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Preface

Oxidative stress in aquatic ecosystems: Selected papers from the First International Conference

The First International Conference on Oxidative Stress in Aquatic Ecosystems was held on November 20–24, 2012 in San José del Cabo, Mexico. The Conference was hosted by the Centro de Investigaciones Biológicas del Noroeste. The academic program of the Conference included lectures from ten invited speakers, nine student awardees, six virtual conferences and 39 poster presentations. The Conference was attended by 53 participants from eleven countries. This special issue of CBP-A includes a selection of the contributions from the Conference, including research articles and reviews. Together, these contributions provide a general overview of current methodologies in free radical and antioxidant biology and an interdisciplinary view of free radical production and antioxidant defenses in organisms that inhabit aquatic environments. Specific topics include methods for detection of reactive oxygen and nitrogen species (ROS, RNS) in living cells, pro- and antioxidant responses of aquatic organisms to contaminants, and strategies employed by aquatic organisms to tolerate situations that potentially elicit oxidative stress.

Recurrent issues in studies of oxidative stress are the extremely short half-life of ROS and RNS and the optimization of the methodologies for non-model organisms. Virtually all of the methods that are commonly used to quantify ROS and RNS were developed for use with terrestrial organisms, mostly humans. Additionally, these indirect methods require cell systems, biological fluids, extracellular material or tissue extracts. González et al. (2013) review the use of electron paramagnetic resonance (EPR) spectroscopy in the analyses of biologically relevant ROS in marine organisms, under both physiological and oxidative stress conditions. General features of EPR spectroscopy and specific details for quantifying the production of the ascorbyl ($A^{\bullet}$) and lipid radicals in a suite of marine invertebrates are presented. By means of EPR spectroscopy, the authors suggest that the ratio of ascorbyl radical/total ascorbate ($A^{\bullet}/AH^{-}$) may be used as an early diagnostic tool of oxidative stress development in the tissues exposed to environmental stressful conditions. In his review, Grisham (2013) analyzes different methods to detect and quantify hydrogen peroxide (H_2O_2) production and introduces a method that allows its measurement in living cells. This manuscript begins by highlighting the importance of H_2O_2 in biological systems, including its role as a major second messenger. Methods for detecting extracellular H_2O_2 in cultured cells, organ cultures and isolated, buffer-perfused tissue preparations, and five methods for detecting intracellular H_2O_2, are described. In this era of genomics, proteomics, metabolomics and epigenetics, Storey and Wu (2013) encourage those who study non-model organisms to adopt the high throughput approaches to the regulatory control of cellular metabolism that have been developed for biomedical models. These approaches allow a multi-faceted assessment of the production, interactions, signaling activity, and metabolism of ROS, RNS and antioxidants in very small sample volumes. All three reviews address the advantages and pitfalls of the methods described therein.

Since aquatic ecosystems are often characterized by spatial and temporal variation in oxygen availability, aquatic organisms can be exposed to environmental conditions of hyperoxia, hypoxia and even anoxia. These environmental conditions are usually considered challenging for aerobic organisms. The implications, in terms of ROS production and antioxidant defenses, for aquatic organisms exposed to environmental hypoxia or anoxia are the topic of the reviews by Welker et al. (2013), Joyner-Matos and Chapman (2013) and Storey et al. (2013). While Welker et al. (2013) focus on animals living in the intertidal zone that are exposed to hypoxia and reoxygenation, Joyner-Matos and Chapman (2013) center their studies on clams (*Sphaerium* sp.) living in freshwater environments with sustained hypoxia, and Storey et al. (2013) describe metabolic and regulatory responses to anoxia and freezing in the intertidal periwinkle (*Littorina littorea*). Novel results and explanations arise from these studies; notably, the suggestion that living in environmental conditions of hypoxia may be, contrary to popular opinion, beneficial.

The identification of appropriate biomarkers is essential for toxicological studies. Oxidative damage indicators and antioxidant defenses are frequently used as biomarkers of exposure to heavy metals and pesticides, although the utility of these biomarkers is often challenged. Changes in oxidative damage indicators and antioxidant enzymes in freshwater green algae and cyanobacteria following exposure to heavy metals and polycyclic aromatic hydrocarbons are reported by Vega-López et al. (2013). Effects of trace elements and PAH on pro- and antioxidants in oysters (*Crassostrea corteziensis*) were analyzed by Girón-Pérez et al. (2013). Tissue-specific differences in trace element concentrations, oxidative damage indicators and antioxidant enzyme activities between sex and maturity cohorts in blue shark (*Prionace glauca*) are presented by Barrera-García et al. (2013) and in mako shark (*Isurus oxyrinchus*) by Vélez-Alavez et al. (2013). These studies confirm that markers of oxidative stress can be reliable biomarkers in toxicological studies of aquatic (freshwater and marine) organisms. Dzul-Caamal et al. (2013) recommend the use of fish mucus as a non-invasive biomarker and report dose-dependent alterations in ROS levels, antioxidant content and oxidative damage indicators in fish (*Goodea gracilis*) exposed to halomethanes. The advantages of using such non-invasive markers when studying endangered species are addressed.

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Sobrino-Figueroa (2013) address the consequences of exposing zebrafish (*Danio rerio*) to detergents, including tissue-specific patterns in oxidative damage indicators and the finding that dishwater detergents (containing biologically active enzymes), more than synthetic detergents, increase oxidative damage. Oliveira Lobato et al. (2013) reported increased concentrations of metallothionenin-like proteins, and changes in ROS production and antioxidant defenses induced by simultaneous exposure to arsenic and cadmium in shrimp (*Litopenaeus vannamei*). These authors also showed that the effects were prevented by pre-treatment of the shrimp with lipoic acid.

Nanoparticles are becoming highly popular and are increasingly being used in several industries, including pharmaceutical and biomedical applications. However, little attention has been directed to potential toxic effects of nanomaterials in aquatic environments. The manuscripts by Da Rocha et al. (2013) and Longaray-Garcia et al. (2013) address this issue. Antioxidant defenses and expression of transcription factors are altered in freshwater zebrafish (*D. rerio*) exposed to fullerenol and single-wall carbon nanotubes (Da Rocha et al., 2013). Time and form of administration (free or in polymeric nanocapsules) were key factors in the differential effects elicited by lipoic acid on oxidative damage, antioxidant enzyme activity and expression in brain, liver and muscle of *Cyprinus carpio*; furthermore, the composition of the nanocapsules per se promoted both antioxidant and prooxidant effects in these fish (Longaray-Garcia et al., 2013). Future studies are required to address the safety of using nanomaterials for aquatic organisms.

The causes of, and responses to, oxidative stress in aquatic organisms merit more attention for several reasons. Aquatic environments are subjected to continuous variation in oxygen, salinity, light and nutrients, both in space (latitudinal, longitudinal, depth) and in time (daily, seasonal, multiannual). The plants and animals in these habitats likely employ a suite of adaptations that minimize the disruptive effects of this variation. Some of these strategies, such as the patterns of expression of antioxidant and chaperone proteins in sessile intertidal organisms, already serve as useful models for biomedical issues (e.g., the ischemia/reperfusion that accompanies myocardial and cerebral infarction). Additionally, aquatic environments are the ultimate destination of solid waste, pesticides, detergents and nanomaterials. Since we regularly harvest for food the organisms that live in these habitats (including some of the study organisms in the contributions presented here), it is critical that we be able to recognize when an organism has been exposed to toxic materials. The hypothesis that aquatic organisms can tolerate (and even thrive) when exposed to environmental hypoxia merits additional attention with respect to aquatic pollution, since aquatic organisms (including harvested species) may be more resistant to pollutants than we thought. Finally, studies of the biochemical mechanisms leading to the production of ROS and RNS, their interactions with other molecules and, particularly, how these mechanisms allow aquatic organisms to tolerate extreme environmental or metabolic conditions may lead to the understanding of the etiology of the diverse human diseases associated with oxidative stress.

As Rebeca Gerschman, a well known pioneer in free radical biochemistry wrote, 'Nature does not easily yield its secrets, what seems the answer to a research inquiry is only the beginning of scrutinizing new aspects of the problem'. This issue presents the current partial answers to inquiries on the molecular mechanisms of oxidative stress and toxicity, as well as oxidative and nitrosative damage in aquatic organisms. The exciting times of discovery and the reflection times of reviewing and summarizing are parts of the same scientific endeavor in its slow progress toward knowledge. We feel that this Conference was a valuable contribution by exploring aspects of research that are intellectual milestones toward the recognition of critical issues in aquatic biochemistry.

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