cer. Toppgene tool (https://toppgene.cchmc.org/) was used to explore the role of the selected genes in gene ontology processes that could affect cancer progression, while GeneMania (https://genemania.org/) determined genes related to the set of genes and the type of interaction between all of them. To determine whether SFN has an effect on selected sets of genes, the Comparative Toxicogenomic Database (CTD, http://ctdbase.org/) was used. In prostate cancer, a total of 13 genes were consistently down-regulated, and 37 genes up-regulated. Down-regulated genes are involved in molecular functions, biological processes and pathways of muscle contraction and channel and enzyme functions, while up-regulated genes regulate processes at the level of the kidneys and the renin-angiotensin-aldosterone system. Network analysis showed that the type of interaction that dominated between downstream or upstream regulated genes and their related genes, was co-expression. Finally, SFN interacted with 21 dysregulated genes and reduced the expression of ERG and TMEFF2, while increased the expression of GSTM3, ACTG2 and CNN1 genes which can lead to positive effects such as improving antioxidant protection, suppressing expansion of tumor tissue and risk of developing bone metastases. In addition, SFN could contribute to the development of prostate cancer, by interacting with the genes already expressed in the tumor tissue. The conducted study indicates that the genomic signature of patients suffering from prostate cancer could be an important factor which determines the benefits and risks of SFN as an adjunctive therapy. It could be suggested that prostate cancer patients with increased expression of ABCC4 and ENTPD5 and decreased expression of MAMDC2, MYLK, PGM5, PPP1R3C and SYNM might not be the best candidates for SFN administration. (Serbia-China project: 451-03-1203/2021-09)

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P06-32
Oral acute toxicity prediction using Cell Painting morphological profiles

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Historically, chemical risk assessment has relied largely on animal-based approaches which are expensive and rapidly becoming socially unacceptable. Regulatory bodies such as the US Environmental Protection Agency and the European Food and Safety Authority have promoted the concept of Next Generation Risk Assessment (NGRA) in which New Approach Methods (NAMs) would bring advantages including a greater focus to the human species and more detailed information on molecular mechanism and kinetic properties of chemicals without the need of animal testing. We are presenting here the results of a case study made on rat oral acute toxicity using Cell Painting as NAM.

Oral acute toxicity is assessed using *in vivo* studies to determine the LD50. In the context of NGRA, non-animal alternatives to determine oral acute toxicity of chemicals in development are needed. *In silico* models exist and use chemical compound structures (QSAR models).

QSAR models work well when predicting the toxicity of molecules structurally close to the compounds used to build the model (model compounds). However, for compounds structurally different from the model compounds the predictions are not reliable. This can be a problem when exploring new chemistry.

To overcome this limitation, one idea is to use compound biological effects instead of chemical structures. One way to capture chemical biological effects is to measure its effect on the morphology of cells.

Cell Painting, an *in vitro* assay developed by the Broad institute, generates morphological profiles of cells perturbated by chemicals. It uses 6 dyes to reveal 8 cell compartments, forming after image analysis a robust and unbiased morphological profile.

We applied Cell Painting for the prediction of oral acute toxicity in the rat. We performed Cell Painting on U2OS cells (Human osteosarcoma) by applying 220 compounds at 3 different concentrations. To assess the potential of Cell Painting we compared morphological profile-based models to QSAR models of the same chemical space.

The results showed that QSAR models perform well when predicting the toxicity of compounds structurally close to the model compounds. Those models do not perform well when the compounds to predict are structurally different. As for morphological profile-based models, they perform almost equally regardless of the compound structure similarities. However, the presence of profiles without any morphological changes compared to control reduces the performance of the model. Only after filtering those profile out, morphological profile-based models outperformed QSAR models for compounds structurally different from the model compounds.

As a conclusion, morphological profiles are informative about oral acute toxicity. Further experiments are needed to characterize in detail its biological applicability domain together with the cell line used. To capture a broader range of biological signal, the usage of additional cell lines needs to be explored.

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Comparative study of positive and negative sulforaphane activity against different tumor types via bioinformatic analysis

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Sulforaphane (SFN) is a compound found in cruciferous vegetables which have been studied as a molecule with the potential in disease prevention and treatment. SFN has demonstrated effectiveness against cancer. However, further research is needed to enhance the understanding of its pharmaco-toxicology profile in different cancer types. Thus, bioinformatics analysis was conducted to determine potential positive and negative SFN activity in 6 cancer varieties: breast,