Sveučilište u Zagrebu
Fakultet kemijskog
inženjerstva i tehnologije
Zavod za industrijsku ekologiju

Kolegij:
PRIMJENA EKOTOKSIKOLOGIJE
4. predavanje

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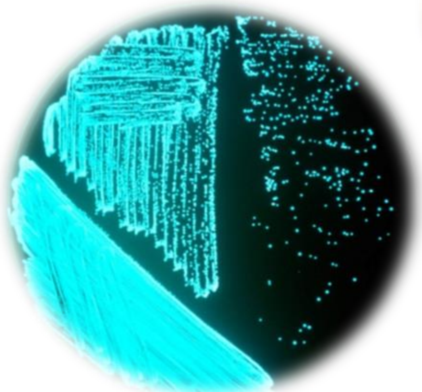
SADRŽAJ

TOKSIČNOST TLA i SEDIMENTA



TOKSIČNOST TLA

- Tlo mješavina živih i neživih tvari
- Na BIORASPOLOŽIVOST i TOKSIČNOST onečišćujuće tvari u tlu utječe: sposobnost tla da adorbira, metabolizira, pohranjuje i akumulira onečišćujuću tvar
- Testovi toksičnosti temelje se na razumijevanju da u određenim uvjetima ispitivanja postoji mjerljiv i progresivan odnos između doze i učinka
- Ispitivanjem toksičnosti mjeri se krajnja točka ili skupine krajnjih točaka (npr. smrtnost, reproduktivna sposobnost, brzina rasta) u rasponu poznatih koncentracija onečišćujuće tvari
- Bitno poznavanje fizikalno-kemijskih karakteristika ispitivane tvari, kao i tla
- Održavanje integriteta tla prilikom uzorkovanja, transporta i laboratorijskih ispitivanja – utječe na stupanj toksičnosti

TOKSIČNOST TLA

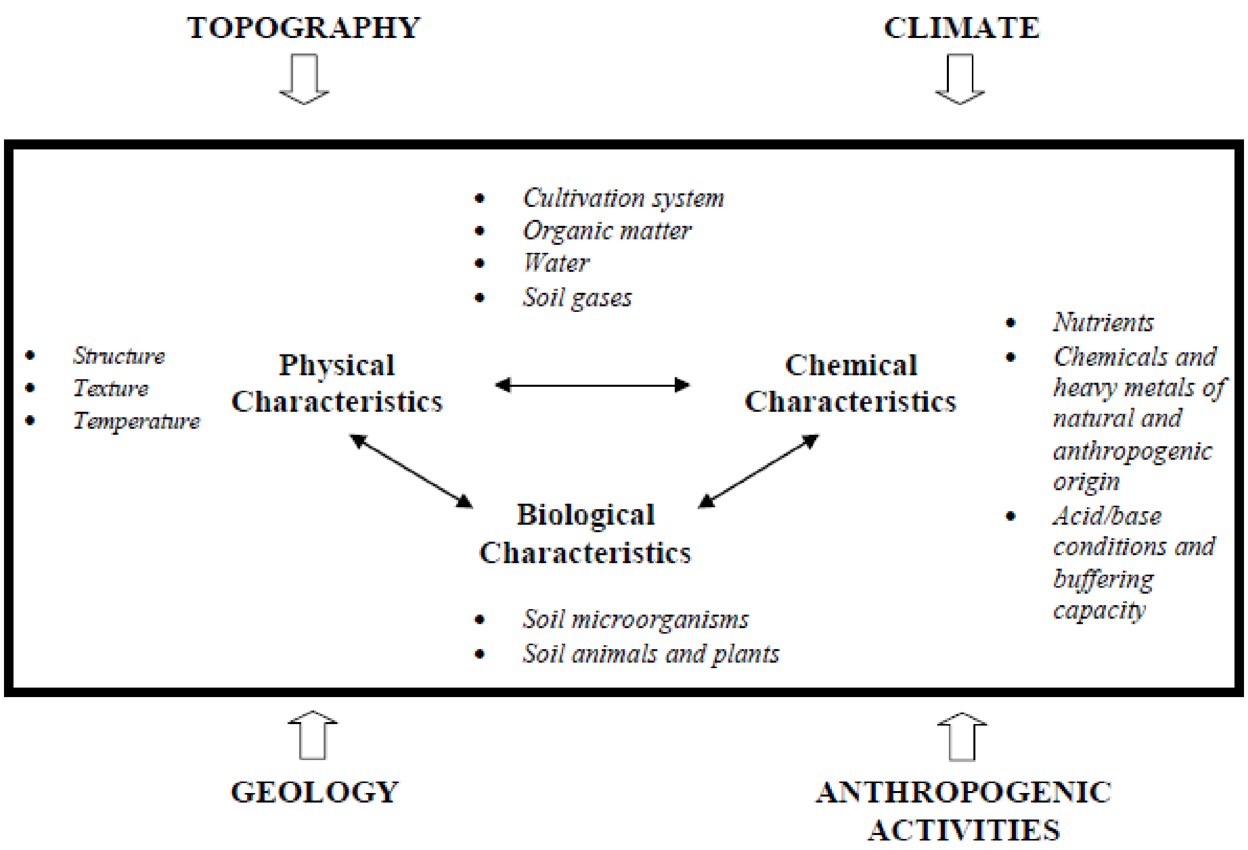


Figure I-1. The complex structure of soil and its influences. Modified from Torstensson 1997, 1998

TOKSIČNOST TLA

- Procjena kakvoće tla uglavnom se temelji na utvrđivanju koncentracija ONEČIŠĆUJUĆE TVARI U TLIMA
- Dobivena koncentracija onečišćujuće tvari se uspoređuje s graničnom (dozvoljenom) koncentracijom tvari te se procjenjuje stupanj onečišćenja tla
- Procjena rizika – predviđanje vjerojatnosti štetnog učinka
- *Procjena rizika po okoliš uključuje*:

✓ opis potencijalno štetnih učinka po okoliš (identifikacija opasnosti) i procjena opsega učinaka (procjena *doza-učinak*);

✓ određivanje potencijalno izloženih receptora (makromolekula u organizmu – na receptor se vežu ligandi (lijekovi, hormoni) – mogu smanjiti ili povećati staničnu funkciju) i uvjeta izloženosti (procjena izloženosti) i

✓ procjena vjerojatnosti nastanka štetnog učinka na temelju usporedbe učinaka i izloženosti

TOKSIČNOST TLA

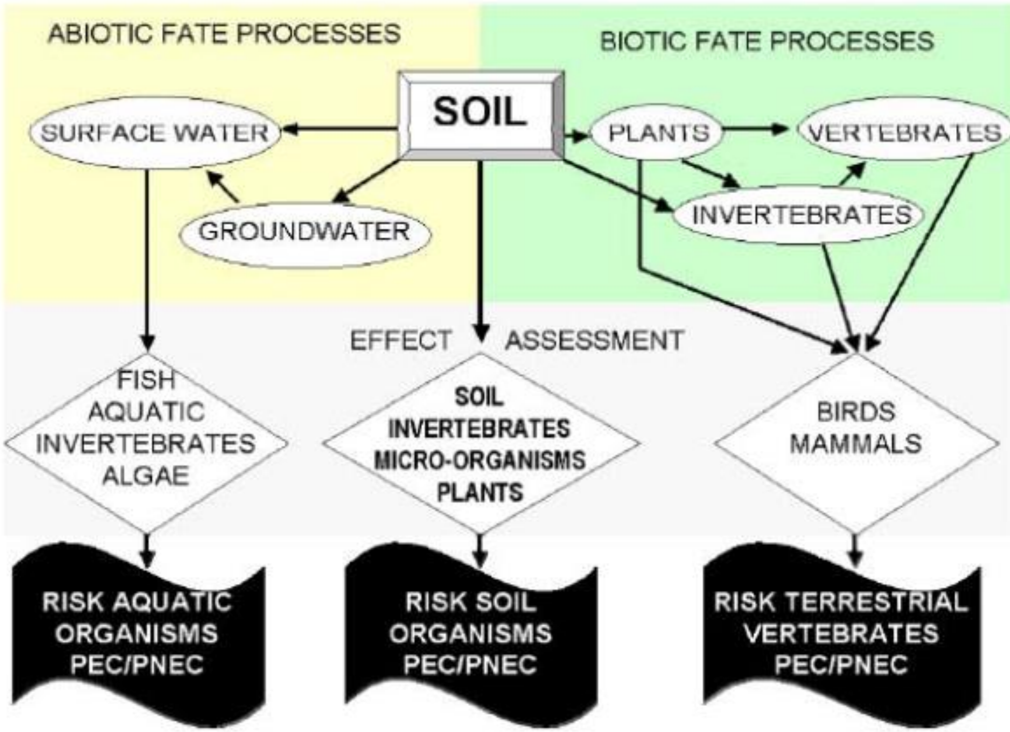


Figure 1. Graphic representation of the generic conceptual model. Symbols represent sources (double square), exposure estimations (ellipses), effect assessments (rhombus) and risk characterization processes (flags).

PROCJENA IZLAGANJA

PROCJENA UČINKA

PROCJENA RIZIKA

CONTAMINANT IN SOIL → EXPOSURE PATTERN → RECEPTOR

TESTOVI TOKSIČNOSTI TLA

- Toksičnost u uzorcima tla se može odrediti na 2 načina:

1. Uzorci tla su „razrijeđeni" s referentnim tlom (tlo koje nije onečišćeno) i različite koncentracije tla su ispitane – ovim pristupom dobije se rezultat doza-učinak kao i kad se ispituje čista tvar
2. Uzorci tla nisu razrijeđeni

- Prednosti i nedostatci prvog načina određivanja toksičnosti:

✓ priprava različitih razrjeđenja u cilju dobivanja LC_{50}
✓ ponekad LC_{50} se ne može odrediti ukoliko uzorak nije dovoljno toksičan
✓ dodatne informacije o onečišćenom tlu koji potencijalno predstavlja rizik po okoliš
✓ bioraspoloživost onečišćujuće tvari

- Oba testa se uspoređuju s referentnim tlom (najbitnija stavka) – na toksičnost organizma uvelike utječu fizikalno-kemijske karakteristike tla

TESTOVI TOKSIČNOSTI TLA

- Za procjenu toksičnosti tla često se koriste VODENI ORGANIZMI – pretpostavka: onečišćujuća tvar djeluje na organizme u tlu putem vodene faze
- Testovi toksičnosti vodenim organizmima – nisu najbolji izbor jer se pretpostavlja *podjednaka osjetljivost* organizama u tlu i vodenih organizama
- Toksičnost tla bi se trebala ispitati na organizmima u tlu, a *leaching* testovi (s vodenim organizmima) bi se trebali samo koristiti kad se procjenjuje utjecaj / procjena rizika onečišćenog tla na podzemne i površinske vode
- Priprema eluata (leaching) – važan korak za određivanje toksičnosti tla
- Destilirana ili deionizirana voda
- Šaržni test, test u koloni ili test na polju
- Šaržni test – tlo u kontaktu s vodom se miješa određeno vrijeme (omjer tla i vode 1:2 ili 1:10) metoda - DIN 38414
- Test u koloni – leachate prolazi kroz tlo koje se nalazi u koloni – simulacija uvjeta u okolišu

TESTOVI TOKSIČNOSTI TLA

- Prilikom provođenja testa toksičnosti – koristiti negativnu, pozitivnu i referentnu kontrolu
- Kontrolna grupa – isti eksperimentalni uvjeti - primjenjuje se u cilju osiguranja integriteta korištenog testa, uključujući:

1. Mjerenje prihvatljivosti testa
2. Osiguranje zdravlja, kvalitete i osjetljivosti testnih organizama
3. Osiguranje uvjeta ispitivanja (npr. rukovanje organizmom, uzgoj i parametri okoliša) prikladni za provođenje testa
4. Dobiveni rezultati omogućuju bolju interpretaciju

- Kod nekih testova nije potrebno korištenje negative, pozitivne i referentne kontrole

TESTOVI TOKSIČNOSTI TLA

- Test toksičnosti na više organizama - u cilju točne procjene stanja okoliša
- Različita osjetljivost testnih organizama na određene onečišćujuće tvari
- Odabrani testni organizmi – dovoljno osjetljivi na onečišćujuću tvar
- ORGANIZMI SE BIRAJU NA TEMELJU:
  - VAŽNOSTI VRSTE,
  - OSJETLJIVOSTI TESTA,
  - JEDNOSTAVNOSTI i
  - EKONOMIČNOSTI
- Različiti standardizirani testovi toksičnosti tla – OECD, ISO, ASTM, DIN, AFNOR, EPA
- BIOKEMIJSKI, FIZIOLOŠKI, REPRODUKTIVNI i BIHEVIORALNI učinci - pružaju podatke o toksičnosti
- Test izravne toksičnosti - mjere se parametri: SMRTONOSNOST, REPRODUKCIJA i RAST

TESTOVI TOKSIČNOSTI TLA

Table 1-1. Test Selection Considerations

Criteria	Considerations
Test Species	Sensitivity, exposure routes, life stage, life strategy, life form, species interaction, habitat
Chemical Factors	Volatility, concentration, distribution patterns, temporal or permanent exposure, persistence, known hazard, physico-chemical properties, water solubility, specific gravity, vapor pressure, adsorption capacity, hydrolysis in water, melting and boiling point, octanol/water partition coefficient (k_{ow})
Soil Characteristics	Soil type, soil source, soil content, soil purpose (e.g., agriculture), pH, moisture, macro- and micro-nutrient levels, water-holding capacity, existing microflora and fauna micro-climatic conditions
Test Conditions	Duration, cost-effectiveness, bias, precision, robustness, ruggedness, reproducibility, standardization, environmental applicability (temperature, pH, salinity, etc.), biological validity, statistical validity
Endpoints	Sensitivity, applicability to the chemical or anticipated exposure, chemical responsiveness, ecological realism

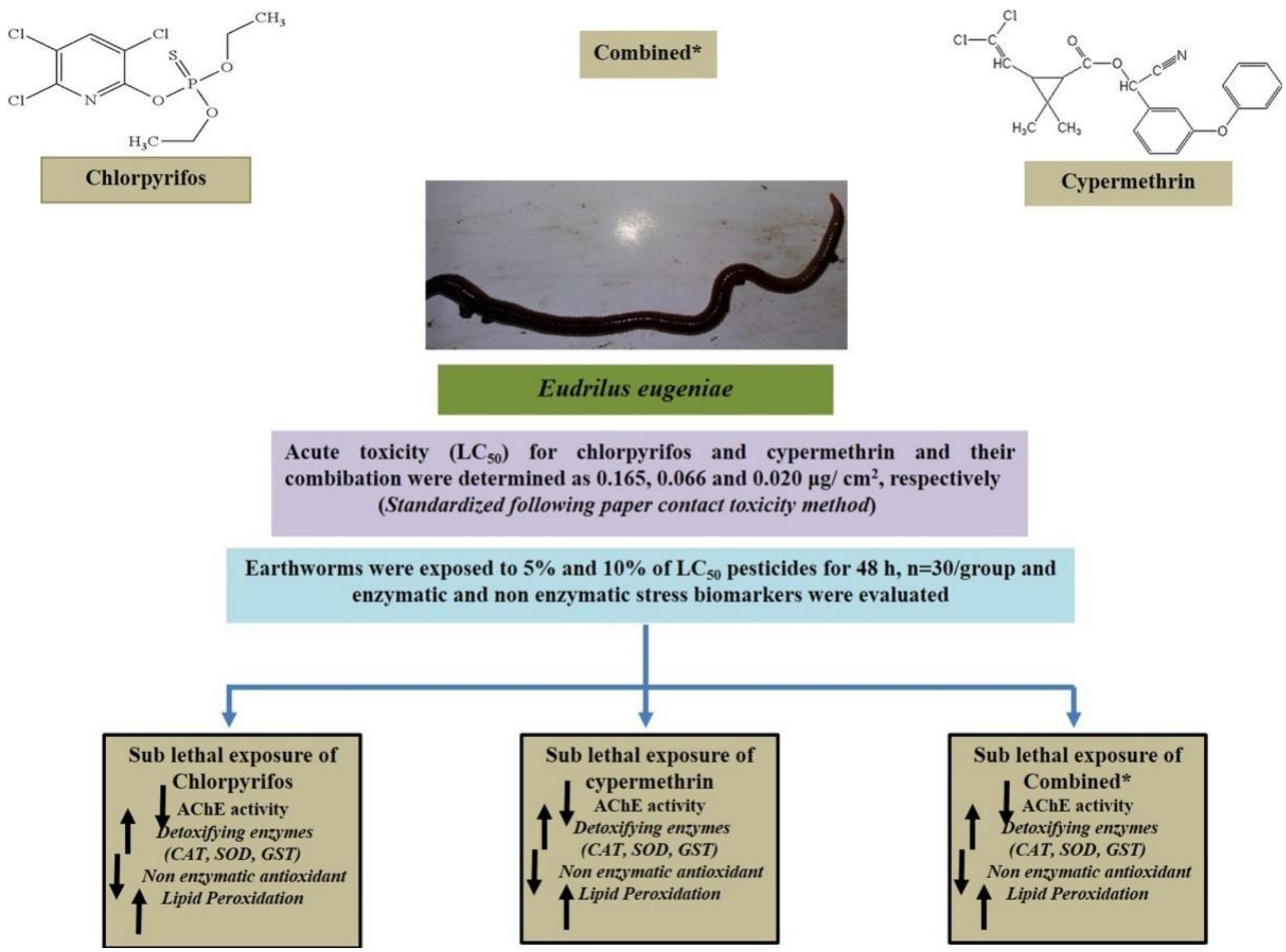
TESTOVI TOKSIČNOSTI TLA

- Najčešće korišteni testovi s BESKRALJEŠNJACIMA tla su:
  - GLISTE,
  - SKOKUNI (*Collembolan*) (člankonošci, nekoliko mm dugi, najbolji skakači u životinjskom svijetu),
  - *Enchytraeidae* (mali zemljani crvi) i
  - IZOPODI (čistači podloge, hrana malim životinjama)

TESTOVI TOKSIČNOSTI TLA

- Testovi s GLISTAMA – najčešće se primjenjuju za procjenu rizika kemijskih tvari – zakonski uvjet
- 2 testa s glistama su standardizirana – SMRTNOST i REPRODUKCIJA se određuje – *Eisenia* sp. (OECD 2004a)
- Može se pratiti i SUBLETALAN učinak (izbjegavanje, gubitak mase i dr.) i BIOKONCENTRACIJA (predviđanje izloženosti predatora štetnim tvarima putem hranidbenog lanca)
- *Enchytraeidae* – ekološki relevantna vrsta za ispitivanje toksičnosti tla
- Vrijeme reprodukcije 4-6 tjedna, gliste – 8 tjedana
- *Enchytraeus albidus* – standardizirani test OECD (2004b)
- BILJKE – ispitivanje kvalitete tla – FITOTOKSIČNOST – smanjenje rasta / biomase (može se pratiti i respiracija i određeni enzimi)
- Prati se i biokoncentracija – izloženost životinja štetnim tvarima putem hranidbenog lanca

TESTOVI TOKSIČNOSTI TLA



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TESTOVI TOKSIČNOSTI TLA

Table 1
Lethal concentrations (LC_{1-99}) of chlorpyrifos for earthworm, *Eudrilus eugeniae* (identified by well-defined clitellum, n = 10).

Lethal Concentration	Chlorpyrifos (μg/cm^2)	95% Confidence limits Lower	Upper
LC_1	0.095	0.033	0.123
LC_5	0.112	0.051	0.137
LC_{10}	0.122	0.064	0.145
LC_{15}	0.129	0.074	0.151
LC_{50}	0.165	0.134	0.186
LC_{85}	0.211	0.187	0.296
LC_{90}	0.223	0.196	0.342
LC_{95}	0.243	0.209	0.425
LC_{99}	0.286	0.233	0.646
Slope ± SEM	9.74 ± 3.10		
Intercept ± SEM	12.62 ± 2.37		
X^2 value	3.02		
P	< 0.05		

Table 2
Lethal concentrations (LC_{1-99}) of cypermethrin for earthworm, *Eudrilus eugeniae* (identified by well-defined clitellum, n = 10).

Lethal Concentration	Cypermethrin (μg/cm^2)	95% Confidence limits Lower	Upper
LC_1	0.037	0.019	0.048
LC_5	0.044	0.027	0.054
LC_{10}	0.048	0.032	0.057
LC_{15}	0.051	0.035	0.060
LC_{50}	0.066	0.056	0.074
LC_{85}	0.086	0.077	0.104
LC_{90}	0.091	0.082	0.115
LC_{95}	0.100	0.088	0.135
LC_{99}	0.118	0.100	0.185
Slope ± SEM	9.29 ± 2.28		
Intercept ± SEM	15.94 ± 2.61		
X^2 value	1.48		
P	< 0.05		

Table 3
Lethal concentrations (LC_{1-99}) of combined pesticide (chlorpyrifos + cypermethrin) for earthworm, *Eudrilus eugeniae* (identified by well-defined clitellum, n = 10).

Lethal Concentration	Combination pesticide[a] (μg/cm^2)	95% Confidence limits Lower	Upper
LC_1	0.004	0.001	0.007
LC_5	0.006	0.002	0.010
LC_{10}	0.008	0.004	0.012
LC_{15}	0.010	0.005	0.013
LC_{50}	0.020	0.015	0.025
LC_{85}	0.040	0.030	0.072
LC_{90}	0.047	0.035	0.096
LC_{95}	0.061	0.042	0.147
LC_{99}	0.097	0.059	0.331
Slope ± SEM	3.35 ± 0.747		
Intercept ± SEM	10.72 ± 1.27		
X^2 value	4.43		
P	< 0.05		

TESTOVI TOKSIČNOSTI TLA

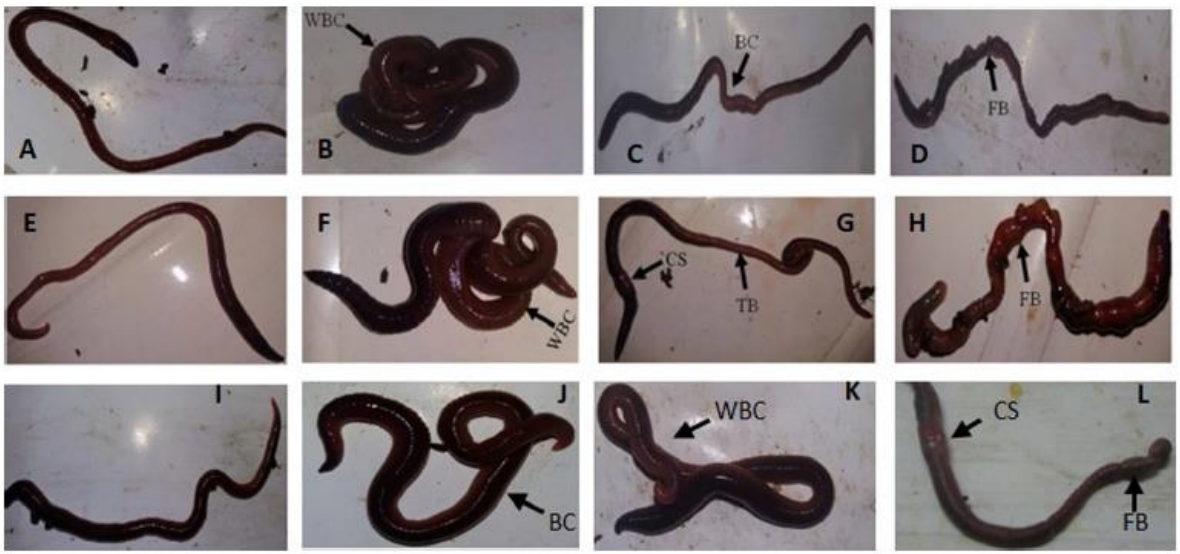


Fig. 1. Morphological changes in earthworm *E. eugeniae* exposed to different concentrations of chlorpyrifos, cypermethrin and their combination (A–L). A: Control and B: 0.112 μg/cm^2, C: 0.196 μg/cm^2, D: 0.224 μg/cm^2 concentrations of chlorpyrifos; E: Control and F: 0.0168 μg/cm^2, G: 0.0279 μg/cm^2, H: 0.0391 μg/cm^2 concentrations of cypermethrin and I: Control and J: 0.055 μg/cm^2 K: 0.075 μg/cm^2 L: 0.095 μg/cm^2 concentrations of their combination (chlorpyrifos + cypermethrin). WBC: Whole Body Coiling, BC: Body Constriction, FB: Fragmentation of Body, TB: Thinning of Body, CS: Clitellar Shrinkage, BC: Body coiling.

TESTOVI TOKSIČNOSTI TLA

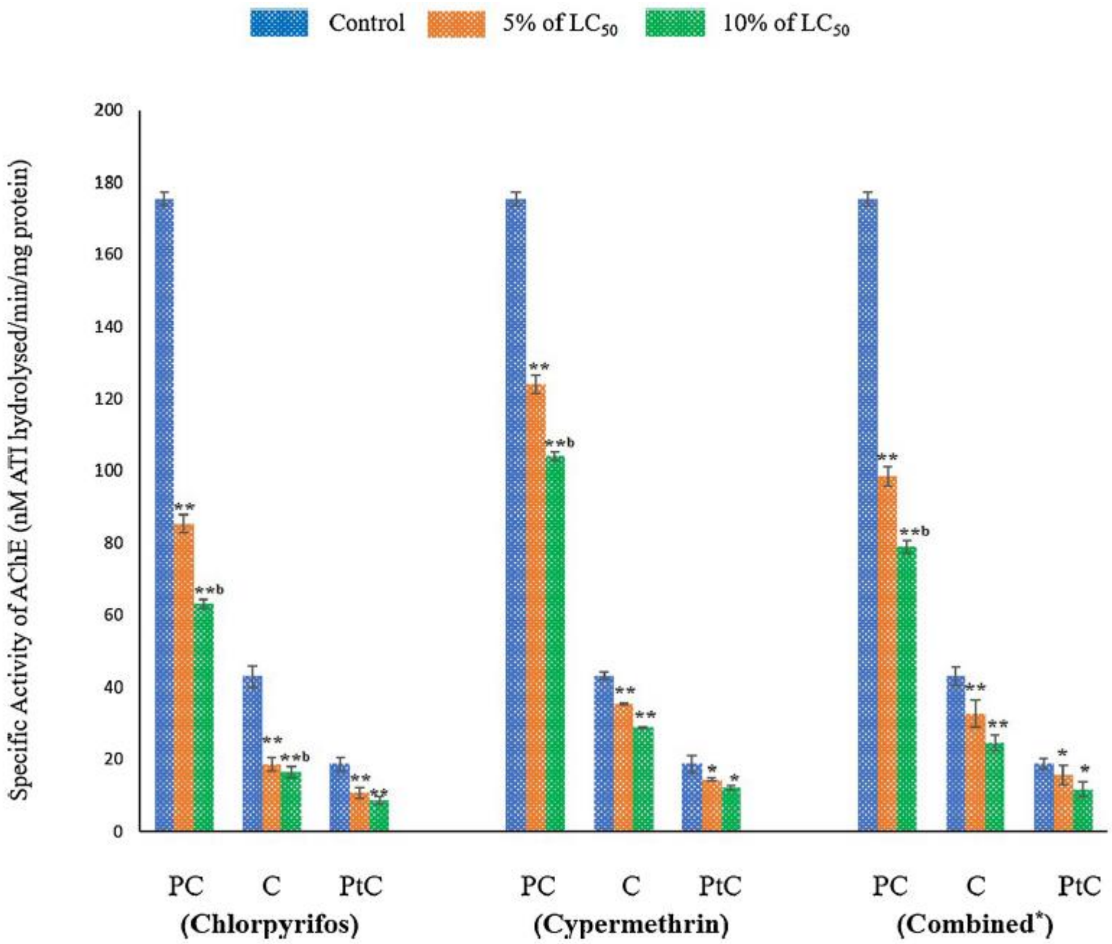
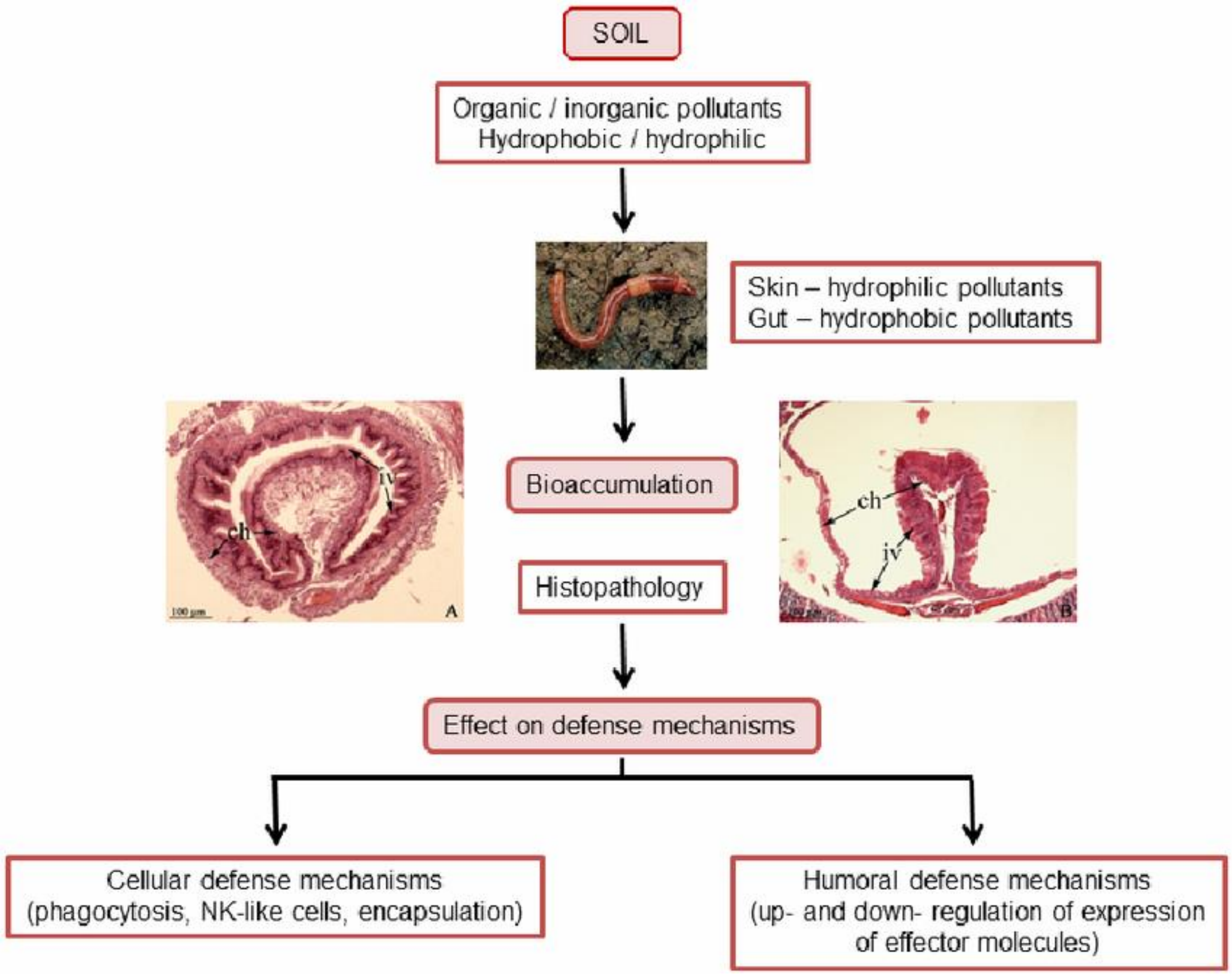


Fig. 2. For chlorpyrifos treated group, data represents mean ± SEM. ** $p < 0.01$ control *vs* 5% (8.4 ng/cm^2) and 10% (16.8 ng/cm^2.) of LC_{50}. For cypermethrin exposed group, data represents mean ± SEM. * $p < 0.05$ control *vs* 5% (1.0 ng/cm^2) and 10% (2.0 ng/cm^2) of LC_{50}, ** $p < 0.01$ control *vs* 5% and 10% of LC_{50} and for the earthworms exposed to combination of pesticide, data represents mean ± SEM * $p < 0.05$ control *vs* 5% and 10% of LC_{50}, ** $p < 0.01$ control *vs* 5% (3.3 ng/cm^2) and 10% (6.6 ng/cm^2) of LC_{50}. Here, PC: Pre-clitellar, C: Clitellar and PtC: Post- clitellar regions of earthworm.

TESTOVI TOKSIČNOSTI TLA



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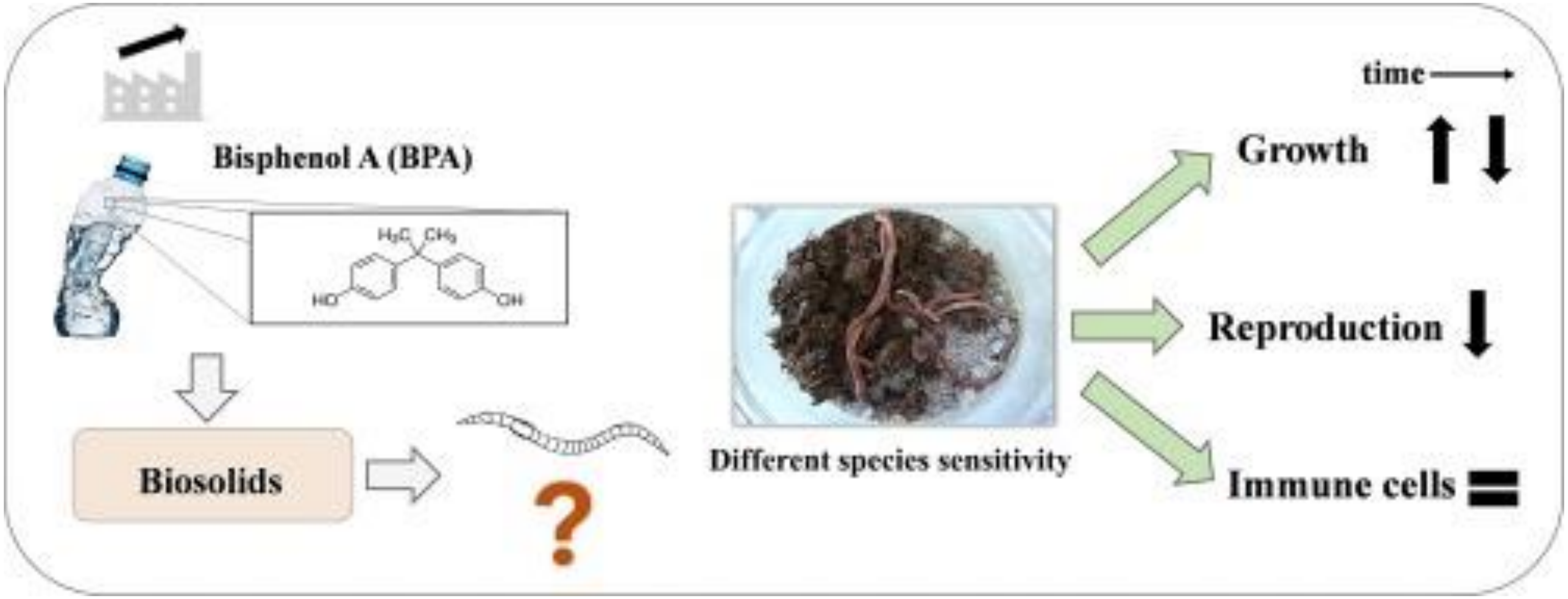


TESTOVI TOKSIČNOSTI TLA

Standardized Soil Toxicity Tests: Invertebrates

The following tests are reviewed in this section:

- Earthworm Acute Toxicity Tests
- Earthworm Sub-chronic Toxicity Test
- Soil toxicity or Bioaccumulation Tests with the Lumbricid Earthworm, Eisenia fetida
- Earthworm Reproduction Test (Eisenia fetida/andrei)
- Enchytraeidae Reproduction Test

Primjer: Eco-toxicity of petroleum hydrocarbon (TPH) contaminated soil

- earthworm acute toxicity
- plant growth experiment
- luminescent bacteria test

3 % TPH – nakon 7 dana sve gliste su umrle

2 % TPH – 90 % glista umrlo

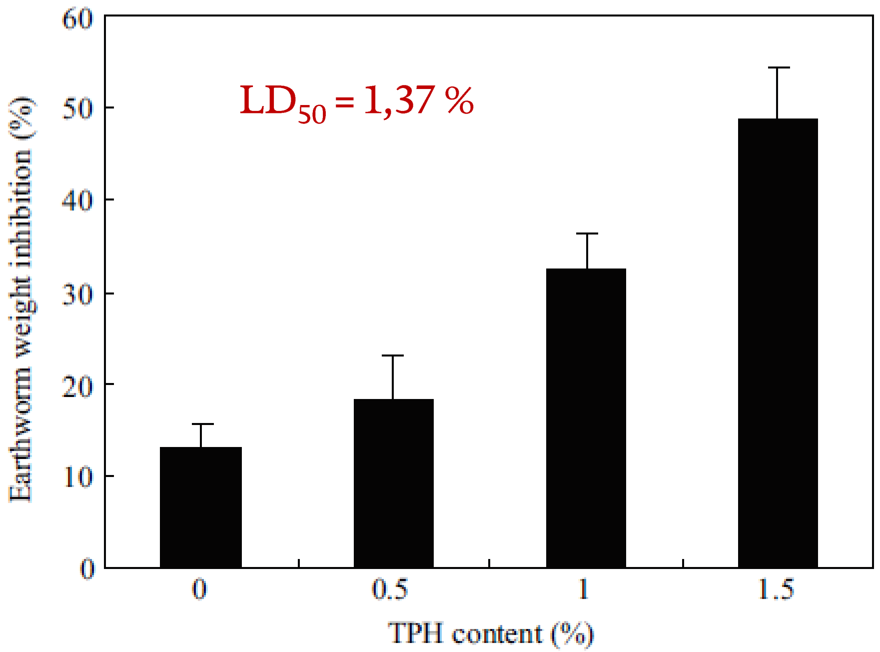


Fig. 2 Inhibition rate of earthworm body weight for 7 days in different TPH contaminated soils.

Primjer: Eco-toxicity of petroleum hydrocarbon (TPH) contaminated soil

Table 2 Comparison of seed germination of five different plant species after 4 and 10 days in TPH contaminated soil (%)

Plant	4 days germination	4 days control	10 days germination	10 days control
Wheat	20	95	60	100
Maize	10	90	45	100
Cotton	0	0	0	10
Mexican corn grass	0	30	0	75
Tall fescue	0	95	10	100

Najtolerantniji na TPH

Primjer: Eco-toxicity of petroleum hydrocarbon (TPH) contaminated soil

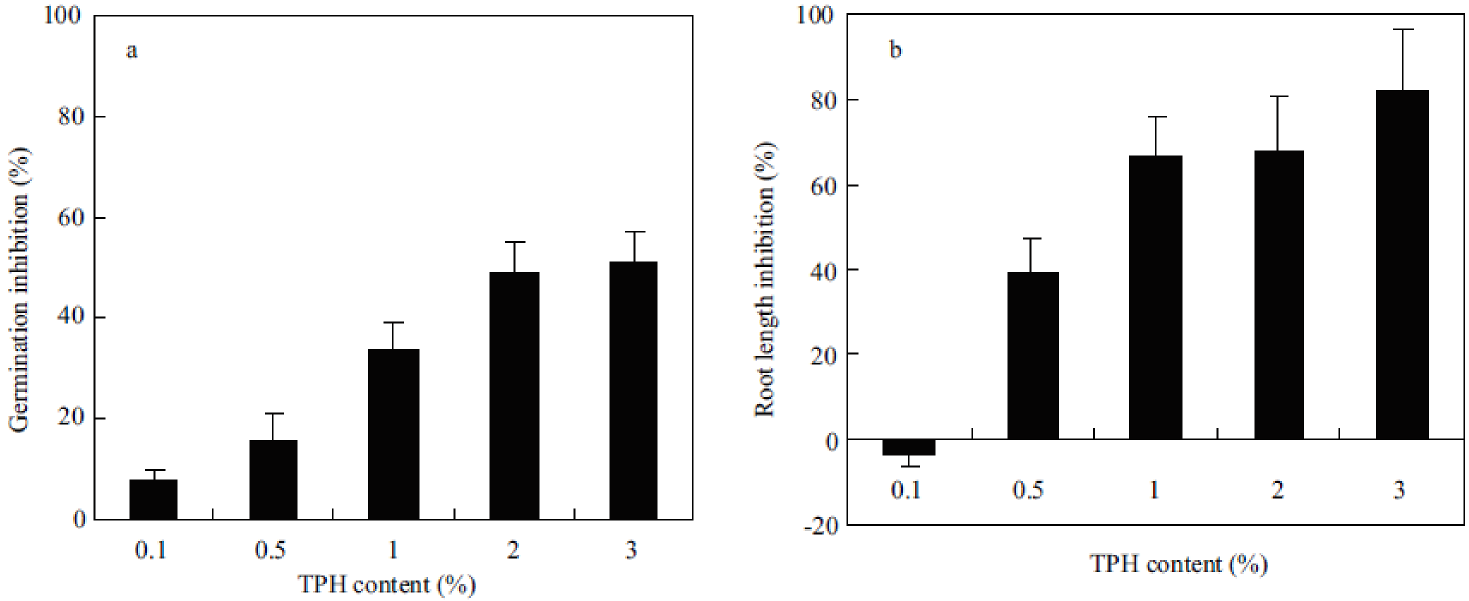


Fig. 3 Average inhibition rate of maize germination (a) and root elongation (b) in 72 hr by different TPH contaminated soils.

The EC_{50} (effective concentration 50%) values on germination and root elongation for maize are 3.04% and 1.11%, respectively, indicating a higher sensitivity of root elongation on TPH ccontamination in soil

Primjer: Eco-toxicity of petroleum hydrocarbon (TPH) contaminated soil

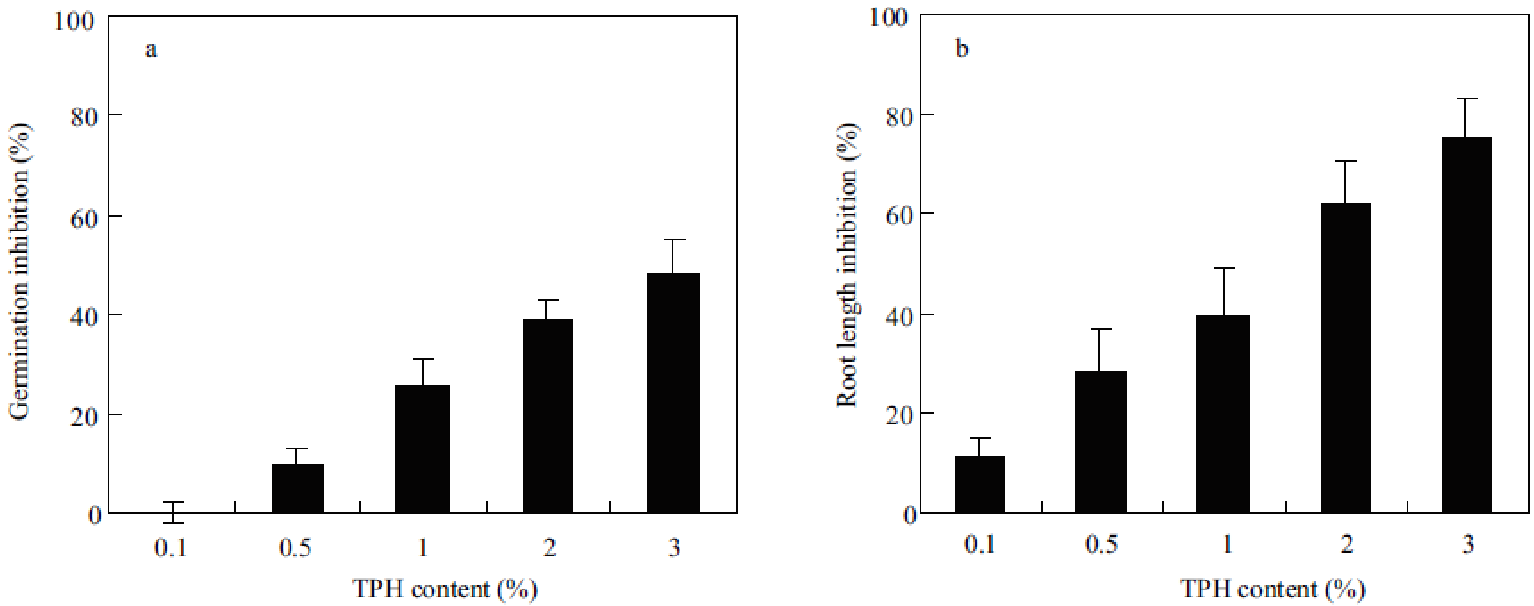


Fig. 4 Average inhibition rate of wheat germination (a) and root elongation (b) in 72 hr by different TPH contaminated soils.

The EC_{50} for germination and root elongation is 2.86% and 1.64%, respectively. As compared with maize, the wheat is more sensitive in germination and less sensitive in root elongation.

Primjer: Eco-toxicity of petroleum hydrocarbon (TPH) contaminated soil

- The EC_{50} value was 0.47% for the petroleum in the soil – *Vibrio fischeri*
- OSJETLJIVOST NA NAFTU:

luminescent bacteria activity > earthworm > plant growth (root elongation and germination)

root length > shoot elongation > germination rate

TESTOVI TOKSIČNOSTI TLA

Phytotoxicity Test: Early Seedling Growth Test	
Summary Description:	The test provides a standard procedure for measuring the toxicities of various chemical substances, industrial and domestic effluents, industrial and domestic sludges, as well as site soils. The test provides data as to whether the test substance or site soil either inhibits or enhances the growth of terrestrial plants. The first days of seedling growth are often the most sensitive stages of plant development. Separate groups of seeds/seedlings are exposed to different concentrations/percents of test substances for at least 21 days post-emergence. The study is terminated no later than 28 days post-planting, and seedling heights, above ground dry weights, and/or root lengths are determined. Visual assessment of plant condition also may be appropriate for determining phytotoxicity effects. A no effect soil concentration, based on the reference and experimental samples, is determined.
Source:	
Standardized:	Yes, published 1994
Reference:	[ASTM] American Society for Testing and Materials. 1994. Standard Practice for Conducting Early Seedling Growth Tests. Annual Book of Standards E 1598-94. West Conshohocken, PA. April, 1994.
Targeted Assessment:	Designed to investigate early life stage developmental toxicity. The first days of seedling growth are often the most sensitive stages of plant development.

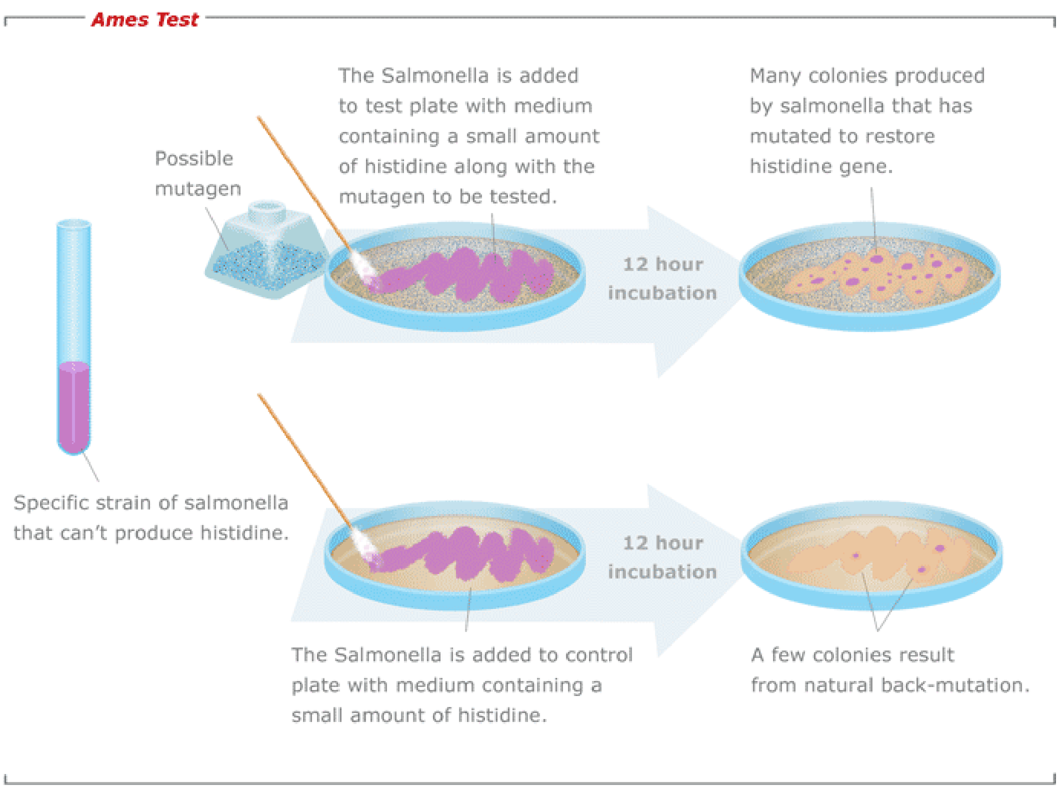
TESTOVI TOKSIČNOSTI TLA

- Dva testa toksičnosti tla pomoću mikroorganizma – standardizirana OECD (2000a, b): PRETVORBA DUŠIKA (MINERALIZACIJA DUŠIKA) i UGLJIKA (RESPIRACIJA TLA)
- Toksičnost na vodene organizme – koncentracija onečišćujuće tvari u tlu ne mora biti ista i u eluatu – adsorpcija na čestice, hlapljenje, razgradnja
- Ribe, dafnije, zelene alge
- Prednost *in vitro* testa toksičnosti tla na ribama – manji volumen eluata je potreban, smanjuje se upotreba kralješnjaka u testovima, brz odgovor, jednostavan test
- BESKRALJEŽNJACI – *Daphnia* - raznolikost u vodenom mediju, važna uloga u hranidbenom lancu (primarni konzumenti) – *Dapnia magna* ili *Daphnia pulex* – akutni i kronični test - OECD (1998a; 2004c) – IMOBILIZACIJA i REPRODUKTIVNOST
- Test toksičnosti na ALGAMA - ASTM, 1990; OECD, 2006b – standardizirani test
- Test rast alga – KRONIČNI TEST - visoka brzina rasta, učinci se mogu vidjeti na nekoliko generacija – *Pseudokirchneriella subcapitata, Desmodesmus subspicatus* i *Chlorella vulgaris*

TESTOVI TOKSIČNOSTI TLA

- Dva testa toksičnosti na bakterije (leaching test):

1. *Vibrio fischeri*
2. *Salmonella typhimurium* – mutagenost (AMES test)



TESTOVI TOKSIČNOSTI

Table 1. OCDE standardized tests for the determination of the toxicity of chemical substances to soil and aquatic organisms

Taxonomic group	Title	Endpoint	Measurement variables	Assay time (days)	Test Number
Terrestrial invertebrates	Earthworm, acute toxicity test	Survival	Number of living worms	14	207
Terrestrial invertebrates	Enchytraeid, reproduction test	Reproduction	Number of juvenile worms	42	220
Terrestrial invertebrates	Earthworm reproduction test	Reproduction	Number of living offspring and cocoon numbers	56	222
Plants	Terrestrial plant test: seedling emergence and seedling growth test	Emergence of seedlings and Inhibition of growth	Emergence, dry shoot weight (fresh weight), shoot weight and assessment of visible detrimental effects	14-21	208
Plants	Terrestrial plants test: vegetative vigor test	Vegetative vigor and growth	Biomass (dry shoot weight) and visible detrimental effects	21-28	227
Microorganisms	Soil microorganisms: Nitrogen transformation tests	Nitrogen transformation	Rate of nitrate production	28	216
Microorganisms	Soil microorganisms: Carbon transformation tests	Carbon transformation	Glucose-induced respiration rates	28	217
Algae Cyanobacteria	Freshwater Alga and Cyanobacteria, Growth inhibition test	Inhibition of growth	Algal biomass: cell counts, cell volume, fluorescence, optical density, etc.	4	201
Aquatic Invertebrates	Daphnia s.p., Acute immobilization test	Survival	Immobilization	1	202
Aquatic Invertebrates	Daphnia magna, reproduction test	Reproduction	Number of living offspring	21	211
Fish	Fish, Acute toxicity test	Survival	Number of living fish	4	203
Fish	Fish, Prolonged toxicity test: 14-day study	Survival, appearance and behavior, and growth	Survival, abnormalities (appearance and behavior), length and weight	14	204

Table 1. (Continued)

Taxonomic group	Title	Endpoint	Measurement variables	Assay time (days)	Test Number
Fish	Fish, early-life stage toxicity test	Survival, appearance and behavior, and growth	Hatching and survival (at different stages), abnormalities (appearance and behavior), length and weight	30-60	210
Fish	Fish, short-term toxicity test on embryo and sac-fry stages	Survival, appearance and behavior, and growth	Hatching and survival (at different stages), abnormalities (appearance and behavior), length and weight	8-55	212
Fish	Fish, juvenile growth test	Inhibition of growth	Weight	≥ 28	215
Aquatic plants	Lemna, sp. Growth inhibition test	Inhibition of growth	Frond number, total frond area, dry weight or fresh weight	7-10	221

TESTOVI TOKSIČNOSTI_tlo _ mikroorganizmi

Table 1-2. Standardized soil testing protocols and guidelines for microorganisms

Species / Duration	Test Guideline Title	Test Organism	Life Stage	Test Duration	Endpoint	Reference
Microorganism						
Acute						
	Soil Microorganisms: Carbon Transformation Test	Soil microbes	N/A	28-100 days	Rate of respiration [mean carbon dioxide released (mg carbon dioxide/kg dry weight soil/h) or mean oxygen consumed (mg oxygen/dry weight soil/h)].	**[OECD] Organization for Economic Cooperation and Development. 2000. OECD Guideline For The Testing Of Chemicals: Soil Microorganisms: Carbon Transformation Test. No. 217. Paris, France. 21 January 2000.**
	Soil Microorganisms: Nitrogen Transformation Test	Soil microbes	N/A	28-100 days	Nitrate production (mg nitrate/kg dry weight soil/day).	[OECD] Organization for Economic Cooperation and Development. 2000b. OECD Guideline For The Testing Of Chemicals: Soil Microorganisms: Carbon Transformation Test. No. 218. Paris, France. 21 January 2000
	Luminescence Bioassay	*Photobacterium phosphoreum* Strain NRRL B-11177	N/A	Approx. 1 hour	Quantitative reduction in light output of luminescent marine bacteria (i.e., IC20 or the calculated concentration of sample that would produce a 20% reduction in the light output of exposed bacteria over a specified time)	[ASTM] American Society for Testing and Materials. 1996. Standard Guide for Assessing the Microbial Detoxification of Chemically Contaminated Water and Soil Using a Toxicity Test with a Luminescent Marine Bacterium. Annual Book of Standards D 5660-96. West Conshohocken, PA. May 1996.
	Soil Microbial Community Toxicity Test	Soil microbes	N/A	28 days	Ammonification and nitrification (measured as NH_3 and NO_3 concentration per gram of soil, respectively) and respiration (CO_2) efflux	[EPA] US Environmental Protection Agency. 1987. Soil Microbial Community Toxicity Test. EPA 40 CFR Part 797.3700. Toxic Substance Control Act Test Guidelines; Proposed rule. 28 September 1987.

(Continued)

Table 1-2. Continued.

Species / Duration	Test Guideline Title	Test Organism	Life Stage	Test Duration	Endpoint	Reference
Microorganism						
Chronic						
	Soil-Core Microcosm Test	Soil microbes	N/A	12 weeks or longer	Effect of chemicals on 1) growth and reproduction of either naturally occurring vegetation or crop(s) of interest, 2) nutrient uptake and cycling within the soil/plant system, 3) potential bioaccumulation (enrichment) of test material into plant tissue and 4) the potential for and rate of transport of the chemical through soil to ground water	[EPA] US Environmental Protection Agency. 1996. Ecological Effects Test Guideline OPPTS 850.2450 Terrestrial (Soil-Core) microcosm Test. EPA 712-C-96-143, April 1996 [ASTM] American Society for Testing and Materials. 1987. Standard Guide for Conducting a Terrestrial Soil-Core Microcosm Test. Annual Book of Standards E 1197-87. West Conshohocken, PA. November 1987.

TESTOVI TOKSIČNOSTI_tlo _ beskralježnjaci

Table 1-5. Handbook of Soil Invertebrate Toxicity Tests

Species	Test Descriptions
Enchytraeid: Oligochaeta	
Cognettia sphagnetorum	Sublethal toxicity test
Annelida: Oligochaeta	
Eisenia fetida	Sublethal toxicity test
Aporrectoda calignosa	Sublethal toxicity test
Acari: Oribatida (Orbatid mite)	
Platynosthrus peltifer	Sublethal toxicity test in soil
Platynosthrus peltifer	Sublethal toxicity test on plaster of Paris with dietary exposure
Collembola: Isotomidae (Springtails)	
Folsomia candida	Sublethal toxicity test: growth and reproduction
Folsomia fimetaria	Sublethal toxicity test
Isotoma viridis	Sublethal toxicity test: growth
Coleoptera: Staphylinidae (Beetles)	
Philonthus cognatus	Sublethal toxicity test – mature beetles
Philonthus cognatus	Sublethal toxicity test – larval stage beetles
Philonthus cognatus	Semi-field test – larval stage beetles
Myriapoda: Chilopoda (Centipedes)	
Lithobius mutabilis	Sublethal toxicity test

TESTOVI TOKSIČNOSTI_tlo _ beskralježnjaci

Diplopoda: Polydesmidae (Millipedes)	
Brachydesmus superus	Sublethal toxicity test
Isopoda: Porcellionidae (Woodlouse)	
Porcellio scaber	Sublethal toxicity test: growth
Porcellio scaber	Sublethal toxicity test: reproduction
Nematoda: Cephalobidae (Nematodes)	
Plectus acuminatus and *Heterocephalobus pauciannulatus*	Sublethal toxicity test: Competition between nematode species
Acari: Gamasida (Gamasid mite)	
Hypoaspis aculeifer	Sublethal toxicity test: Predation of a gamasid mite on the collembolan *Folsomia fimetaria*

(Løkke and van Gestel 1998)

TESTOVI TOKSIČNOSTI_tlo _ beskralježnjaci

Table 1-6. Standardized soil testing protocols and guidelines for invertebrates

Species / Duration	Test Guideline Title	Test Organism	Life Stage	Test Duration	Endpoint	Reference
Earthworms						
Acute						
	Earthworm, Acute Toxicity Tests	*Eisenia fetida*	Adult	***Filter Paper Test:*** **48-72 hours** *Artificial Soil Test*: 14 days	Mortality and other noted abnormal behaviors	[OECD] Organization for Economic Cooperation and Development. 1984. OECD Guideline For The Testing Of Chemicals: Earthworm, Acute Toxicity Test. No. 207. Paris, France. April 1984.
	Toxicity in Earthworms					[EEC] European Economic Community. 1985. Directive79/8331, V, Part C: Methods for the Determination of Ecotoxicity – Level 1. DG XI/127-129/82, Rev. 1: Toxicity for Earthworms. Commission of the European Community, Brussels.

(Continued)

TESTOVI TOKSIČNOSTI_tlo _ beskralježnjaci

Table 1-6. Continued.

Species / Duration	Test Guideline Title	Test Organism	Life Stage	Test Duration	Endpoint	Reference
Earthworms						
Subchronic						
	Earthworm Subchronic Toxicity Test	*Eisenia fetida*	Adult	28 days	Mortality and other noted abnormal behaviors, pathological conditions, or weight loss	[EPA] US Environmental Protection Agency. 1996. Ecological Effects Test Guideline OPPTS 850.6200 Earthworm Subchronic Toxicity Test. EPA 712-C-96-167, April 1996
	Soil toxicity or Bioaccumulation Tests with the Lumbricid Earthworm *Eisenia fetida*	*Eisenia fetida*	Adult	14-28 days	Endpoints are dependent on purpose of the test and study design but may include animal weight, lethality, sublethal behaviors, pathological changes (segmental constriction, lesions, stiffness, etc.), reproduction, tissue accumulation, etc. Other endpoint analysis may include kinetic studies with estimate uptake, depuration rates, and time to steady state, lipid normalization and normalizing soil concentrations of non-ionic organics to total organic carbon. Reproductive endpoints might include number and growth of young worms, rate of clitellum development, number of cocoons produced, cocoon mass, number of hatchlings per cocoon, and biomass of hatchlings.	[ASTM] American Society for Testing and Materials. 1998. Standard Guide for Conducting Laboratory Soil Toxicity or Bioaccumulation Tests with the Lumbricid Earthworm *Eisenia fetida*. Annual Book of Standards E 1676-97. West Conshohocken, PA. February 1998.

(Continued)

TESTOVI TOKSIČNOSTI_tlo _ beskralježnjaci

Table 1-6. Continued.

Species / Duration	Test Guideline Title	Test Organism	Life Stage	Test Duration	Endpoint	Reference
Earthworm						
Chronic						
	Earthworm Reproduction Test (*Eisenia fetida/andrei*)	*Eisenia fetida/andrei*	Adult	8 weeks observation Adult earthworms are removed after the fourth week	Adult mortality and other signs of toxicity, reproductive success as measured by number of juveniles produced.	[OECD] Organization for Economic Cooperation and Development. 2004. OECD Guideline For The Testing Of Chemicals: Test No. 222, Earthworm Reproduction Test (*Eisenia fetida/andrei*). Paris, France.

TESTOVI TOKSIČNOSTI_tlo _ kralježnjaci

Table 1-7. Standardized Vertebrate Toxicity Tests

TEST NAME	Source of Test Guideline: US EPA OPPTS Number[1]	OECD Number	ASTM Number
Ecological Effects - Terrestrial Wildlife Test Guidelines			
Avian acute oral toxicity test	850.2100		E555-92
Avian dietary toxicity test	850.2200	205	E857-87 (1997)
Avian reproduction test	850.2300	206	E1062-86
Wild mammal acute toxicity	850.2400		
Field testing for terrestrial wildlife (draft)	850-2500		
Health Effects – Acute Toxicity Test Guidelines			
Acute toxicity testing-background	870.1000	420, 423, 425	
Acute oral toxicity	870.1100	401	E1163.98
Acute dermal toxicity	870.1200	402	
Acute inhalation toxicity	870.1300	403	
Acute inhalation toxicity with histopathology	870.1350		

TESTOVI TOKSIČNOSTI_tlo _ kralježnjaci

Health Effects – Subchronic Toxicity Test Guidelines

90-Day oral toxicity	870.3100	408	E1372-95 (1999)
Subchronic nonrodent oral toxicity 90-day	870.3150	409	
Repeated dose dermal toxicity--21/28 days	870.3200	410	
Subchronic dermal toxicity--90 days	870.3250	411	E1103-96 (2000)
Subchronic inhalation toxicity	870.3465	413	E1373-92 (1996)
Preliminary developmental toxicity screen	870.3500		

(Continued)

Table 1-7. Continued

TEST NAME	Source of Test Guideline: US EPA OPPTS Number[1]	OECD Number	ASTM Number
Inhalation developmental toxicity study	870.3600		
Prenatal developmental toxicity study	870.3700	414	E1483-92 (2000)
Reproduction and fertility effects	870.3800	416	
Health Effects – Chronic Toxicity Test Guidelines			
Chronic toxicity	870.4100	452	E1619-95 (1999)
Carcinogenicity	870.4200	451	
Combined chronic toxicity carcinogenicity	870.4300	453	E1811-96
Health Effects – Genetic Toxicity Test Guidelines			
Mouse biochemical specific locus test	870.5195		
Mouse visible specific locus test	870.5200		
Detection of gene mutations in somatic cells in culture	870.5300	476	
Standard guide for performing the mouse lymphoma assay for mammalian cell mutagenicity			E1280-97
Standard guide for performance of the Chinese hamster ovary cell/hypoxanthine guanine phosphoribosyl transferase mutation assay			E1262-88 (1996)
In vitro mammalian cytogenetics	870.5375	473	
In vivo mammalian cytogenetics tests: spermatogonial chromosomal aberrations	870.5380		
In vivo mammalian cytogenetics tests: Bone marrow chromosomal analysis	870.5385	475	
In vivo mammalian cytogenetics tests: Erythrocyte micronucleus assay	870.5395	474	
Rodent dominant lethal assay	870.5450	478	

(Continued)

Table 1-7. Continued

TEST NAME	Source of Test Guideline: US EPA OPPTS Number[1]	OECD Number	ASTM Number
Rodent heritable translocation assays	870.5460	485	
Unscheduled DNA synthesis in mammalian cells in culture	870.5550	482	
In vitro sister chromatid exchange assay	870.5900	479	
In vivo sister chromatid exchange assay	870.5915		
Health Effects – Neurotoxicity Test Guidelines			
Delayed neurotoxicity of organophosphorus substances following acute and 28-day exposure	870.6100	418, 419	
Neurotoxicity screening battery	870.6200		
Developmental neurotoxicity study	870.6300		
Schedule-controlled operant behavior	870.6500		
Peripheral nerve function	870.6850		
Neurophysiology: Sensory evoked potentials	870.6855		
Health Effects – Special Studies Test Guidelines			
Domestic animal safety	870.7200		
Metabolism and pharmacokinetics	870.7485	417	
Dermal penetration	870.7600		
Immunotoxicity	870.7800		
Pharmacokinetic test	870.8223		
Dermal pharmacokinetics of DGBE and DGBA	870 8245		
Oral/dermal pharmacokinetics	870.8320		

(Continued)

TESTOVI TOKSIČNOSTI_zaštita bilja_pesticidi _ kralježnjaci_ptice

- AKUTNA TOKSIČNOST tvari za:

1. PREPELICE (japanska prepelica – *Coturnix coturnix japonica* ili virdžinijska prepelica – *Colinus virginianus*) ili
2. DIVLJU PATKU (*Anas platyrhynchos*)

- Najveća doza pri testiranju ne bi trebala biti veća od 2 000 mg/kg tjelesne težine

- MOŽE SE ODREDITI:
  - LD_{50} vrijednost,
  - Najniža smrtna doza (LLC),
  - Vrijeme učinka i oporavka
  - NOEL
  - Makropatološki nalazi

Coturnix coturnix japonica

Colinus virginianus

Anas platyrhynchos

Smjernice za istraživanje Setac – Postupci za procjenu utjecaja pesticida na okoliš i ekotoksičnosti pesticida

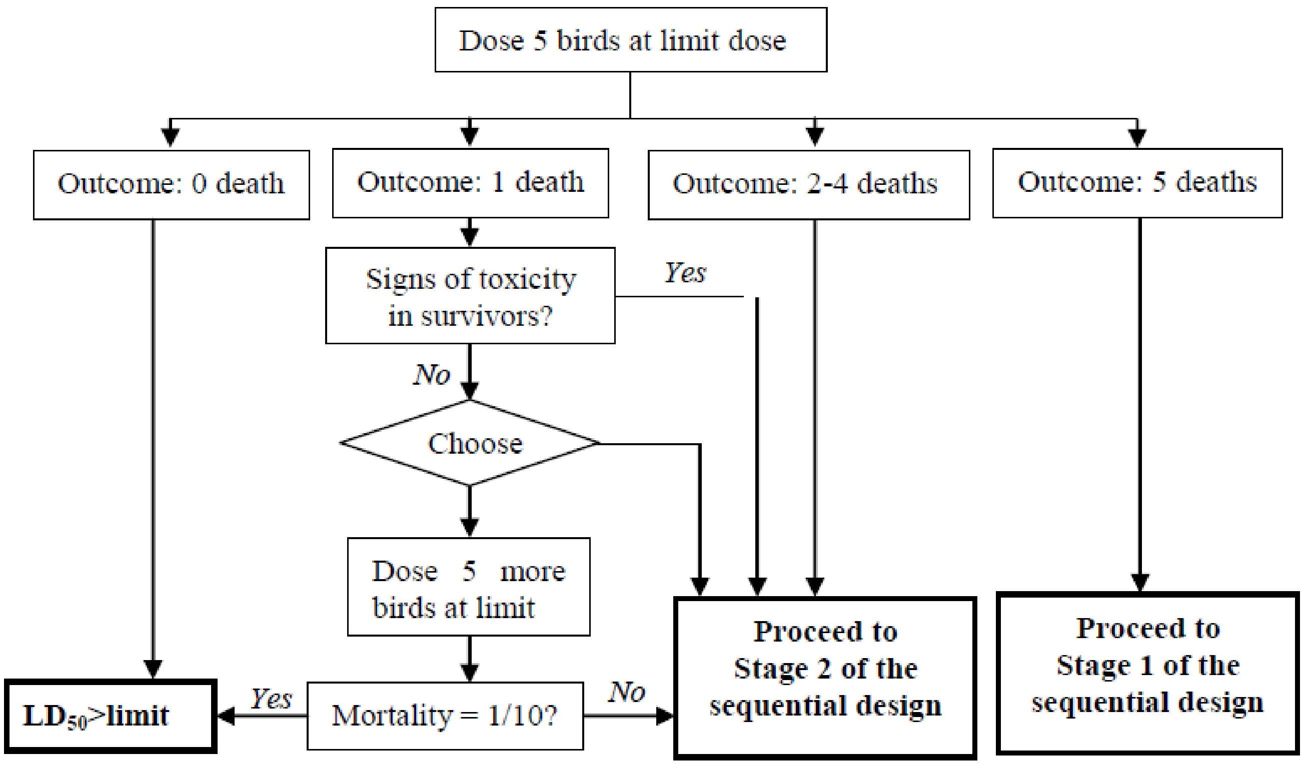
TESTOVI TOKSIČNOSTI_zaštita

bilja_pesticidi _ kralježnjaci_ptice

- KRATKOTRAJNA ORALNA TOKSIČNOST
- Test bi trebao dati podatke o:
- kratkoročnoj oralnoj toksičnosti (vrijednosti LC 50 , najnižoj smrtnoj koncentracija (LLC), ako je moguće koncentraciji bez učinaka (NOEC), vremenu odgovora i oporavka) i
- uključivati relevantne makropatološke nalaze
- PETODNEVNO ISTRAŽIVANJE toksičnosti za ptice pri unosu aktivne tvari (pesticida) s hranom,
- Ako je akutni oralni NOEL ≤ 500 mg/kg tjelesne težine ili ako je kratkotrajni NOEC < 500 mg/kg hrane - TEST SE MORA OBAVITI NA JOŠ JEDNOJ VRSTI - ako se mora testirati druga vrsta, ona NE BI SMJELA BITI SRODNA PRVOJ TESTIRANOJ VRSTI

TESTOVI TOKSIČNOSTI_tlo _ kralježnjaci_ptice

Figure 1: Limit dose test procedure; figure does not include control birds

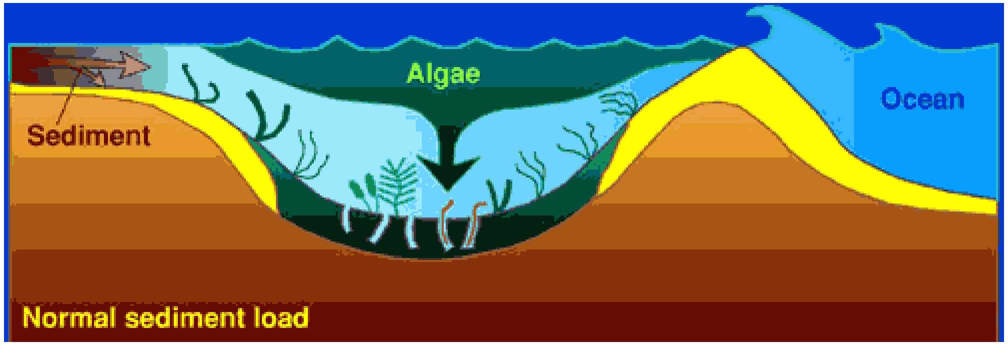


TESTOVI TOKSIČNOSTI_tlo _ kralježnjaci_ptice

Toxicity Test: Avian Acute Oral Toxicity Test	
Summary Description:	This test is one of a series of tests originally designed to assess the toxicity of pesticides and toxic substances to birds. In the US, this test is designed to meet the data requirements of the US EPA under the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA) and Toxic Substance Control Act (TSCA). The purpose of this test is to develop data on the acute oral toxicity to northern bobwhite and mallard of chemical substances and mixtures of interest to assess the acute hazard of these chemicals to birds. Following a 14-day acclimation period, adult birds are gavaged with a single oral dose of the test material in a range of doses. Birds are closely monitored for signs of intoxication for at least 14 days or until all signs of intoxication are not observed for 72 hours. Bird weight and food consumption are monitored at least weekly. The mortality pattern is examined and the LD50, confidence limits, and slope of the dose-response line are determined through appropriate statistical analysis. Signs of intoxication, bird weights, food consumption, necropsy results and the mortality results are reported.
Source:	
Standardized:	Yes
Reference:	[EPA] US Environmental Protection Agency. 1996. Avian Acute Oral Toxicity Test, OPPTS 850.2100, EPA 712-C-96-139. Washington, DC. April 1996. See also [EPA] US Environmental Protection Agency. 1998b. Acute Oral Toxicity, OPPTS 870.1100 Acute Oral Toxicity, EPA 712-C-98-190. Washington, DC. August 1998. [ASTM] American Society for Testing and Materials. 1995. Standard Practice for Determining Acute Oral LD50 for Testing Vertebrate Control Agents. Annual Book of Standards E 555-95. West Conshohocken, PA.
Targeted Assessment:	Designed to investigate the acute hazard of pesticides and toxic chemicals to birds by establishing the LD50, the empirically derived dose of the test substance that is expected to result in mortality of 50 percent of the birds treated with a single oral dose under the test conditions.

TOKSIČNOST SEDIMENTA

- SEDIMENT – „skladište” za onečišćujuće tvari (porijeklom iz industrije, poljoprivrede, komunalnih voda)
- Sediment – bitna komponenta vodenih ekosustava, STANIŠTE RAZLIČITIH BENTONSKIH VRSTA
- BENTON- skupni je naziv za sve BILJNE I ŽIVOTINJSKE ORGANIZME KOJI ŽIVE NA DNU MORA ili slatke vode
- Organizmi koji žive bentoski VRLO SU VAŽNA HRANA ZA RIBE i druge životinje koje žive u otvorenim vodama, za nekton, ali i za organizme koji razgrađuju organske tvari
- Prijenos onečišćujućih tvari putem HRANIDBENOG LANCA



TOKSIČNOST SEDIMENTA

- Testovi toksičnosti sedimenta – procjenjuju stupanj toksičnosti sedimenta prema organizmima koji se nalaze na dnu
- Stupanj toksičnosti se može zaključiti iz – KEMIJSKIH ANALIZA – određivanje koncentracija onečišćujućih tvari – daje nam podatak o KVALITETI SEDIMENTA – velika ili mala vjerojatnost da sediment može imati potencijalno štetne učinke na organizme
- PROBLEM: KEMIJSKIM ANALIZAMA se NE MOGU PROCIJENITI potencijalni štetni učinci mješavina onečišćujućih tvari i utjecaj ostalih fizikalnih karakteristika sedimenta (poput veličine čestica) koji mogu stupiti u interakciju i pri tome pridonijeti štetnim učincima
- TESTOVIMA TOKSIČNOSTI SEDIMENTA – mogu se procijeniti potencijalni štetni učinci onečišćujućih tvari, mješavina onečišćujućih tvari te ostalih karakteristika sedimenta – DIREKTNO UTJEČU NA ISPITIVANU VRSTU

TOKSIČNOST SEDIMENTA

- PRIJE PROVEDBE TESTA TOKSIČNOSTI MORA SE UZETI U OBZIR:
  - Broj i vrsta organizma
  - Osjetljivost i trajanje testa
  - Broj ponavljanja testa ili broj koncentracija izloženosti
  - Utjecaj fizikalno-kemijskih parametra na provedbu testa
  - Potreba za unosom hrane, utjecaj dodatka hrane na ponašanje organizma i unos onečišćujuće tvari
  - Statični ili dinamični testovi
  - Da li se koncentracija odnosno bioraspoloživost onečišćujuće tvari mijenja s vremenom (hlapiva organska onečišćenja)

TOKSIČNOST SEDIMENTA

- ODABIR TESTNOG ORGANIZMA:
  - Organizam koji ima kontakt sa sedimentom
  - Osjetljivost na različite onečišćujuće tvari
  - Tolerantan na fizikalno-kemijske karakteristike sedimenta
  - Standardizirana metoda
  - Dostupna vrsta
  - Lako se održava u laboratoriju
  - Lako se identificira
  - Kratak životni ciklus (dani do tjedana) – kratki testovi

Koncentracija onečišćujućih tvari u sedimentu 100 do 10 000 puta nego u površinskim i podzemnim vodama

TOKSIČNOST SEDIMENTA

- Ne postoji organizam koji je osjetljiv na sve onečišćujuće tvari – preporuka koristiti više vrsta testova (različiti organizmi (rod))
- Neke vrste su neosjetljive na mnoge onečišćujuće tvari – morski crv (*Nereis virens*) i škamp (*Artemia* sp.)
- Ovi organizmi se koriste za druge testove – npr. TESTOVI BIOAKUMULACIJE (USEPA, 1993) – zbog velike veličine i zbog sposobnosti da žive u onečišćenom sedimentu
- Bakterija – *V. fischeri* – problem – mutnoća sedimenta
- Alge – prati se učinak na obavljanje fotosinteze, ne na rast – na rast utječe amonijak i koncentracija nutrijenta koji se nalaze u sedimentu

FIGURE 7.1 Whole-sediment toxicity test species: *(top, left to right)* amphipod, copepod, mysid, and crab (crustaceans); *(bottom, left to right)* benthic algae (plant), bivalve/clam (mollusk), polychaete worm (annelid), and snail (gastropod).

TABLE 7.1 Estuarine and Marine Whole-Sediment Toxicity Tests

Organism	Test species	Duration/Endpoint	Acute/Chronic	References
Bacteria	*Vibrio fischeri* (Microtox)	20-min luminescence	Acute	Azur Environmental (1998), Environment Canada (2002)
Protozoa	*Euplotes crassus*	Elutriate tests: 8-h cell viability; 24-h replication	Acute	Gomiero et al. (2013)
Miroalga	*Entomoneis* cf *punctulata*	24-h enzyme (esterase) inhibition	Acute	Adams and Stauber (2004), Adams (2016)
Microalga	*Cylindrotheca closterium* (formerly *Nitzschia closterium*)	72-h growth rate	Chronic	Moreno-Garrido et al. (2003a,b, 2007), Araujo et al. (2010)
Copepod	*Amphiascus tenuiremis* and *Microarthridion littorale*	14-day survival and reproduction; 21-day full life cycle	Chronic	Chandler and Green (1996), Kovatch et al. (1999), Kennedy et al. (2009)
Copepod	*Nitocra spinipes*	10-day reproduction	Chronic	Perez-Landa and Simpson (2011), Simpson and Spadaro (2011), Krull et al. (2014), Araujo et al. (2013), Spadaro and Simpson (2016a)
Copepod	*Robertsonia propinqua*	24-day life-cycle test	Chronic	Hack et al. (2008)
Copepod	*Tisbe biminiensis*	7-day reproduction	Chronic	Araujo et al. (2013)
Mysid	*Americamysis bahia*	10-day survival	Acute	Kennedy et al. (2009)
Amphipod	*Ampelisca brevicornis*	28-day survival, fecundity, and growth	Chronic	Costa et al. (1998, 2005)
Amphipod	*Corophium multisetosum*	10-day survival; 21-day fecundity and growth	Acute Chronic	Casado-Martinez et al. (2006), Castro et al. (2006)
Amphipod	*Corophium volutator*	28-day survival and growth; 28-day and 76-day survival, growth, and reproduction	Chronic	Scarlett et al. (2007a), Fox et al. (2014)
Amphipod	*Ampelisca brevicornis*, *Corophium volutator*, *Eohaustorius estuarius*, *Leptocheirus plumulosus*, *Rhepoxynius abronius*	10-day survival	Acute	Rodriguez-Romero et al. (2013), ASTM (2014), Greenstein et al. (2008)
Amphipod	*Gammarus locusta*	28-day survival, fecundity, and growth	Chronic	Costa et al. (1998, 2005)

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FKITMCMXIX

TABLE 7.1 Estuarine and Marine Whole-Sediment Toxicity Tests—cont'd

Organism	Test species	Duration/Endpoint	Acute/Chronic	References
Amphipod	*Hyalella azteca* (up to 15 ppt)	10-day and 28-day survival and growth; 42-day survival, growth, and reproduction	Chronic	ASTM (2010)
Amphipod	*Leptocheirus plumulosus*	28-day reproduction and growth	Chronic	ASTM (2008a), Kennedy et al. (2009)
Amphipod	*Melita plumulosa*	10-day juvenile survival	Acute	Spadaro et al. (2008), Strom et al. (2011)
Amphipod	*Melita plumulosa*	10-day reproduction	Chronic	Mann et al. (2009), Simpson and Spadaro (2011), Spadaro and Simpson (2016b)
Bivalve	*Mercenaria mercenaria*	7-day juvenile growth	Sublethal	Ringwood and Keppler (1998), Keppler and Ringwood (2002)
Bivalve	*Tellina deltoidalis*	10-day survival	Acute	King et al. (2010)
Bivalve	*Tellina deltoidalis*	30-day survival and growth	Chronic	Campana et al. (2013), Spadaro and Simpson (2016c)
Polychaete worm	*Arenicola marina*	10-day, 21-day survival	Acute	Bat and Raffaelli (1998), Morales-Caselles et al. (2008)
Polychaete worm	*Neanthes arenaceodentata*	20-day survival, 28-day growth	Chronic	Bridges and Farrar (1997), Farrar and Bridges (2011)
Polychaete worm	*Nereis virens*	7-day avoidance and damage to body condition	Acute	Van Geest et al. (2014a,b)
Mussel	*Mytilus galloprovincialis*	2-day embryo development at sediment–water interface	Sublethal	Anderson et al. (1996), Greenstein et al. (2008)
Snail	*Hydrobia ulvae*	48-h postexposure feeding 24-h avoidance	Sublethal	Krell et al. (2011), Araujo et al. (2012)

TOKSIČNOST SEDIMENTA


M. spicatum

Table 1

Overview of species for which relevant toxicity data were found.

Species	Taxonomic group	Environment
Ampelisca abdita	Amphipod, Crustacea	Estuarine
Amphiascus tenuiremis	Copepod, Crustacea	Estuarine
Asellus aquaticus	Isopod, Crustacea	Fresh
Chironomus dilutus (tentans)	Chironomid, Insecta	Fresh
Chironomus riparius	Chironomid, Insecta	Fresh
Corophium volutator	Amphipod, Crustacea	Marine
Eohaustorius estuarius	Amphipod, Crustacea	Estuarine
Ephoron virgo	Trichopteran; Insecta	Fresh
Hyalella azteca	Amphipod, Crustacea	Fresh / estuarine
Jappa kutera	Ephemeropteran, Insecta	Fresh
Leptocheirus plumosus	Amphipod, Crustacea	Estuarine
Lumbriculus variegatus	Worm, Oligochaeta	Fresh
Mercenaria mercenaria	Bivalve, Mollusca	Marine
Microarthridion littorale	Copepod, Crustacea	Estuarine
Myriophyllum aquaticum	Rooted macrophyte	Fresh
Myriophyllum spicatum	Rooted macrophyte	Fresh
Neanthes arenaceodentata	Worm, Polychaeta	Marine
Nereis diversicolor	Worm, Polychaeta	Marine
Nereis virens	Worm. Polychaeta	Marine
Potamopyrgus antipodarum	Snail, Mollusca	Fresh
Paronychocamptus wilsoni	Copepod, Crustacea	Marine
Tubifex tubifex	Worm, Oligochaeta	Fresh

Mekušac

M. mercenaria

Table 2

Acute and semi-chronic (duration ≤ 12 d) sediment toxicity data for organochlorine compounds.

Compound	Species	Effect endpoint	LC50/EC50/NOEC (µg/g OC) Mean (Range)	endpoint	References
Kepone	*C. dilutus*	Mortality	584 (293 – 1000)	10d LC50	Adams et al., 1985; Di Toro et al., 1991
Kepone	*C. dilutus*	Growth/ weight	495 (311 – 662)	10d EC50	Adams et al., 1985; Di Toro et al., 1991
DDT	*H. azteca*	Mortality	420 (367 – 473)	10d LC50	Nebeker et al., 1989
DDT	*H. azteca*	Mortality	121 (101 – 140)	10d LC50	Schuytema et al., 1989
DDT	*H. azteca*	Mortality	135 (128 – 142)	10d LC50	Amweg and Weston, 2007
DDT	*H. azteca*	Mortality	259 (221 – 296)	10d LC50	Weston et al., 2009
DDT	*M. mercenaria*	Mortality	829	10d LC50	Chung et al., 2007
DDD	*H. azteca*	Mortality Length	260 240	10d LC50 10d EC50	Ingersoll et al., 2005
Dicofol	*H. azteca* *C. dilutus* *C. dilutus*	Mortality Mortality Growth	>573[a] 915 630	10d LC50 10d LC50 10d NOEC	Ding et al., 2011
Dieldrin	*C. dilutus*	Mortality	57 (35-78)	10d LC50	US EPA 1993
Dieldrin	*H. azteca*	Mortality	2000 (1100-3700)	10d LC50	US EPA 1993
Endrin	*C. dilutus*	Mortality	4.22	10d LC50	Weston et al. 2004 You et al. 2004
Endrin	*H. azteca*	Mortality	100 (54 – 147)	10d LC50	Nebeker et al., 1989
Endrin	*H. azteca*	Mortality	153 (136 – 170)	10d LC50	Schuytema et al., 1989
Endosulfan (sulfate)	*C. dilutus*	Mortality	5.22	10d LC50	Weston et al. 2004 You et al. 2004
Endosulfan (sulfate)	*C. dilutus*	Mortality	100	10d LC50	Sappington, 2013
Endosulfan (sulfate)	*L. plumulosus*	Mortality	144	10d LC50	Sappington, 2013
Endosulfan (sulfate)	*H. azteca*	Mortality	873	10d LC50	Weston et al. 2004 You et al. 2004
Endosulfan (sulfate)	*J. kutera*	Mortality	324[b]	10d LC50	Leonard et al. 2001
Endosulfan (sulfate)	*N. virens*	Mortality	17	12-d LC50	McLeese et al., 1982
Alpha-endosulfan	*C. dilutus*	Mortality	0.96	10d LC50	Weston et al. 2004 You et al. 2004
Alpha-endosulfan	*H. azteca*	Mortality	51.7	10d LC50	Weston et al. 2004 You et al. 2004
Beta-endosulfan	*C. dilutus*	Mortality	3.24	10d LC50	Weston et al. 2004 You et al. 2004
Lindane	*T. tubifex*	Lethality Avoidance Autotomy	>1000 224 200	3d LC50 3d EC50 3d EC50	Meller et al., 1998
Alpha-chlordane	*H. azteca*	Lethality	516	10d LC50	Trimble et al., 2009
Gamma-chlordane	*H. azteca*	Lethality	889	10d LC50	Trimble et al., 2009
Methoxychlor	*C. dilutus* *H. azteca*	Mortality Mortality	36.7 85.8	10d LC50 10d LC50	Weston et al. 2004 You et al. 2004

[a] Range of values given as >1230, >274 and >215 µg/g OC for three sediments.

[b] Recalculated on basis of 95% moisture content and 1% OC content of sediment used.

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Table 3

Comparison of 10-12 d sediment toxicity data for organochlorine compounds between the amphipods Hyalella azteca and Leptocheirus plumulosus, the insects Chironomus dilutus and Jappa kutera, the polychaete Nereis virens and the mollusc Mercenaria mercenaria on basis of the endpoint mortality (background data,see Table 2).

Compound	Geometric mean 10-12 LC50 of values reported in Table 2 (µg/g OC)					
	H. azteca	*L. plumulosus*	*C. dilutus*	*J. kutera*	*N. virens*	*M. mercenaria*
DDT	205					829
Dicofol	>573		915			
Dieldrin	2000		57			
Endrin	124		4.22			
Endosulfan (sulfate)	873	144	23	324	17	
Alpha-endosulfan	51.7		0.96			
Methoxychlor	85.8		36.7			

Table 4

Chronic sediment toxicity data for organochlorine compounds.

Compound	Species	Effect endpoint	L(E)C50/NOEC (µg/g OC)		References
			Mean	Toxicity endpoint	
DDD	*Hyalella azteca*	Mortality	250	28d LC50	Ingersoll et al., 2005
		Length	240	28d EC50	
		Length	250	42d EC50	
		Reproduction	120	42d EC50	
DDT	*Neanthes arenaceodentata*	Reproduction	17100	120d NOEC	Murdoch et al. 1997

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Table 8

Acute and semi-chronic sediment toxicity data for organophosphorous and carbamate insecticides.

Compound	Species	Effect endpoint	L(E)C50/NOEC (µg/g OC) Mean (Range)	Toxicity endpoint	References
Azinphos-methyl	*M. littorale* *A. tenuiremis*	Mortality Mortality	370 21.8	4d LC50 4d LC50	Klosterhaus et al., 2003
Carbaryl	*C. riparius*	Mortality	14.7	1d LC50	Fisher et al., 1993
Carbofuran	*C. riparius*	Mortality	0.26	1d LC50	Fisher et al., 1993
Carbofuran	*C. dilutus*	Mortality	0.43	10d LC50	Douglas et al., 1993
Chlorpyrifos	*H. azteca*	Mortality	1.77	10d LC50	Amweg & Weston, 2007
Chlorpyrifos	*H. azteca*	Mortality	4.4	10d LC50	Hintzen et al., 2009
Chlorpyrifos	*H. azteca*	Mortality	4.1 (4.1-4.2)	10d LC50	Weston et al., 2009
Chlorpyrifos	*C. dilutus*	Mortality	7.74 (5.51-9.96)	10d LC50	Ankley et al., 1994
Chlorpyrifos	*C. dilutus*	Mortality	6.68	10d LC50	Harwood et al., 2009
Chlorpyrifos	*A. abdita* *E. estuarius*	Mortality Mortality	15.9 13.2	10d LC50	Anderson et al., 2008
Chlorpyrifos	*A. tenuiremis*	Mortality (adult)	1.74	4d LC50	Green et al., 1996
Diazinon	*H. azteca* *C. dilutus* *C. dilutus*	Mortality Mortality Growth	15.4 (2.8-24.4) 54.3 15.9	10d LC50 10d LC50 10d NOEC	Ding et al., 2011
Dichlorvos	*C. riparius*	Mortality	0.56	1d LC50	Fisher et al., 1993
Ethion	*C. riparius*	Mortality	13.1	1d LC50	Fisher et al., 1993
Leptophos	*C. riparius*	Mortality	>167	1d LC50	Fisher et al., 1993
Malathion	*C. riparius*	Mortality	0.22	1d LC50	Fisher et al., 1993
Methyl-parathion	*C. riparius*	Mortality	0.13	1d LC50	Fisher et al., 1993
Methyl-parathion	*H. azteca* *C. dilutus* *C. dilutus*	Mortality Mortality Growth	6.9 (2.8-12.7) 318 115	10d LC50 10d LC50 10d NOEC	Ding et al., 2011
Mevinphos[x]	*C. riparius*	Mortality	0.88	1d LC50	Fisher et al., 1993
Mexacarbate	*C. riparius*	Mortality	0.85	1d LC50	Fisher et al., 1993
Parathion	*C. riparius*	Mortality	2.4	1d LC50	Fisher et al., 1993
Propoxur	*C. riparius*	Mortality	3.6	1d LC50	Fisher et al., 1993

[x] In the original paper denoted as Phosdrin.

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Table 9

Comparison of acute and semi-chronic sediment toxicity data for organophosphorous and carbamate insecticides between the amphipods Hyalella azteca, Eohaustorius estuarius and Ampelisca abdita, the copepods Amphiascus tenuiremis and Microarthridion littorale and the insect Chironomus dilutus and Chironomus riparius (for background data see Table 8).

Compound	Geometric mean of acute LC50 values in Table 8 (µg/g OC)						
	H. azteca	*E. estuarius*	*A. abdita*	*A. tenuiremis*	*M. littorale*	*C. dilutus*	*C. riparius*
Azinphos-methyl				21.8 [b]	370 [b]		
Carbofuran						0.43 [a]	0.26 [c]
Chlorpyrifos	2.8 [a]	13.2 [a]	15.9 [a]	1.74 [b]		7.2 [a]	
Diazinon	15.4 [a]					54.3 [a]	
Methyl-parathion	6.9 [a]					318 [a]	0.13 [c]

[a] 10d LC50; [b] 4d LC50; [c] 1d LC50

Table 10

Semi-chronic sediment toxicity data for insecticides, other than organochlorines, pyrethroids and acetyl-cholinesterase inhibitors.

Compound	Species	Effect endpoint	EC50/LC50/NOEC (µg/g OC)		References
			Mean (Range)	Toxicity endpoint	
Biopesticides					
Abamectin	*H. azteca*	Mortality	19.9 (11.3-26.2)	10d LC50	Ding et al., 2011
	C. dilutus	Mortality	0.18	10d LC50	
	C. dilutus	Growth	0.10	10d NOEC	
Emamectin (benzoate)	*E. estuarius*	Mortality	>29	10d LC50	Kuo et al., 2010
Emamectin (benzoate)	*C. volutator*	Mortality	9.56	10 d LC50	Mayor et al., 2008
	N. diversicolor	Mortality	85.5	10 d LC50	
Benzoylureas					
Lufenuron	*C. riparius*	Wet weight	0.6 [b]	10d NOEC	Hooper et al. 2005
		Mortality	1.2 [b]	10d NOEC	
Teflubenzuron	*C. riparius parents*	Emergence	4.0 [a]	10d NOEC	Tassou & Schulz, 2011
	C. riparius offspring		2.48 [a]	10d NOEC	
Other insecticides					
Fipronil	*H. azteca*	Mortality	4.1	10d LC50	Hintzen et al., 2009
Fipronil	*C. dilutus*	Mortality	0.13	10d LC50	Maul et al., 2008b
		Growth	0.12	10d EC50	
Imidacloprid	*L. variegatus*	Mortality	>925.9	10d LC50	Sardo and Soares, 2010
		Avoidance	<9.3	10d EC50	
		Growth	<9.3	10d NOEC	
Indoxacarb	*H. azteca*	Mortality	>1419 [c]	10d LC50	Ding et al., 2011
	C. dilutus	Mortality	11.3	10d LC50	
	C. dilutus	Growth	3.2	10d NOEC	
Metaflumizone	*C. riparius*	AFDW	48.0 [a]	10d EC50	EU DAR
			10.4 [a]	10d NOEC	
Acaricides					
Etoxazole	*C. riparius*	Growth rate	1000 [a]	10d NOEC	EU DAR
Propargite	*H. azteca*	Mortality	576 [e]	10d LC50	Ding et al., 2011
	C. dilutus	Mortality	964	10d LC50	
	C. dilutus	Growth	633	10d NOEC	

[a] Assuming 2.5% organic carbon content in OECD 218 standard sediment.

[b] Assuming 5% organic carbon in OECD 207 standard sediment.

[c] Range of values given as >2630, >1010 and >616 µg/g OC for three sediments.

[e] Range of values given as 576, >408 and >416 µg/g OC for three sediments.

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Table 14

Semi-chronic sediment toxicity data for herbicides and the benthic invertebrates Chironomus dilutus, Chironomus riparius and Hyalella azteca and the rooted macrophyte Myriophyllum aquaticum.

Compound	Species	Effect endpoint	EC50 / LC50 (µg/g OC) Mean (Range)	Toxicity endpoint	References
Atrazin	*C. dilutus*	Mortality	>186	10d LC50	Douglas et al., 1993
Atrazin	*M. aquaticum*	RGR fresh weight Yield fresh weight	28[a] 20.4[a]	10d E_rC50 10d E_yC50	Teodorovic et al., 2012
Atrazin	*M. aquaticum*	Yield fresh weight	116	13d E_yC50	Schreiber et al. 2011
Bentazone	*Ch. riparius*	Mortality Growth	276000 (209000-325000) 10850 (7200-14500)	10d LC50 10d NOEC	Mäenpää et al., 2003
DNOC	*M. aquaticum*	Yield fresh weight	348.4	13d E_yC50	Schreiber et al. 2011
Ioxynil	*C. riparius*	Growth	787	10d NOEC	Mäenpää et al., 2003
Metsulfuron methyl	*M. aquaticum*	Yield fresh weight	0.021	13d E_yC50	Schreiber et al. 2011
Oxyfluorfen	*H. azteca* *C. dilutus* *C. dilutus*	Mortality Mortality Growth	>6140[b] 630 312	10d LC50 10d LC50 10d NOEC	Ding et al., 2011

[a] Assuming 2.5% organic carbon content in OECD 218 standard sediment,

[b] Range of values given as >12300, >3400 and >2720 µg/g OC for three sediments.