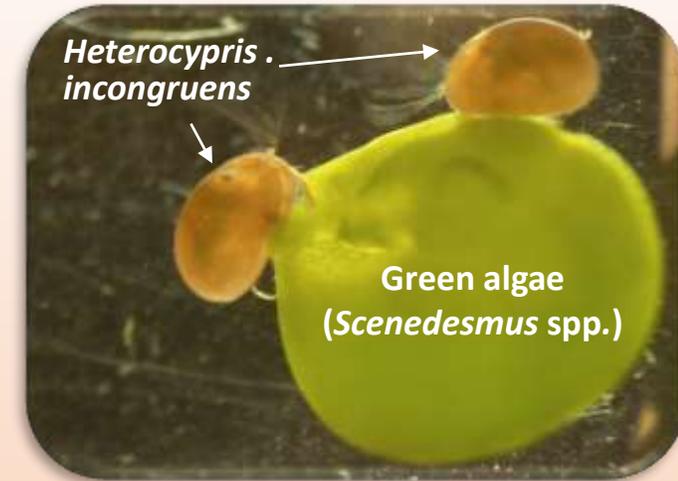


THE OSTRACOD *HETEROCYPRIS INCONGRUENS* TOXICITY TEST FOR EVALUATING CHEMICAL MIXTURE EFFECTS IN EXCAVATED SOIL



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Earth Pressure Balance - Tunnel Boring Machines (EPB-TBMs) functioning requires the use of additives, such as commercial foaming products (FA). FA are mixtures of various chemicals; most of them contain the anionic surfactant sodium lauryl ether sulphate (SLES)



Once excavated, the soil conditioned with FAs (spoil material) requires an environmental compatibility evaluation for its re-use, in accordance with the Italian Ministry for Environment and the current legislation (Italian Decree 120/2017).



Set-up of soil microcosms using conditioned soil from the real tunnel excavation site

- 2 soil types - 2 different foaming products **FAX** and **FAY**

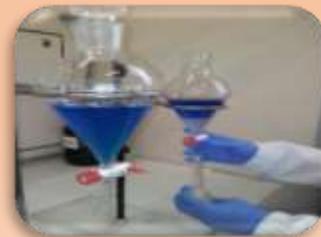


The soil samples were treated at the real treatment ratio used for tunnelling; Soil samples were collected and analysed at 3 different times (0, 14, 28 days)

Chemical analysis

SLES residual concentrations (mg/ Kg)

- Pressurized Liquid Extraction (PLE).
- MBAS spectrophotometric method ($\lambda=650$ nm)



Ecotoxicological analysis

ISO 14371:2012 (Water quality — Determination of fresh water sediment toxicity to *Heterocypris incongruens*, Crustacea, Ostracoda)



Sub-chronic "direct contact" test

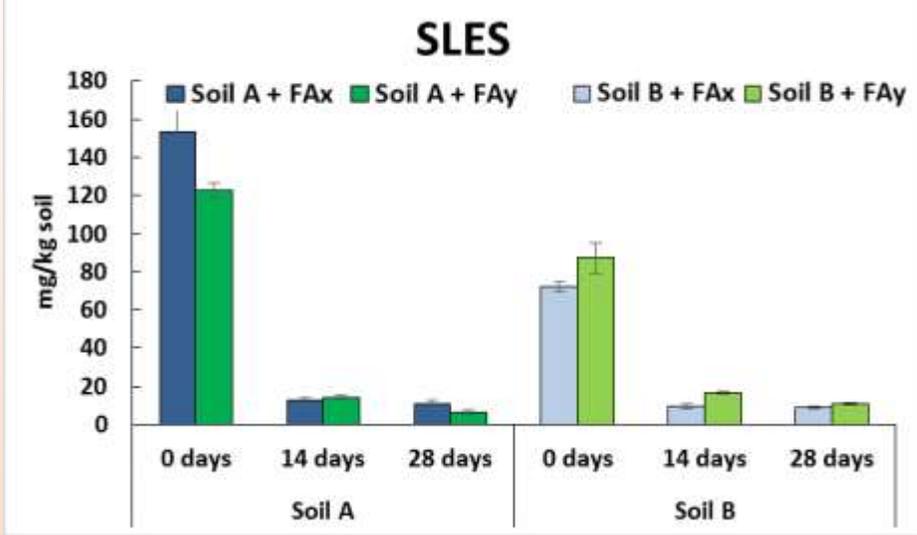


Endpoint (6 days):
• Mortality
• Growth Inhibition

Results



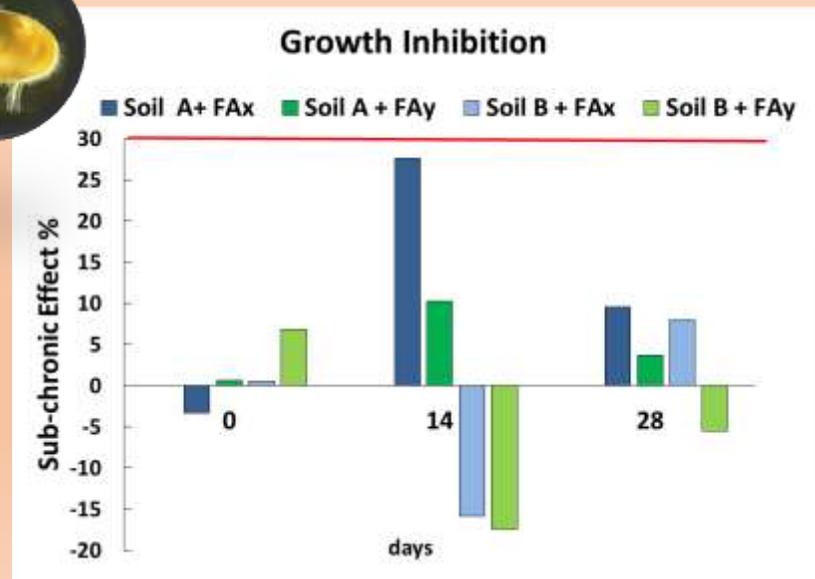
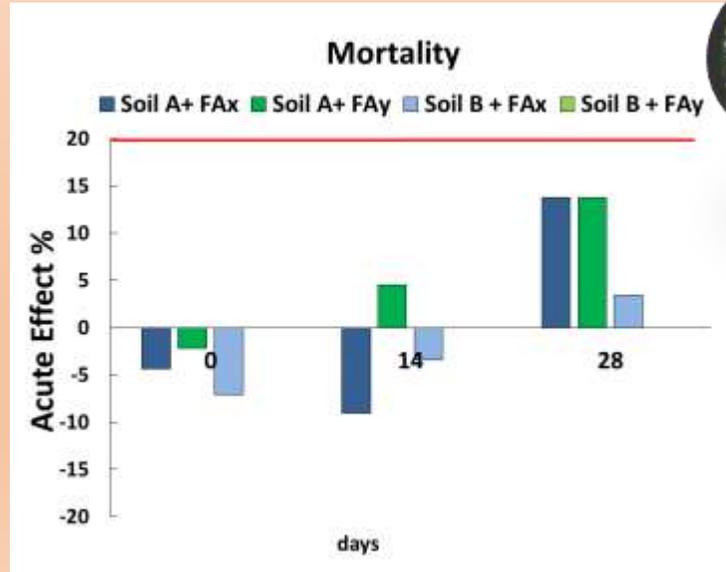
SLES degradation in soil: at 14 and 28 days residual concentrations <20% (in the range of 5-19%) were found in all conditions, corresponding to ca. 10 mg/kg soil (in the range of 6-17 mg/kg).



H. incongruens was directly exposed to the conditioned and unconditioned soils (control). The ecotoxicological responses were calculated net of the effect of the untreated soil. No acute and sub-chronic toxicity effects were recorded.

All mortality values were < 20% (threshold toxicity value).

Py + soil B showed no acute effect throughout the duration of the experiment (0-28 days).



All growth inhibition values were < 30% (threshold value for sub-chronic toxicity).

Py (green columns) showed lower growth inhibition values for both soil type (A and B) at 14 and 28 days.

Negative values correspond to a higher organism survival or growth in the soils treated with Px and Py

Concluding remarks

- ✓ *Heterocypris incongruens* test is a valuable and practical tool for the evaluation of possible toxicological effects due to a chemical mixture in an environmental matrix, as in the case of soils conditioned with foaming products.
- ✓ The advantage of this test relies on the fact that the organism is in contact with the substances contained both in the soil and in the fraction released in water, because the crustacean lives in soil/water interface.
- ✓ It is used in a battery test for evaluating the environmental compatibility of spoil material, whatever its final reuse, including in areas in contact with water bodies.
- ✓ Ecotoxicity tests make it possible to assess the impact of environmental matrices as a whole, evaluating also the possible effects of un-known contaminants (e.g. those non declared in the commercial products).

