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Population
Radiobiology
Lab

Stress memory in *Plantago major* from the zone of radioactive contamination (East Ural Radioactive Trace, Russia)

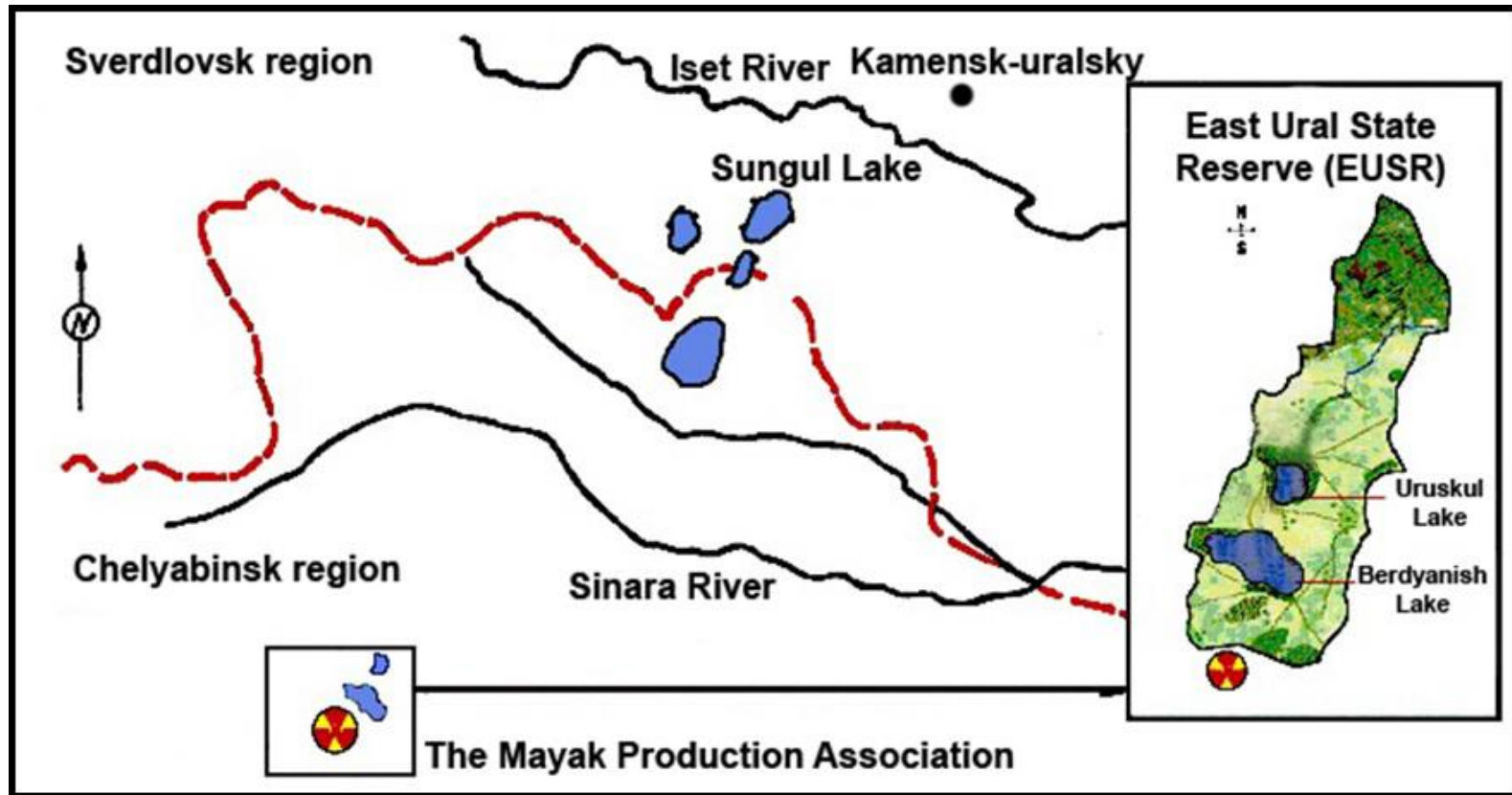
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The East Ural Radioactive Trace



The East Ural Radioactive Trace (EURT) appeared in 1957 after the accident at the Mayak Production Association, where a tank with radioactive waste exploded (Urals, Russia). The main contaminant in the EURT is ^{90}Sr . Additional contamination of this zone occurred in 1967 as a result of the wind transfer of radioactive sand from the shores of Lake Karachay, which had been used as an open reservoir for radioactive wastes (with ^{137}Cs being the main contaminant).

Absorbed doze rates for parental *P. major* plants

| Plot | Specific radioactivity in the soil, Bq/kg (average and min-max) | | | Concentration ratios | | | Absorbed dose rate, $\mu\text{Gy/h}^*$ |
|----------------|--|-------------------|----------------------|----------------------|-------------------|----------------------|--|
| | ^{90}Sr | ^{137}Cs | 239, 240 Pu | ^{90}Sr | ^{137}Cs | 239, 240 Pu | |
| Backgr ound | 6.92 (2.0-12.95) | 9.89 (3.65-6.5) | n.o. | 0.66 | 0.054 | 0.0014 | 0.108 |
| EURT-10 | 90900 (155-271000) | 4400 (138-18500) | 235 (47-399) | | | | 73.1 |
| EURT-5 | 195000 (88300-292000) | 9860 (3990-17000) | 514 (60-1350) | | | | 157.1 |

*Absorbed dose rate calculated including background radiation which equal to 0.1 $\mu\text{Gy/h}$ for the Ural region

Karimullina E., Mikhailovskaya L.N., Pozolotina V., Antonova E. Radionuclide uptake and dose assessment of 14 herbaceous species from the East-Ural Radioactive Trace area using the ERICA Tool // Environmental Science and Pollution Research. 2018.

<http://dx.doi.org/10.1007/s11356-018-1544-y>



Research purpose

Study of the persistence of changes in viability, lipid peroxidation and antioxidant capacity in two generations of greater plantain (*Plantago major* L.) after the cessation of ionizing radiation impact.



Stages of the study

- 1) To obtain two generations (F2 and F3) of *P. major* seed progeny under "clean" leveled agricultural conditions from seeds collected directly from the East Ural Radioactive Trace and background plots (F1-generation);
- 2) To estimate viability of F1, F2 and F3 generations of seed progeny of *P. major* from different zones during laboratory experiment on seed germination by roll culture method;
- 3) To determine the content of malondialdehyde (a marker of oxidative stress), superoxide dismutase activity and the total content of low molecular weight antioxidants in the seedlings obtained in the laboratory experiment.

Obtaining of seed material

#1 Collection of F1-generation seeds in natural populations in the EURT zone and background plots



#2 Growing plants of F1-generation on a clean, leveled agricultural background



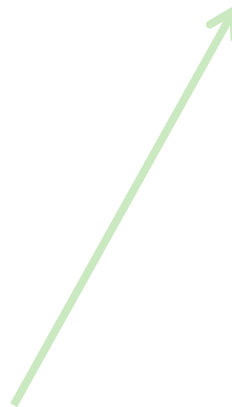
#3 Collecting of F2-generation seeds



#4 Growing plants of F2-generation on a clean, leveled agricultural background



#5 Collecting of F3-generation seeds



Methods



21-day-old *P. major* seedlings grown by the roll culture method



Viability parameters:

- survival rate of seedlings at the stage of leaf formation (at 21 day), % to the number of sown seeds ;
- root length, mm.

A mixture of seeds from each sample was used for germination; 50 seeds were sown in 5 replications.

A total of 1500 seeds of each generation were used.

Mann-Whitney (U-test) and Kruskal-Wallis (H) nonparametric criteria were used to test statistical hypotheses. Calculations were performed in the program STATISTICA 10.0 (StatSoft Inc., 2011).

Biochemical parameters:

- Content of malondialdehyde* (MDA, a product of lipid peroxidation)
- Superoxide dismutase activity* (SOD, enzyme of antioxidant defense)
- The total content of low molecular weight antioxidants ** (LMWA)

*Measurements were performed using a microplate spectrophotometer SpectraMax Plus 384 (Molecular Devices, США);

**Measurements were performed using a spectrophotometer SPEKOL 1300 (Analytik Jena AG, Германия).

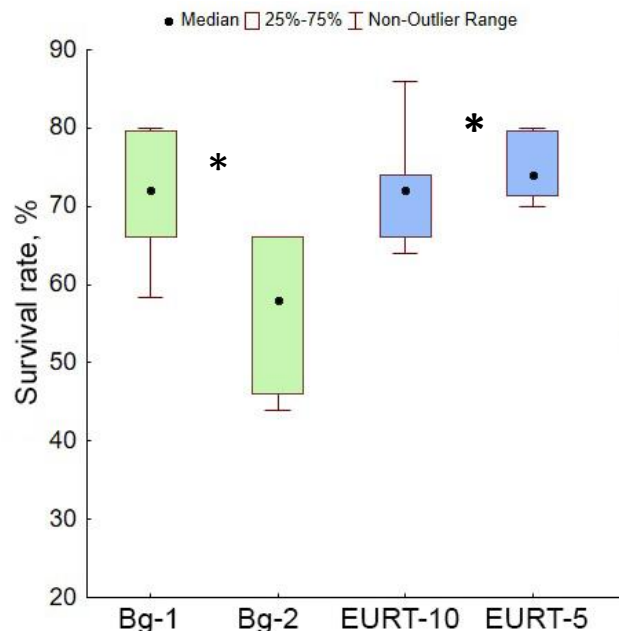
Detailed description of methods is presented in our paper:

Shimalina N.S., Pozolotina V.N., Orekhova N.A., Antonova E.V. Assessment of Biological Effects in *Plantago major* L. Seed Progeny in the Zone of Impact from a Copper Smelter // Russian Journal of Ecology. 2017. V. 48. № 6. P. 513–523.

<https://doi.org/10.1134/S1067413617060108>

Results: survival rate of seedlings

F1



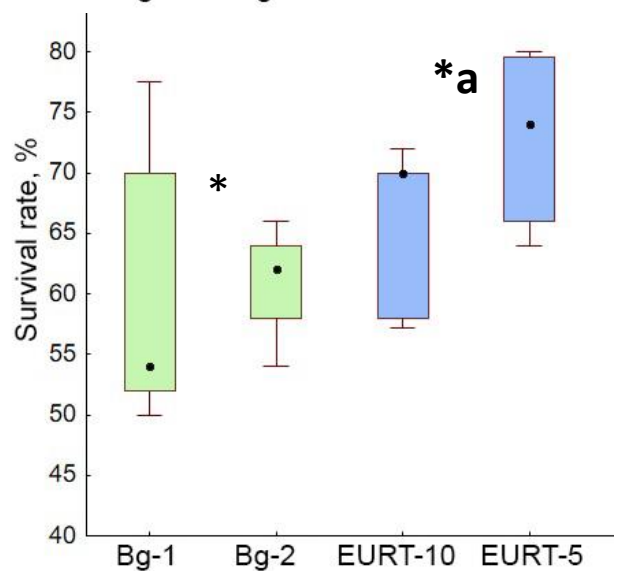
In a succession of generations of *P. major* from the EURT zone, it is impossible to identify a certain relationship between the survival rate of seedlings and the radiation load that their ancestors experienced over a long period of time due to the high variability of the parameter.

*no significant differences between samples within zones, *U*-test, $n=5$, $p=0.347-0.602$;

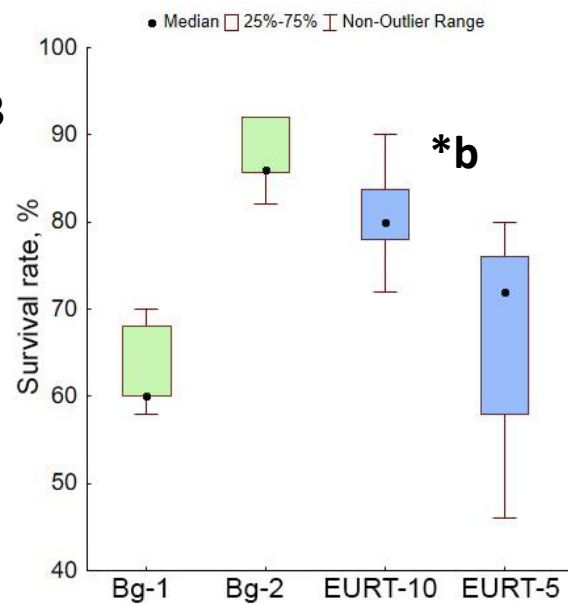
a – significant differences in comparison to the pooled background sample, $p=0.041$;

b – significant differences in comparison to Bg-2, $p=0.012$.

F2

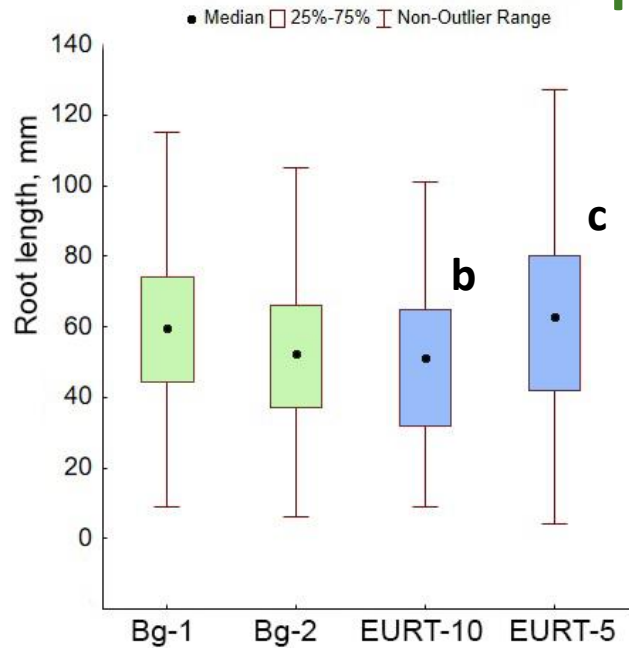


F3



Root length

F1



The highest root length was observed in the EURT-5 sample in all generations.

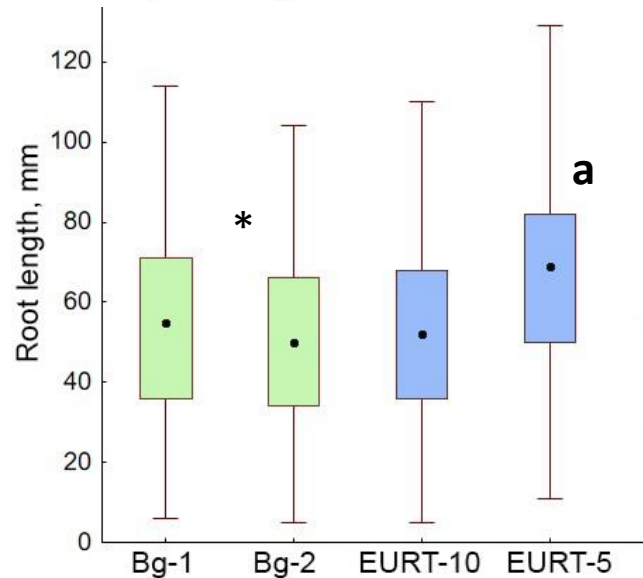
*No significant differences between samples within zones, *U*-test, $n=117-234$, $p=0.054-0.103$;

a – significant differences in comparison to the pooled background sample, $p<0.001$;

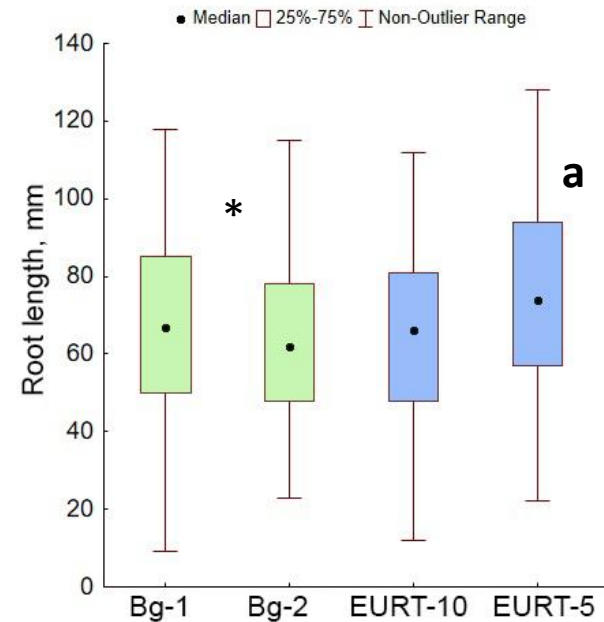
b – significant differences in comparison to Bg-1, $p<0.001$;

c – significant differences in comparison to Bg-2, $p<0.001$.

F2

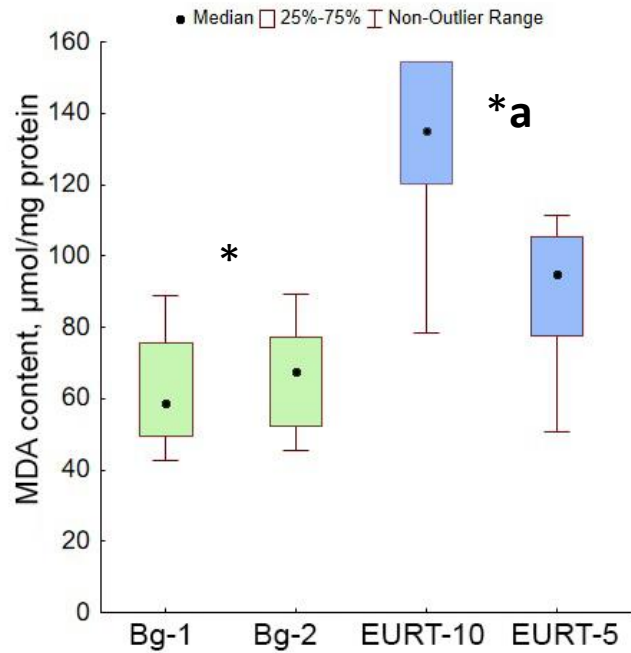


F3



MDA content

F1

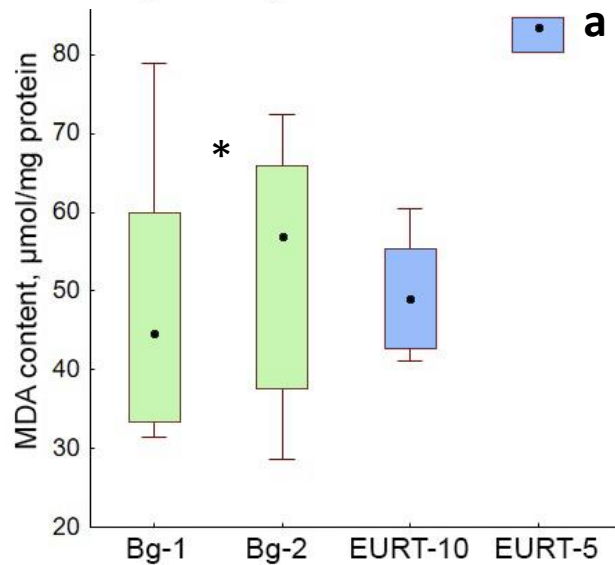


The increase in MDA content in F1 seedlings from the EURT-5 site with the highest level of radiation exposure proved persisted in F2 and F3 generations.

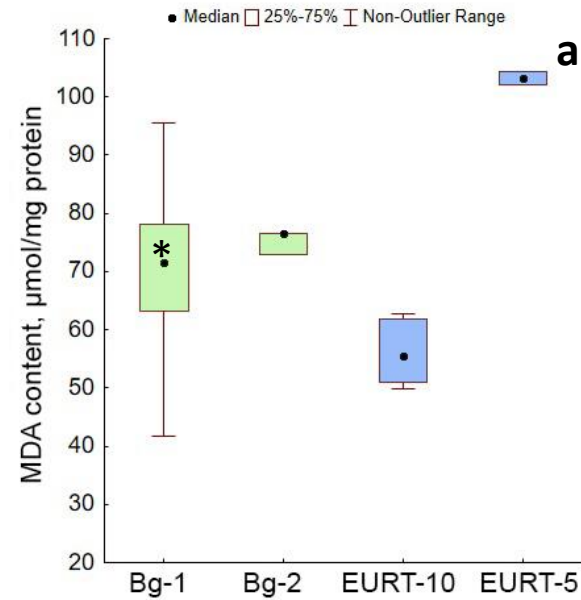
*no significant differences between samples within zones, *U*-test, $n=5$, $p=0.060-0.676$;

a – significant differences in comparison to the pooled background sample, $p=0.004$.

F2

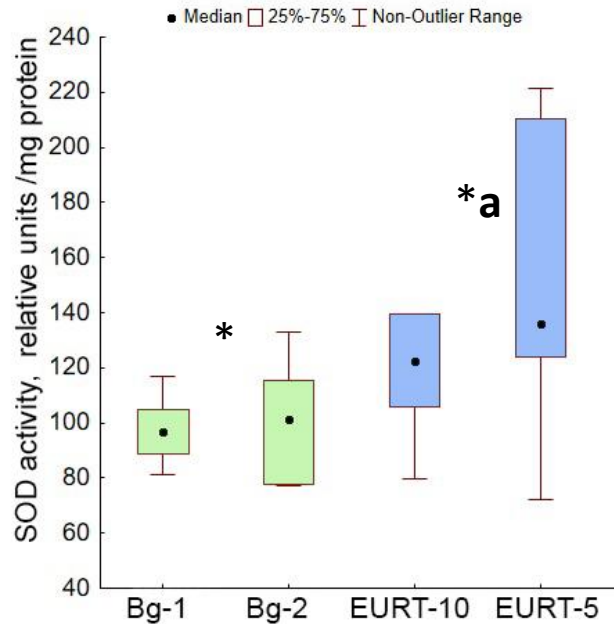


F3



Superoxide dismutase activity

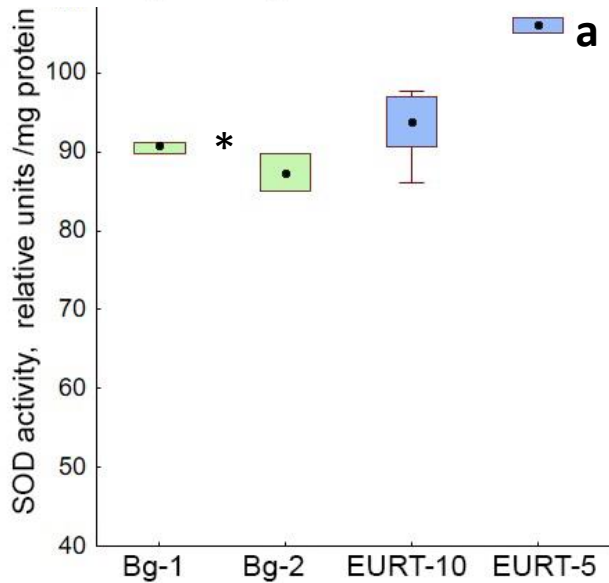
F1



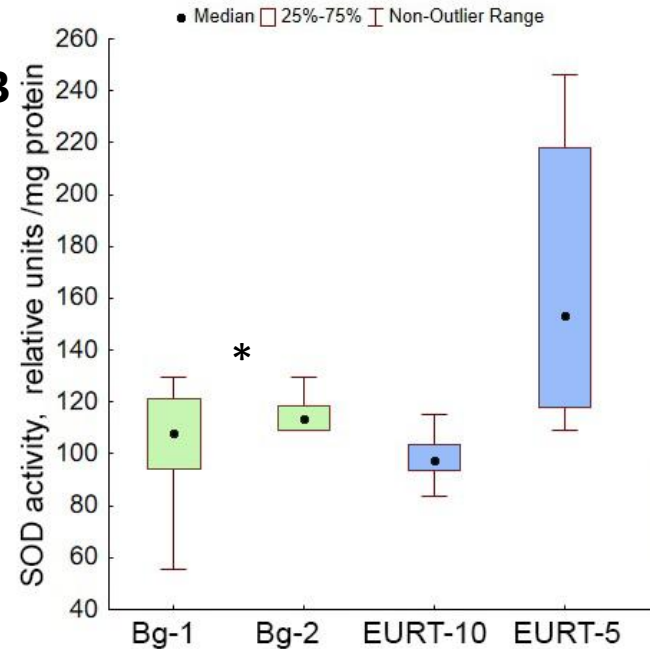
The increase in SOD activity was found in the F1 and F2 generations (EURT-5). The effect was leveled in the F3 generation.

*no significant differences between samples within zones, *U*-test, $n=5$, $p=0.531-1.000$;
a – significant differences in comparison to the pooled background sample, $p=0.017-0.045$.

F2

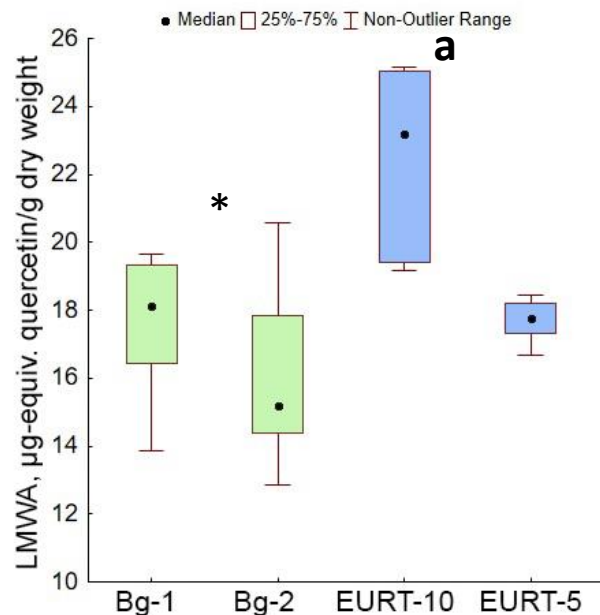


F3



The total content of low molecular weight antioxidants

F1

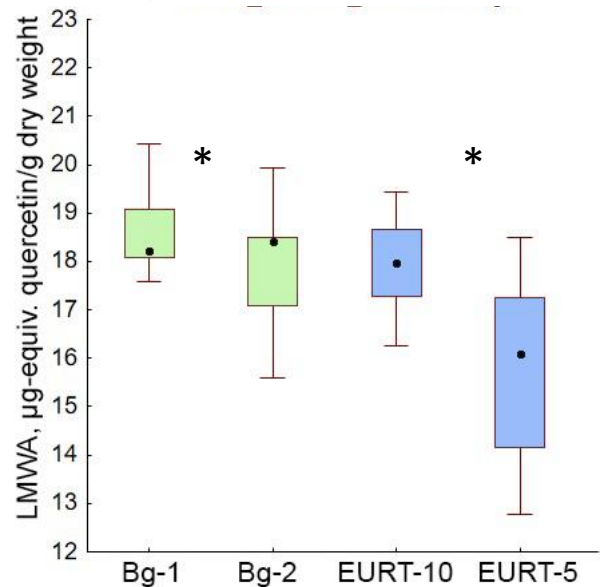


The increase of total LMWA content in the seed progeny of F1 generation from EURT-10 site did not persist in the next generations after the stress removal.

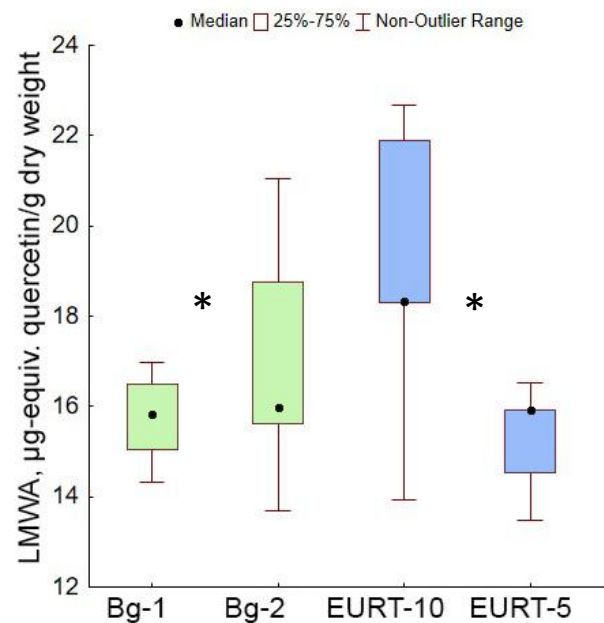
*no significant differences between samples within zones, *U*-тест, $n=5$, $p=0.095-0.676$;

a – significant differences in comparison to the pooled background sample, $p=0.017$.

F2



F3



Conclusion

| Parameter | Samples from the East Ural Radioactive Trace compared to background | | |
|----------------------------|---|----------|----------|
| | F1 | F2 | F3 |
| Survival rate of seedlings | ≈ | ↑ | ≈↑ |
| Root length | ↓↑ | EURT-5 ↑ | EURT-5 ↑ |
| MDA content | ↑ | EURT-5 ↑ | EURT-5 ↑ |
| SOD activity | ↑ | EURT-5 ↑ | ≈ |
| LMWA content | EURT-10 ↑ | ≈ | ≈ |

Current study with the example of *P. major* showed that the features of oxidative stress in plant populations affected by low-dose irradiation could persist in two subsequent generations after the cessation of radiation exposure.



Acknowledgements

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Natalya A. Orekhova

Thank you for your attention!