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of TBT induces GluR2 decrease and neurons become susceptible to glutamate toxicity.

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P307-022
Correlation between trace elements in blood and urine and human reproductive indicators

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The great concern with exposure to metals and their effects on human health has been expressed in research related to occupational health. However, few studies have been developed to correlate such exposure and reproductive health. It is well known that a long term exposure to metals such as lead (Pb) and cadmium (Cd) is correlated to adverse effects in the reproduction of many mammalian species. Thus the development of studies with fertile and infertile population is mandatory. The aim of this work was to investigate the correlation between metal levels and fertility in the general population using blood, urine, follicular fluid (FF) and seminal liquid (SL). The concentrations of those metals in biological fluids from 25 infertile couples under IVF were determined by electrothermal atomic absorption spectrometry. Results presented are preliminary since only 16 individuals were involved and categorized by gender. Among men, medians were Pb-B = 3.8 $\mu g\,dL^{-1}$, Cd-B = 1.05 $\mu g\,L^{-1}$ and Cd-U = 0.28 $\mu g\,L^{-1}$, while the results found for Pb-SL and Cd-SL were 0.78 $\mu g\,L^{-1}$ and 0.10 $\mu g\,L^{-1}$, respectively. In this category, just the relationship between Cd-S and Cd-U showed a mild correlation (Spearman's coefficient = 0.647) statistically significant (90%, $p = 0.08$). Regarding to women, the medians were Pb-B = 3.2 $\mu g\,dL^{-1}$, Cd-B = 1.05 $\mu g\,L^{-1}$ and Cd-U = 0.28 $\mu g\,L^{-1}$, while the results found for Pb and Cd in FF were 1.25 $\mu g\,L^{-1}$ and 0.76 $\mu g\,L^{-1}$, respectively. There was only a mild to strong correlation ($r = 0.745$) with statistical significance (95%, $p = 0.05$), between Pb-FF and Cd-FF. As men group showed an inverse correlation ($r = -0.240$) with no statistical significance ($p = 0.568$) for Pb-SL and Cd-SL, it may be that the statistically significant correlation for Pb-FF and Cd-FF is a characteristic of the gender. Nevertheless the results were not conclusive since the sample size was still small.

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P307-023
Important role of antioxidant enzymes in the Saccharomyces cerevisiae survival to the toxicity of vanadium pentoxide

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Vanadium pentoxide, V_2O_5 (V^{5+}) behaves as powerful oxidizing agent which participates in free radicals generation. Although some biological studies reveals that vanadium compounds have mutagenic and genotoxic effects, other authors reveal that vanadium exerts antitumoral effects by activating signaling pathways leading to apoptosis or inducing xenobiotic metabolizing enzymes which degrade active carcinogens. In other hand, several industrial processes contributes to increase the vanadium concentration in the environment, making it a pollutant. Following from the above, the main objective of this work was to evaluate the survival and enzymatic antioxidant response to V_2O_5, by Saccharomyces cerevisiae: UE-ME3, a wild-type strain; Red Fruit, a wine commercial strain and a BY4741 strain. Cells at mid-exponential phase were inoculated in YEPD medium with 2% (w/v) glucose and incubated during 72 h in a water bath with orbital stirring, at 28 °C, in the absence or in the presence of 2 mM V_2O_5. Samples from each treatment were used to obtain growth curves and to determine mitochondrial, microsomal and cytosolic fractions, used for determination of NADH cit c reductase, (NADHred), NADPH(P450) reductase, (NADPH(P450)red), glutathione peroxidase (GPx), glucose-6-phosphate dehydrogenase (G6PD) and catalase T (CAT T) activities. The results show that V_2O_5 inhibited cell growth of any yeast strains in study, although drastically pronounced in the case of BY4741 strain. Inhibition of the respiratory chain at the NADHred activity, as well as the activation of the microsomal electrons transport, via microsomal NADPH(P450)red, by vanadium appear to be important sources of ROS and toxicity for studied strains. However higher levels of CAT T in native and wine commercial strains are vital to significantly prevent cell death by V_2O_5, since GPx decrease significantly.

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P307-024
Ascorbate prevents pro-oxidant effects of vanadium pentoxide on wild-type Saccharomyces cerevisiae UE-ME3

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Vanadium pentoxide, V_2O_5, (V^{5+}) the most toxic compound of vanadium behaves as an amphoteric oxide and a powerful oxidizing agent which may be an oxidative stress inducer. The vanadium (V^{5+}) is generally reduced by living cells to vanadium (V^{4+}), less toxic, using enzymes which mobilize the reducing equivalents of NADPH, or non-enzymatically using ascorbate. Nevertheless, species generated by vanadium (V^{4+}) from H_2O_2 and lipid peroxidation, via Fenton reaction can have a significant role in the metabolism of vanadium and induce cell damage in physiological conditions. Although vanadium is an element with ubiquitous environmental distribution, combustion of fossil fuels represents an important source of vanadium in the environment. Biological studies to evaluate the influence of vanadium on living organisms has shown that is mutagenic and genotoxic. Having in account that toxicity mechanisms of vanadium on eukaryotic cells are not entirely clear, the main objectives of this work was to evaluate the synergistic effects of 0.025 mM ascorbate vs 2 mM V_2O_5 on cell survival, alkaline phosphatase (ALP), catalases A and T (CAT A, T) and glutathione reductase (GR) activities of Saccharomyces cerevisiae UE-ME3. Cells at mid-exponential phase were inoculated in YEPD medium with 2% (w/v) glucose and incubated during 72 h in a water bath with orbital stirring, at 28 °C, in the absence or in presence of 2 mM V_2O_5, or 0.025 mM ascorbate plus 2 mM V_2O_5. Samples from each treatment were used to obtain growth curves and to prepare post-12,000 g supernatant, used for enzymatic activities determination. The results shown that ascorbate counteracted growth inhibition, the decrease of ALP and CAT activities, as well as

the increase of GR antioxidants activities, caused by 2 mM V_2O_5, to values similar of control cells.

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P307-025
Comparison of synthetic chelates and compost at enhancing phytoextraction of Cd, Ni and Pb from contaminated soil under canola cultivation

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The plant which can be used to clean up the soil of heavy metals contamination are named phytoremedation. Phytoremedation has received increasing attention because of its low environmental impact and cost-effectiveness. But, it is slowly process and needs long time. Synthetic chelates and low molecular weight organic acids are the most common chemical amendment that have been used in chemically assisted phytoextraction of metals from soils. The objective of this work was comparison of EDTA and sugarcane by product compost in enhancing phytoextraction of Cd, Pb, and Ni by canola in an artificially contaminated soil. Two levels of contamination (800 and 1600 ppm) were performed. The soil was placed in dark condition for 2 weeks and compost of sugarcane was applied in two levels (20 and 50 ton per hectare). A number of 5 canola seeds with grower power 95% germination were cultivated .in discrete pots. After two weeks, the treatments included EDTA (0, 10 and 20 mmol/kg soil) in irrigated water, were added to some pots. Eight weeks after cultivating the plants were cut and the soils as well as the plants were analyzed. All treatments significantly increased the concentrations of Cd, Pb and Ni in the shoots of plants compared with the control. Therefore, the influence of EDTA and compost were observed more powerful for enhanced phytoextraction of the heavy metals. The effectiveness of EDTA and compost to stimulating the accumulation of the Cd, Pb and in shoots plants were (4.3 and 4.1), and (2.8 and 2.9) times more respectively, than the control. Also, the result of this study indicated that all treatments were superior in terms of solubilizing soil Pb, Cd and Ni for root uptake and translocation into shoots canola but, in different levels.

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P307-026
Particulate cobalt and soluble cobalt: Same metal but different toxicities as assessed by cellular biology and innovative proteomic approaches

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Cobalt is widely used in the industry as it is included in the production of drying agents, pigments, and catalysts, and it is a major constituent of hard metal alloys. Radioactive isotopes of cobalt are also used in industry, medicine and nuclear research. In nuclear power plants, ^{59}Co-containing alloys can be activated into radioactive ^{60}Co oxides, dispersed in the cooling water, and represent a major concern. Occupational exposure to Co occurs mainly via inhalation leading to various lung diseases, such as pneumonitis, fibrosis and asthma. Although the chemical toxicity of cobalt has been proven, the molecular mechanisms of its toxicity are not well described and in vitro toxicity studies are mainly focussed on cobalt chloride. Cobalt is genotoxic and induces oxidizing stress and apoptosis. As the risk of exposure to cobalt oxide is a main industrial concern, we compared the toxicity of soluble chloride cobalt and particulate oxide cobalt in in vitro tests. Since the lung is the main target organ of cobalt toxicity, we made this comparison on the human BEAS-2B lung cell line. First, we characterized the size and aggregation of particles using SEM, TEM, Specific Surface Area determination, and Dynamic Light Scattering. We followed the particle internalization pathway by measuring the cell side scatter increase. The cellular cytotoxicity, ROS generation, cell cycle modulation and hypoxia were compared for both cobalt forms. We also explored the secretomes from cell lines exposed to these toxicants using a proteomic shotgun approach and a high-resolution tandem mass spectrometer. The modulation of secretion of growth factors, cytokines, and novel peptides is currently in progress to better describe the stress response mechanism and the intercellular cross-talk induced by exposure to soluble and particulate cobalt forms.

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P307-027
Interaction of cadmium and manganese in cellular uptake and toxicity

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It has long been considered that cadmium is incorporated into cells via the pathway for calcium or iron. However, recent studies have revealed a role of zinc transporter, ZIP8, for cellular cadmium uptake. In a previous study, we have established cadmium-resistant cell lines from embryonic fibroblasts of metallothionein-null mice, and found that the uptake rates of both cadmium and manganese in cadmium-resistant cells were markedly lower than those of parental cells. We also found that the down-regulation of ZIP8 gene in cadmium-resistant cells is responsible for the decreased uptake of cadmium. To further investigate the interaction of cadmium and manganese, we examined cytotoxicity and cellular accumulation of cadmium and manganese among several rat cell lines, such as PC12, TRL1210, H9c2, and RBL-2H3 cells. We found that RBL-2H3 cells established from rat basophilic leukemia cells showed the highest sensitivity to cadmium and manganese among the cell lines examined. Cellular uptake rates of cadmium and manganese in RBL-2H3 cells were also the highest, while the excretion rates of cadmium and manganese were similar among the cell lines. The addition of manganese in the medium decreased both cellular uptake and cytotoxicity of cadmium. Furthermore, the addition of bicarbonate in the medium clearly increased the uptake rates of both metals. These data suggest that either ZIP8 or ZIP14 may be involved in the high uptake rates of cadmium and manganese in RBL-2H3 cells. Therefore, we introduced siRNA of ZIP8 or ZIP14 to RBL-2H3 cells. As a result, the knockdown of ZIP8, but not ZIP14, clearly decreased the uptake of both cadmium and manganese. These results suggest that RBL-2H3 cells serve as a good model for investigating the interaction of cadmium and manganese, and the role of ZIP8 in the uptake of these metals.

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