

DAIMON Toolbox Fact Sheets:

Methods to Study the Impact of Dumped Munitions on Marine Biota

Assessment category 3: Biological effects

Toolbox component: General stress

Fact Sheet 3.11: Superoxide dismutase activity (SOD)

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What is it?

Superoxide dismutase (SOD) is a major enzyme in the antioxidant defence system (ADS) functioning against reactive oxygen species (ROS). SOD catalyzes the conversion of the superoxide anion $O_2^{\cdot-}$ to the less harmful hydrogen peroxide (H_2O_2), which is further broken down by catalase (CAT) and glutathione peroxidase (GPx).

What does it tell you?

Elevated levels of SOD activity indicate that the cellular ADS has been activated due to an accelerated production of ROS that need to be neutralized to prevent oxidative damage to macromolecules. On the other hand, lowered SOD levels may indicate that the ADS is already overloaded and the organism cannot properly cope with the excessive amounts of ROS (the so-called bell-shape response).

Type of Indicator (tick box)

- non-specific stress indicator
- specific for groups of contaminants incl. CWA or explosives
- CWA-specific indicator
- specific for substances related to explosives (e.g. TNT)

How to measure it?

Species: SOD can be measured in a large variety of organisms, including fish and mussels.

Matrix: Fish liver tissue and mussel digestive gland and gill tissue homogenates.

Equipment: Spectrophotometer/microplate reader able to measure at 450 nm; microplates/cuvettes; basic laboratory equipment (pipettes, decanters).

Measurements and units: Measurement of SOD activity is recommended to be performed with a commercial kit, e.g., Sigma-Aldrich 19160 SOD Determination Kit (Sigma-Aldrich, 2014). The activity of SOD (Units/ml) is adjusted to the protein concentration of the sample, measured with, e.g., the Bradford method (Bradford, 1976).

Calculations: According to the kit protocol for a non-kinetic measurement.

Sample size: Measurements are made from at least 15-20 individual specimens from each study site.

How to analyze and assess the data?

Compare the SOD activity levels measured from organisms collected from the target area to those from the reference area. An elevated or lowered activity level (bell-shape response) compared to the reference area indicate a negative effect. If the difference in mean activity level is more than one standard deviation (SD) of the mean values measured in the reference area, stress is considered moderate. If the level differs more than two SDs, stress is severe.

References

Bradford, M.M. 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical Biochemistry*. 72:248-254.

Sigma-Aldrich. 2014. Product Information 19160 SOD determination kit. Sigma-Aldrich Co, St. Louis USA.