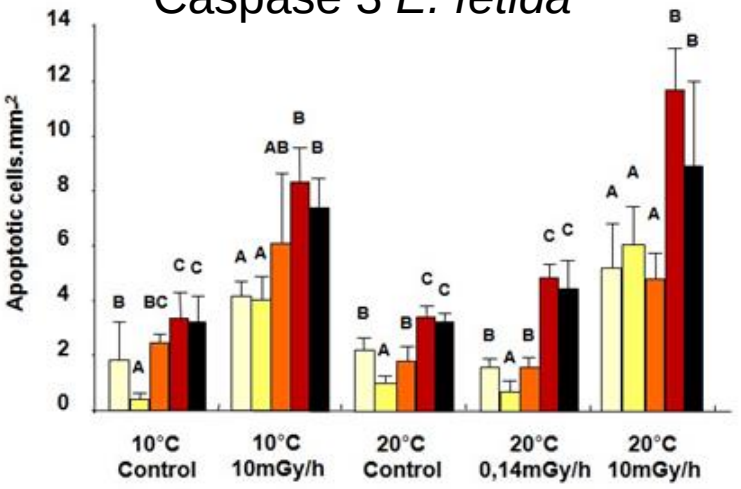
FIGARO Exposures: Apoptosis assessed by Tunel, Apostain and Caspase 3

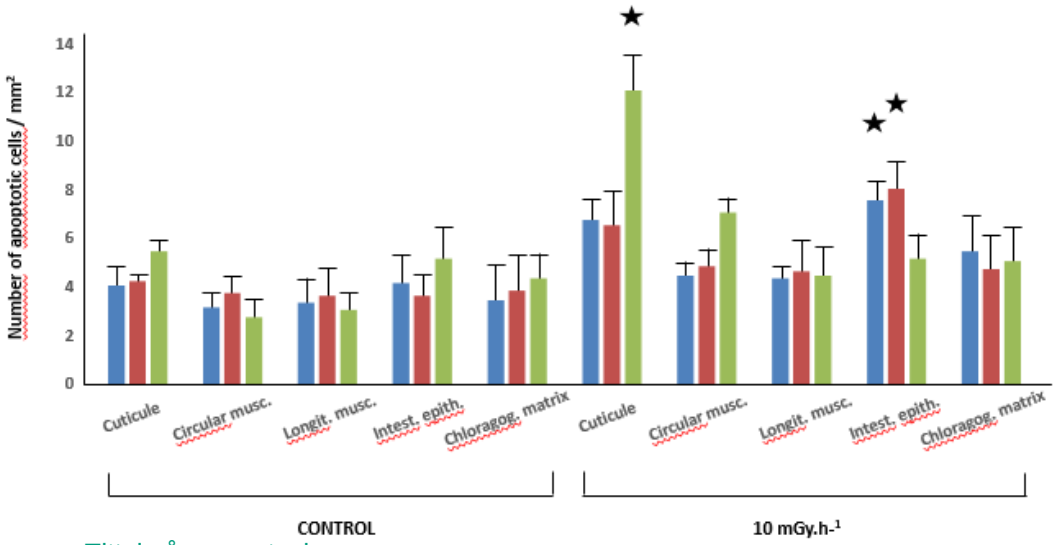


Apostain

Increase in apoptic cells seen after exposure to gamma radiation: 10 mGy/hr, 2 weeks exposure

Caspase 3 *E. fetida*





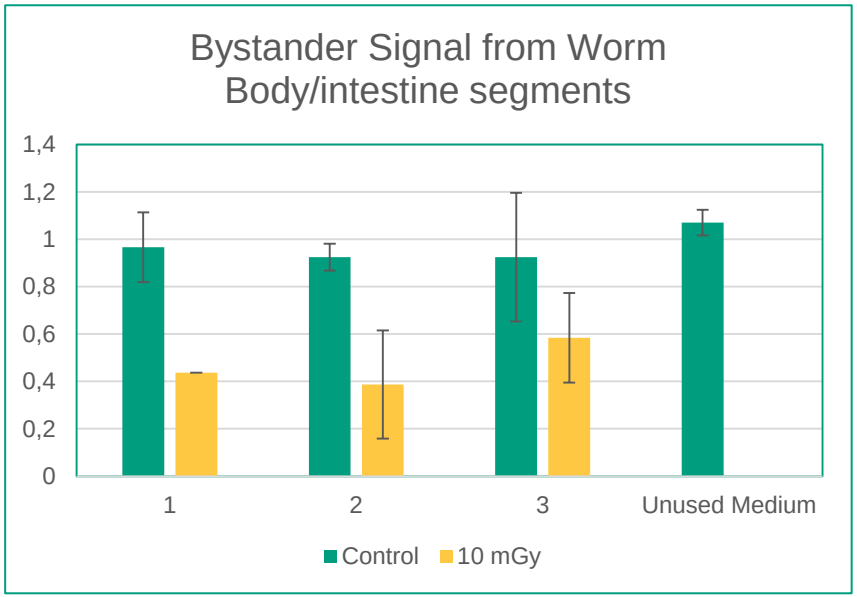
Cuticule
Circular musculature
Longitudinal musculature
Intestinal epithelium
Chloragogenous matrix

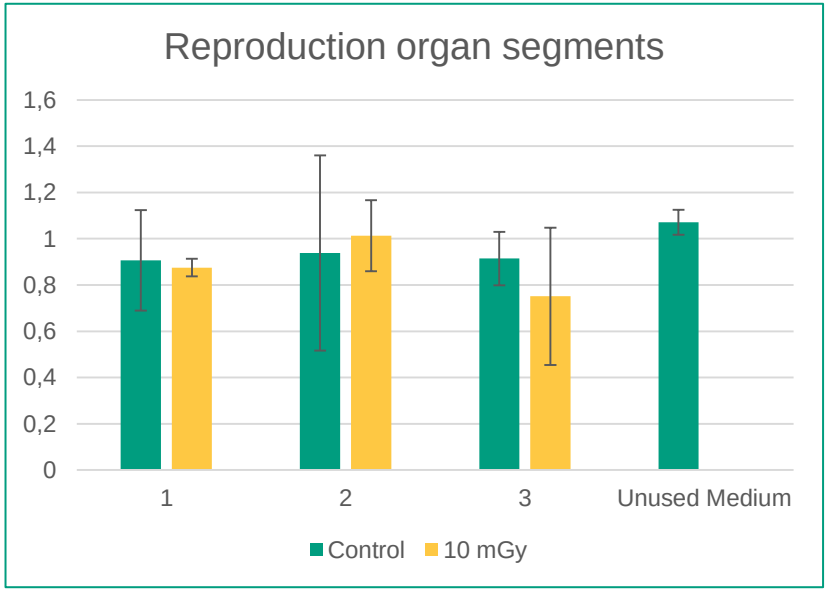
Aporrectodea rosea (endogeic species)
Aporrectodea caliginosa (endogeic species)
Dendrodrilus rubidus (epigeic species)

Field site species

Lapied et al, Environment International,; Lapied et al, in prer

FIGARO Exposures: Bystander signal assessed by clonogenic assay, 24 hr at 10 mGy/hr





1 and 2 – A. calignosa

3 – D. rubidius

- Comparison with same species collected at Chernobyl (*A. caliginosa*) and Fen (*A. caliginosa, D. rubidus*) and ongoing gut microbiome and mitochondria analysis

Lapied et al, Environment International,; Lapied et al, in prer

Thank you!

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Turid Hertel-Aas (NMBU), Gunnar Brunborg (FHI), Alicja Jaworska (NRPA), Brit Salbu (NMBU). Elena Belykh, Anna Kaneva, Illa Velegzhaninov, Tatiana Evseeva, Vladimir Zainullin, (The Institute of Biology, Komi Scientific center of UB RAS) and many more…

… must recognise that correlations and interpretation of cause and effect is challenging in environmental science

Species identification

COI sequencing.

The dominant species was defined as *Aporrectodea calliginosa and Lumbricus rubellus.*

… genetically separated from samples collected in the US and Europe.