

Journal of Biological Research

Bollettino della Società Italiana di Biologia Sperimentale



**91st SIBS Congress on the role of the
Italian Society for Experimental Biology
in the Italian research**

Ancona, Italy, 9-10 November 2018

ABSTRACT BOOK

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species is possible thanks to prominent membranes which create the surface, making it possible to overcome significant distances in the air. Using routine methods of LM, TEM and SEM we have studied the skin from gliding membrane, toe, and dewlap. We observed a conspicuous bundles of tonofilaments in the cytoplasm of epithelial cells, allowing this species the locomotion and the mechanism of adhesion to surfaces (gliding membrane, toe, dewlap). In the toe outer layer, are present prismatic cells; furthermore in the epidermis of the gliding membrane we have observed a layer of keratinized cells near the surface. In the connective tissue under the epithelium there are numerous mucous glands, an abundance of nerves and several types of cells including: melanophores, iridiophores and xanthophores. Our future studies are aimed to compare *Rhacophorus*'s skin structures with those of other species exhibiting similar behaviour (climbing/flying) (*Rana leucomystax*, *Litoria caerulea*) and to characterize Merkel Cells and Langerhans cells, both cells present in the skin of all vertebrates.

THE COMBINED EXPOSITION TO POLYBROMINATED COMPOUNDS AND β -BLOCKERS MODULATES VITALITY AND OXIDATIVE STRESS PARAMETERS IN THE MARINE FISH CELL LINE SAF-1: PRELIMINARY RESULTS

Concetta M. Messina^{1*}, Cristobal Espinosa Ruiz¹, Simona Manuguerra¹, Maria Morghese¹, Rosaria Arena¹, Giuseppe Renda¹, Alberto Cuesta¹, Maria Esteban², Andrea Santulli^{1,3}

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Pharmaceutically active compounds have been detected in wastewater treatment plants and surface waters across the world. Among these compounds, some non-steroidal anti-inflammatory drugs as well as β -blockers used to treat human hypertension, such as atenolol (ATL), is one of the most abundant reported at a very variable range, in relation to the site. The uncertainty surrounding the behavior of these pharmaceutically active compounds could increase due to the presence of others pollutants in the marine environment. Polybrominated diphenyl ethers (PBDEs) are a class of brominated compounds which leads to accumulate in the marine environment, being ubiquitous and toxic. The interactions between PBDE and ATL and its effects on marine organisms are still unknown and *in vitro* system are ideal for screening of toxicity, dose-time exposure and studies on synergisms. In the present study are reported some preliminary results obtained in an experiment with the *Sparus aurata* fibroblast cell line (SAF-1), exposed to different concentrations of BDE-47, ATL and a mix of both, until 72 hours, to evaluate the cytotoxicity and reactive oxygen species (ROS) production. Evaluation of molecular markers related to toxicity, oxidative stress and cell cycle is in progress. At 72 hours the SAF-1 cells exposed to both compounds showed a decreased viability. Our preliminary results suggested the synergic effects of these class of compounds, which not only could remark the importance of the development of effective treatment strategies for remove these compounds from the marine environment, but also the prevention of the discharge.

EFFECTS OF SUB-LETHAL DOSES OF THE POLYBROMINATED BDE-47 ON SOME INNATE IMMUNE RELATED PARAMETERS OF MYTILUS GALLOPROVINCIALIS

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Polybrominated diphenyl ethers (PBDEs) belong to the class of the flame retardants and are persistent and toxic pollutants, extremely stables. In fact, PBDEs have demonstrated to produce several negative effects on marine organisms. Marine bivalves are susceptible to be exposed to a large quantity of pollutants due to its feeding habits. However, the effects of PBDEs on the innate immune system from mussels remains unclear. With the aim to shed light on this issue, within the project CISAS, specimens of *Mytilus galloprovincialis* were allowed in aquaria, fed on microalgae contaminated with different concentrations of PBDE-47. The treatment was maintained for 15 days. Samples of haemolymph were obtained from T0 to 30 days of the experiment. Both immune and humoral related parameters were evaluated and demonstrated to be affected by the treatment. In addition, the results indicated that after 15 days of detoxification, immune parameters did not restore its basal levels, but some of them resulted overexpressed. BDE-47 exposure, at environmentally realistic levels, may affect various aspects of immune function in *M. galloprovincialis*, acting as a stressor that can compromise the ability to counteract external stimuli, such as pathogens, reducing animal welfare.

ELECTROPHYSIOLOGICAL PROPERTIES OF CRUDE VENOM FROM PELAGIA NOCTILUCA (CNIDARIA: SCYPHOZOA) NEMATOCYSTS

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Cnidarians are known to negatively impact on ecosystems and human economic activities, and represent a health problem consequent to their stings. However, their venom is a rich source of bioactive compounds, and is only partially characterized. *Pelagia noctiluca* (Cnidaria: Scyphozoa) is the most abundant jellyfish in the Mediterranean Sea, and it is mostly found in the Strait of Messina (Italy). To investigate the properties of *P. noctiluca* venom, cultured human cells were exposed to the crude venom (0,025 ug/ul). Changes in the cellular volume were monitored by phase contrast microscopy and the membrane conductance was measured by patch clamp. The exposure of cells to the venom induced a dramatic cell swelling and profoundly altered the ion conductance of the plasma membrane, thus affecting the homeostatic functions of the maintenance and regulation of cellular volume. Venom-treated cells exhibited a large inwardly rectifying current, mainly due to permeation of Na⁺ and Cl⁻ and sensitive to amiloride. Cell swelling and the venom-induced current were completely abrogated following

removal of NaCl from the extracellular solution and thermal treatment of venom extract. The current-inducing activity was also observed following delivery of venom to the cytosolic side of the plasma membrane, consistent with a pore-forming mechanism. This is the first study exploring in detail the ability of *P. noctiluca* venom to modify the electrophysiological properties of mammalian cells. In this light, the study i) provides essential information on a possible use of bioactive substances; ii) provides new strategies in the treatment of envenomation.

ANTIMICROBIAL ACTIVITIES OF EXTRACTS FROM *GRACILARIA GRACILIS* (RHODOPHYTA)

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Marine algae produce a wide variety of bioactive metabolites as antimicrobial, anti-feedant, antihelminthic and cytotoxic agents. These compounds include alkaloids, polyketides, cyclic peptides, polysaccharides, phlorotannins, diterpenoids, sterols, quinones, lipids and glycerols. The antimicrobial potential differs among the different classes of seaweed, with the most promising being Rhodophyceae, followed by Chlorophyceae and Phaeophyceae. In this study, the antimicrobial potential of *Gracilaria gracilis*, from the Natural Reserve of Capo Peloro (Sicily, Italy), was evaluated. Soxhlet extractions were carried out using five different solvents (methanol, ethanol, acetone, chloroform and ethyl ether). Extracts were tested against Gram positive and Gram negative pathogens (i.e. *Vibrio cholerae*, *Pseudomonas aeruginosa*, *Salmonella* sp., *Bacillus subtilis*, *Aeromonas hydrophila*, *Vibrio fischeri*). The antimicrobial susceptibility was assessed using the "agar disk diffusion method": 20 µl of each extract (at three different concentrations) were tested. Results showed that all the extracts were active against *B. subtilis*, with the best result that was obtained using the methanolic extract (inhibition halo diameter: 19 mm). A slight activity was also observed towards *Vibrio fischeri*. Our results enlarge the existing knowledge about the antimicrobial potential of red macroalgae. Further studies will be necessary to deepen the activity of *Gracilaria gracilis* against other Gram positive bacteria. Moreover, considering that *Bacillus subtilis* is often associated with food poisoning outbreaks, it could be suggested the use of *G. gracilis* extracts in the complexation of food preservatives, useful to reduce the risks deriving from the contamination by such microorganism.

THE EFFECTS OF ACUTE EXPOSURE OF NEONICOTINOID INSECTICIDE ON DIFFERENT SPECIES OF CRAYFISH

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Neonicotinoids are one of the newest class of pesticides, used in agriculture like insecticides from the beginning of 90 years. They have become very popular and widely used all over the world for their low toxicity to vertebrates. Together, in last years some of the active substances in the neonicotinoid group (clothianidin, imidacloprid a thiamethoxam) have been banned for their secondary impact on non-target organisms and the environment. The aim of this study was to investigate the acute toxicity 96hLC50 of Calypso 480 SC (containing 48% neonicotinoid thiacloprid) on three species of crayfish. We used two different developmental stages: marbled crayfish (*Procambarus fallax* f. *virginalis*), red swamp crayfish (*Procambarus clarkii*) and common yabby (*Cherax destructor*), which were tested separately. The Calypso 480 SC had a more toxic effect on to younger stages of crayfish, the 96hLC50 in mg/L was: 1.60 and 27.30 marbled crayfish, 1.13 and 1.94 red swamp crayfish, 0.52 and 7.65 mg/L common yabby, values are sorted consecutively younger and then older. Also changes in behaviour in crayfish exposed to insecticide were observed, as is reduced aggressiveness, slowing movement of the limbs, reduced breathing, turning the crayfish on the back, apathy of organisms and subsequent deaths. This study provides and complements other important results for evaluating the toxicity effect of thiacloprid on non-target species, respectively crayfish. *Acknowledgements:* Supported by the Ministry of Education, Youth and Sports of the Czech Republic - projects CENAKVA (No. CZ.1.05/2.1.00/01.0024) and CENAKVA II (No. LO1205 under the NPU I program), and project Development of University of South Bohemia: International Mobility MSCA IF (no. CZ.02.2.69/0.0/0.0/17_050/0008486).

METHADONE VERSUS BUPRENORPHINE: DATA DETECTED FROM CALTANISSETTA SER.T

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In last years, heroin-addicted have exponentially increased: this has made it necessary to identify a pharmacological strategy as effective as possible. With this purpose, a statistical investigation was conducted in a sample of individuals, aged between 18 and 50. They were diagnosed and subjected to different treatments at Ser.T of Caltanissetta (Sicily-Italy) during the period 2013-2017. The analyzed patients were treated with three different pharmacological therapy: methadone 0.1%, methadone 0.5%, buprenorphine and suboxone. We obtained percentages of RESPONDERS, LOW RESPONDERS and NON RESPONDERS patients from data processing, based on used therapy. Considering pharmacological responses of the sample examined, it is possible to observe that the treatment with buprenorphine has led to 71.98% of RESPONDERS subjects, 23.52% of LOW RESPONDERS and 4.5% of NON RESPONDERS. Instead, the administration of methadone 0.1 % has produced 82.82% of RESPONDERS subjects, 11.08% of LOW RESPONDERS, 6.1% of NON RESPONDERS. The therapy with methadone