

Marine Invertebrate Caffeine Exposure: stimulated stress response in *Hemigrapsus* *oregonensis*

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Caffeine Occurrence and Impact in Marine Environment

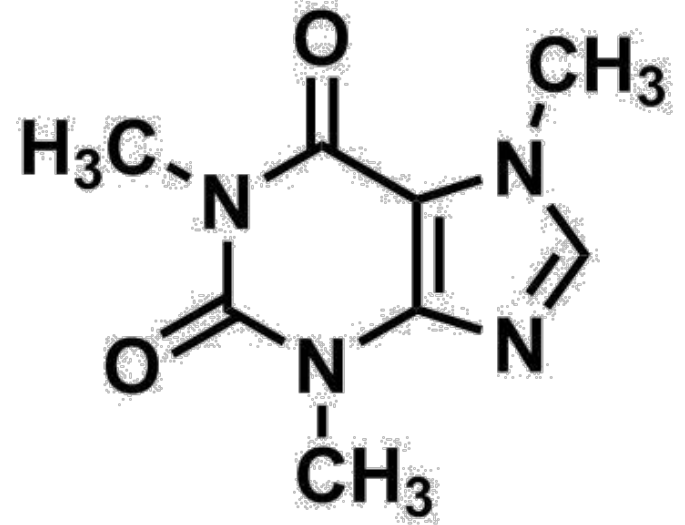
- Considered a representative pharmaceutically active compound pollutant (Li et al., 2020)
- Primarily sourced from wastewater, used as a marker of anthropogenic activities (Hillebrand et al., 2012)
- Relevant caffeine concentrations in marine animal tissues after long-term exposure (Vieira et al., 2022)

Caffeine Effects on Marine Animals and Ecosystems

- Delayed hatch time and embryo development (Garcia et al., 2014)
- Establishes oxidative stress and an increase in metabolic activity (Cruz et al., 2016) (Aguirre-Martínez et al., 2013)
- Negatively influences species' abundance, biomass, and fitness (Vieira et al., 2022)

Crabs and Caffeine: Digestive Tract

- Depolarizes membrane potential (Hermann, 1981)
- Decreases the threshold for bursting pacemaker PD neurons (Hermann, 1981)
- Inhibition of acetylcholinesterase activity (Baracchini et al., 2024)
- Decreases in the lysosomal membrane stability (Aguirre-Martínez et al., 2012)



(FD, 2017)

Crabs and Caffeine: Muscle

- Induces tension in depolarized muscle by lowering threshold (Huddart, 1969)
- Accelerates the release of Ca^{2+} from the SR in the muscles (Huddart, 1969)
- Suppresses the active binding of mycoplasma Ca^{2+} (Huddart, 1969)

Crabs and Caffeine: Gill Membrane

- Release of calcium from the ER and increased flux in intracellular Cd and Cu in gill cells (Ortega et al., 2014; Sá & Zanotto, 2013)
- Effects on the Na⁺/K⁺-ATPase transporter (Ortega et al., 2014; Sá & Zanotto, 2013)

Research Question

How do different environmental concentrations of caffeine facilitate physiological stress responses in *Hemigrapsus oregonensis*?

Hypotheses:

Null: Different environmental concentrations of caffeine will have no effect on the physiological stress response in *Hemigrapsus oregonensis*.

Alternative: Higher environmental concentrations of caffeine will have an increased effect on the physiological stress response in *Hemigrapsus oregonensis*.

Experimental Design: Set-Up

- Four tanks (approx. 5 crabs ea.):
 - Control → 0 $\mu\text{g/L}$ of caffeine
 - Low → 5 $\mu\text{g/L}$ of caffeine
 - Medium → 15 $\mu\text{g/L}$ of caffeine
 - High → 20 $\mu\text{g/L}$ of caffeine

Similar to previous studies (Baracchini et al. 2024, Aguirre-Martinex et al. 2012)

Experimental Design: Methods

- Day 0:
 - Initial respiratory and righting time
 - Establish caffeine concentrations
- Week 1:
 - Record current caffeine concentrations
 - Record hemolymph, respiratory and righting time
- Week 2:
 - Record final caffeine concentrations
 - Record hemolymph, respiratory and righting time
- Concentration of caffeine will not be maintained

Experimental Design: Data and Analysis

- Oxygen consumption and righting time
- Hemolymph Tests:
 - BCA Proteins
 - Lactate

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Questions?