

Cellular immune functions of *Eisenia fetida* are affected with sub-lethal dose of Polycyclic Aromatic Hydrocarbons

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Exposure to ubiquitous environmental chemicals, such as polycyclic aromatic hydrocarbons (PAH), may affect to earthworm cellular immune-defense. We examined cellular functions of coelomocytes such as phagocytosis, H₂O₂ activity, NK activity and P450 by flow cytometry after exposure contaminated soil with 7,12-dimethylbenzanthracene (DMBA) and benzo-a-pyrene (BaP). In addition, presence of cytochrome P450 examined by immunohistochemistry, western blots and flow cytometry and RT-PCR. Sexually matured *Eisenia fetida* kept in dark at 18°C with various dose of DMBA and BaP (0-100 µg) contaminated artificial soil (20g each in glass jar) and periods (0 to 2 weeks). Phagocytosis was assessed by fluorescence beads ingestion test and H₂O₂ activity examined using 2',7'-dichloro-fluorescein diacetate that measured by flow cytometry after 2h incubation, respectively. NK activity accessed by 4h MMT assay using K563 cells as a target. The result indicated that those cell functions were down regulated by sub-lethal dose and time dependent manner. While, increased expression of P450 (40 to 60 %) was detected by flow cytometry and western bolt as well as mRNA transcript. Immunohistochemical analysis of cytochrome P450 revealed that skin and coelomocytes were major catabolized sites for PAH in *Eisenia fetida*. These non-specific cellular functions permit us to evaluate biological activity of earthworm and the effect of soil contamination for ecotoxicological studies.