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Review Article

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Sea Urchin Bioassays in Toxicity Testing: I. Inorganics, Organics, Complex Mixtures and Natural Products

Giovanni Pagano[1,2*], Marco Guida[1], Marco Trifuoggi[1], Philippe Thomas[3], Anna Palumbo[2], Giovanna Romano[2] and Rahime Oral[4]

Abstract

Based on established knowledge in physiology and embryology, sea urchins have proven to be excellent bioassay models since the early 1970's when evaluating the impacts to early life stages and cellular functions after exposure to a number of xenobiotics. This review attempts to build a comprehensive survey of reports on sea urchin gametes and embryos used in toxicity testing of different classes of xenobiotics including inorganics, organics, different complex mixtures, and natural products. Our results provide support for common endpoints used when evaluating adverse effects on sea urchin early life stages, including frequency of developmental defects and/or cytogenetic abnormalities, changes in sperm fertilization success and offspring damage, and other endpoints related to redox alterations, DNA damage and other molecular biomarkers. Current studies using sea urchin bioassays continue to provide support for this tool in toxicology including, amongst others, studies of ocean acidification, action mechanisms of candidate new drugs, and novel xenobiotic releases to the environment.

Keywords

Sea urchin; Fertilization; Embryogenesis; Cytogenetic abnormalities; Redox alterations

Introduction

Sea urchins have proven to be extremely helpful in providing scientific insight in a number of biological disciplines, including physiology, embryology, biochemistry and genetics, in studies dating as far back as the late 19th century to the early 20th century [1-6]. Those studies provided a body of knowledge that became foundation in what we know in basic cellular events such as mitosis with the first reports on chromosomes [2], fertilization and embryogenesis [3-5], with implications well beyond echinoderm biology and thereafter translated into general biology and medicine. Relatively few reports on the exposures of sea urchin embryos to various chemicals were available, and focused primarily on the influence of some agents in modulating normal embryogenesis, rather than on investigating any adverse effects of chemicals, e.g. lithium-induced "animalization" [6,7].

***Corresponding author:** Giovanni Pagano, Federico II Naples University, via Cinthia, I-80126 Naples, Italy, Tel: +39-335-7907261; Fax: +39-825-1911712; E-mail: gbpagano@tin.it

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In the wave of the early concern in radiobiology, drug toxicity, and environmental pollution, sea urchins were first utilized in pioneering studies evaluating the effects of sperm irradiation [8], marine pollution [9], pharmaceuticals and pesticides [10-12], and the action mechanisms of carcinogens [13,14], in bioassays viewed as complementary to the classical murine models. Those studies dating up to 1970's set the grounds for continuing investigations up to present.

The body of literature of toxicity testing in the sea urchin model includes reports focused on a number of morphological and/or molecular endpoints assessing adverse effects. This prevents attempting extensive comparisons (e.g. scaling EC50's) in-between several agents. Though with this limitation, the present review may both provide an unprecedented archive of sea urchin toxicity testing reports and a useful contribution in future study design.

Inorganic agents

Seawater acidification: As shown in Table 1, pH changes were initially studied in sea urchin embryos and sperm as a subject of bioassay standardization, or for their possible implications in freshwater systems, related to the recognized phenomenon termed acid rain [15]. Early reports found that developing embryos under slightly decreased pH conditions, in the order of 0.5 pH units, underwent damage to embryogenesis and mitotic aberrations, with stronger effects when treatment was performed before hatching than in later developmental stages (blastula/gastrula) [16,17]. When sea urchin sperm were exposed to pH changes, fertilization was inhibited by either pH decrease (<7) or increase (>8.5). Furthermore, transmissible damage to the offspring of low pH-exposed sperm was observed, both in terms of developmental defects and of decreased mitotic activity [18,19]. It should be noted that slightly decreased pH (such as 7.5) enhanced fertilization success, as confirmed in a more recent report [20]. A series of studies by Stumpp et al. [21-24] showed that elevated seawater pCO_2, closely resembling the expected values by the end of this century, altered the expression of 26 representative genes important for metabolism, calcification and ion regulation, along with impacting growth and resulting in developmental delay of echinoid larvae. Adult sea urchins (*P. lividus*) were tested by Lewis et al. [25] for their sensitivity to micromolar copper levels in ambient *vs.* acidified seawater, resulting in DNA damage and oxidative stress responses, beyond the mere data of metal speciation.

Altogether, the available literature on the effects of pH decrease in sea urchin bioassays corroborates the of global concern about ocean acidification.

Other inorganics: A number of inorganics (Cd, As, Cr, Pb, Cl, Ag, Mn, La, Ce, Gd) were tested for their adverse effects on sea urchin development, fertilization success, mitotic activity, as well as other effects as induction of apoptosis, calcification defects, DNA damage and/or increasing production of reactive oxygen species (ROS) amongst others [26-59] (Table 1). Groups of inorganics were tested in an extensive body of studies providing comparative toxicity datasets for various agents [40,59]. This was the case for early studies [41,42] testing groups of inorganics as Ag, Cd, Cr, Hg and Pb; other studies reported on the effects of individual inorganics *vs.* their mixtures [43,54].