**Materials and methods:** Approximately  $5 \times 10^6$  cells were grown in culture flasks ( $25\,\mathrm{cm}^2$ ), exposed for  $24\,\mathrm{h}$  at non-cytotoxic concentrations of putrescine, corresponding to 70%, 50%, 30% and 10% of the LD<sub>50</sub>, which is 463 mg/kg for rats. The assay was performed in triplicate, and 3000 cells were analyzed per treatment. Normal binucleate cells and binucleate cells bearing bridges, buds, and/or micronuclei were counted. Statistical analysis was performed by ANOVA, pos test Tukey (p < 0.05).

**Results:** According to the results of the micronucleus test, all the tested concentrations exhibited significant frequency for this marker. For the formation of bridges and nuclear buds, the results were significant only for the highest tested concentration (70%).

**Conclusions:** Several studies have been conducted to evaluate the necrochurume potential contamination, but little is known about its physical and chemical properties. The obtained information on the effects of putrescine present in necrochurume is extremely important, because besides of alerting for the potential as an environmental pollutant, it can lead to a better understand of the action mechanisms of this substance.

Financial support: FAPESP.

http://dx.doi.org/10.1016/j.toxlet.2016.07.436

### PP18.9

## Evaluation of cytotoxic and mutagenic effects of CactiNea<sup>TM</sup> nutraceutical in A. Cepa



F.F. Navarro  $^{1,2}$ , F.D.C. Pereira  $^2$ , B.S. Oliveira  $^2$ , M.A. Marin-Morales  $^1$ 

**Introduction:** Natural products and functional foods have received special attention by health professionals and the general population due to the constant pursuit of welfare, as well as combating diseases. *Opuntia ficus indica* L., a species in the cactus family *Cactaceae*, it is a plant growing in dry and hot climates: northern Mexico, south-western United States, Africa, Mediterranean countries and Europe. The fruits are used in the traditional medicine. NEXIRA Health Gere Cacti-Nea<sup>TM</sup>, a cactus fruit extract with natural diuretic properties, is a dehydrated water extract of the fruits of the prickly pear cactus *Opuntia ficus indica*, obtained by a process designed to preserve the nutritional and functional properties of the fruit.

**Objective:** This study used as test organism the *Allium cepa* to conduct the evaluation of the cytotoxic and mutagenic potential of CactiNea nutraceutical.

**Materials and methods:** The assay was performed in meristematic cells of *A. cepa* exposed to eight concentrations 0.12; 0.06; 0.03; 0.01; 0.008; 0.006; 0.004; 0.002 g/mL.

**Results:** The results showed that *A. cepa* at any concentration analyzed showed significant levels of mutagenicity when compared to the positive control after the treatment period. The concentrations 0.12 and 0.06 g/mL showed a decrease in mitotic index, which shows that within this concentration range the CactiNea solutions interfere on the cell cycle and division of cells of *A. cepa*.

**Conclusions:** These results demonstrate the necessity for toxicological tests in nutraceuticals, other tests will be performed to evaluate other acute and chronic toxicological parameters.

http://dx.doi.org/10.1016/j.toxlet.2016.07.437

#### PP18.10

# Comparative study of cytotoxicity and genotoxicity of commercial Jeffamines<sup>®</sup> and polyethylenimine in CHO-K1 cells



L. Castan, C.J. Da Silva, R. Dos Santos, E. Molina

Núcleo de Pesquisas em Ciências Exatas e Tecnológicas, Universidade de Franca, Ave. Dr. Armando Salles de Oliveira, Franca, São Paulo, Brazil

**Introduction:** Jeffamines<sup>®</sup> are a family of polymers containing primary amine groups attached to the extremities of a polyether backbone, which can be used as biomaterials. They have been used in combination with polyethylenimine (PEI) to improve the biocompatibility in drug and gene delivery systems. Despite these facts, very few studies have been done on the cytotoxicity and genotoxicity of pure Jeffamines<sup>®</sup> or compared with PEI.

**Objective:** The present study aimed to evaluate and compare the cytotoxic and genotoxic effects of Jeffamines® and PEI in CHO-K1 cells. Specifically, polypropylene oxide 2000 (PPO 2000, Jeffamine® D series), polyethylene oxide 2000 (PEO 1900, Jeffamine® ED series), branched 25 kDa PEI and linear 20 kDa PEI were evaluated at different concentrations.

**Materials and methods:** Cell viability and proliferation were assessed by XTT and BrdU assays, respectively. Genotoxicity was evaluated using single cell gel electrophoresis and cytokinesis-blocked micronucleus assays.

**Results:** PPO 2000 was the most cytotoxic Jeffamine<sup>®</sup>, whereas PEO 1900 did not cause significant cell death at any tested concentration. Branched PEI was more cytotoxic than LPEI and both were more cytotoxic than Jeffamines<sup>®</sup>. Only PPO 2000 induced DNA damage when evaluated in the comet assay, probably due to its cytotoxicity. PPO 2000, PEO 1900 and PEI did not increase the frequency of micronuclei when tested at sub-cytotoxic concentrations.

**Conclusions:** This work provides new insights about the biocompatibility of Jeffamines<sup>®</sup> and PEI and suggests the genotoxicological safety for further investigations of PEO 1900 in drug and gene delivery systems.

**Financial support:** FAPESP, grant number 2014/12465-0. CAPES, grant number 12467-13-8.

http://dx.doi.org/10.1016/j.toxlet.2016.07.438

### PP18.11

## The influence of washing methods on the DNA damage levels assessed by Comet assay



A. Tirsina <sup>1,2</sup>, C. Costa <sup>2,3</sup>, S. Costa <sup>2,3</sup>, J.P. Teixeira <sup>2,3</sup>

**Introduction:** In the last years, the reliability and importance of the comet assay is significantly increasing. The protocol of the assay is continuously developing and although the main principles are the same, small differences in the procedure from one laboratory to the other seem to affect the sensitivity and outcomes. Therefore, among other crucial steps in performing the comet assay protocol is the washing method as this may introduce additional damage

<sup>&</sup>lt;sup>1</sup> Departamento de Biologia, Instituto de Biociencias, Universidade Estadual Paulista (UNESP), Rio Claro, SP, Brazil

<sup>&</sup>lt;sup>2</sup> Centro Universitário Hermínio Ometto – UNIARARAS, Araras, SP, Brazil

<sup>&</sup>lt;sup>1</sup> Department of Chemicals Surveillance, Centre of Chemical Safety and Toxicology, National Center of Public Health, Chisinau, Republic of Moldova

<sup>&</sup>lt;sup>2</sup> Department of Environmental Health, Portuguese National Institute of Health, Porto, Portugal

<sup>&</sup>lt;sup>3</sup> 4EPIUnit-Institute of Public Health, University of Porto, Porto, Portugal