In silico prediction of potential mixture toxicity mechanisms underlying endocrine disorders as a result of e-waste recycling activities: Exposure to organophosphate flame retardants and toxic metal(oid)s

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Introduction

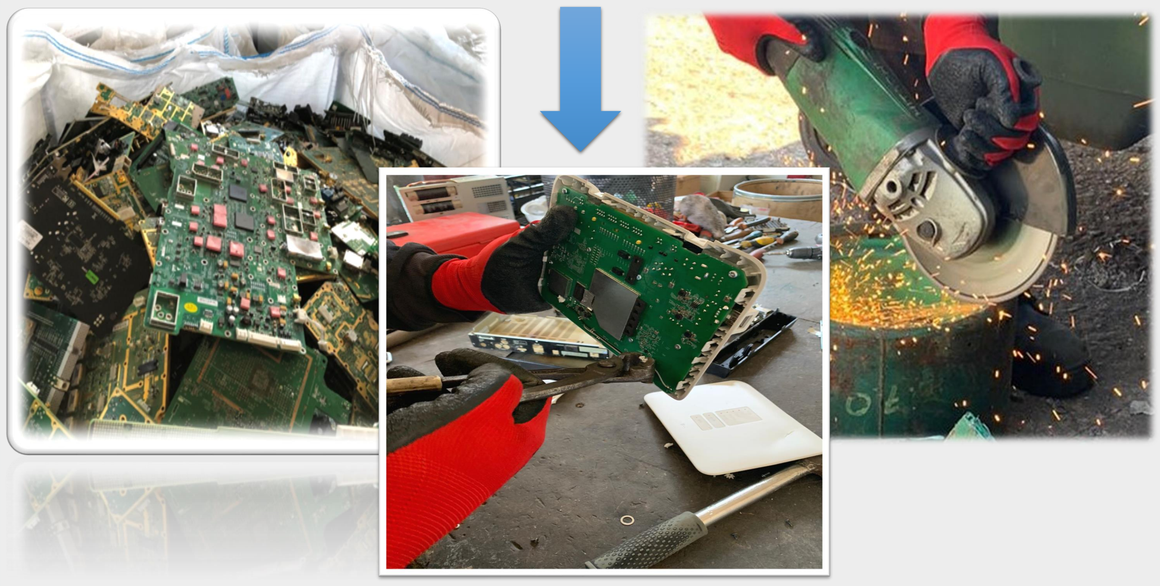
- **Waste from Electrical and Electronic Equipment (WEEE) or E-waste as one of the world's rapidly growing waste streams.**
- **Dust generated during e-waste dismantling activities constitutes a significant source of various hazardous compounds (such as persistent organic pollutants (POPs) or toxic metal(oid)s) to which workers or residents living near e-waste recycling facilities could be constantly exposed.**
- **The ban of commonly used brominated flame retardants (BFRs) by the Stockholm Convention on POPs resulted in an increase in the use of their alternatives, such as organophosphate esters.**
- **Both organophosphate flame retardants (OPFRs) and toxic metal(oid)s are identified as endocrine disrupting chemicals (EDCs).**
- **Considering their simultaneous presence in the workplace and living environment, additional investigation is needed to asses the adverse effects on human health due to the combined exposure to low doses of their mixture.**

The current *in silico* toxicogenomics data mining approach aims to asses the linkage between the exposure to toxic fumes from e-waste recycling activities consisting of environmentally relevant toxic mixture (OPFRs and toxic metal(oid)s), and endocrine system diseases (ESD) and to reveal the possible molecular mechanisms underlying endocrine toxicity.

Material and methods

- **The Comparative Toxicogenomics Database (CTD, http://CTD.mdibl.org) as the main data mining source for assessing genes linked to investigated toxic mixture and involved in endocrine system diseases development.**
- **GeneMANIA prediction server (https://genemania.org) for constructing tight networks of genes interactions affected by the exposure to the mixture of toxic substances.**
- **ToppGene Suite portal (https://toppgene.cchmc.org) for gene ontology (GO) enrichment analysis.**
- **GeneCards database (https://www.genecards.org) for obtaining functional information about the predicted genes.**

E-waste dismantling activities as a source of exposure to an environmentally relevant mixture of toxic substances (Pb, Cd, As, Hg, TPP and TDCPP)



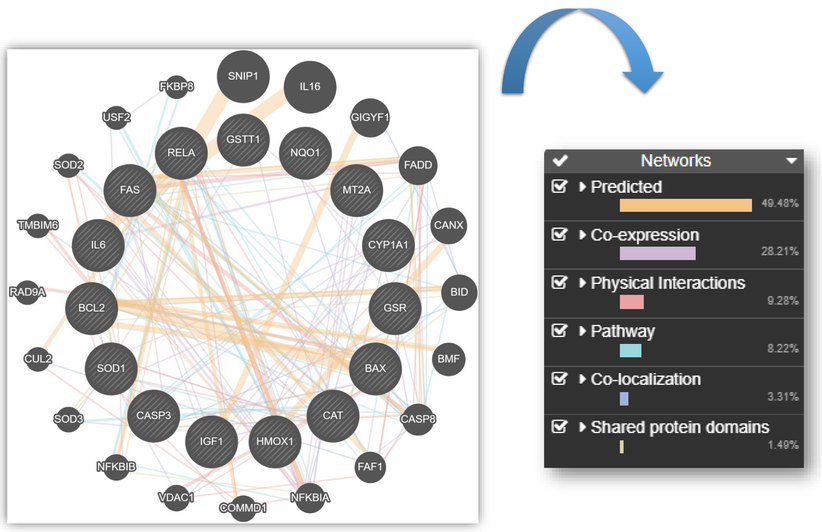


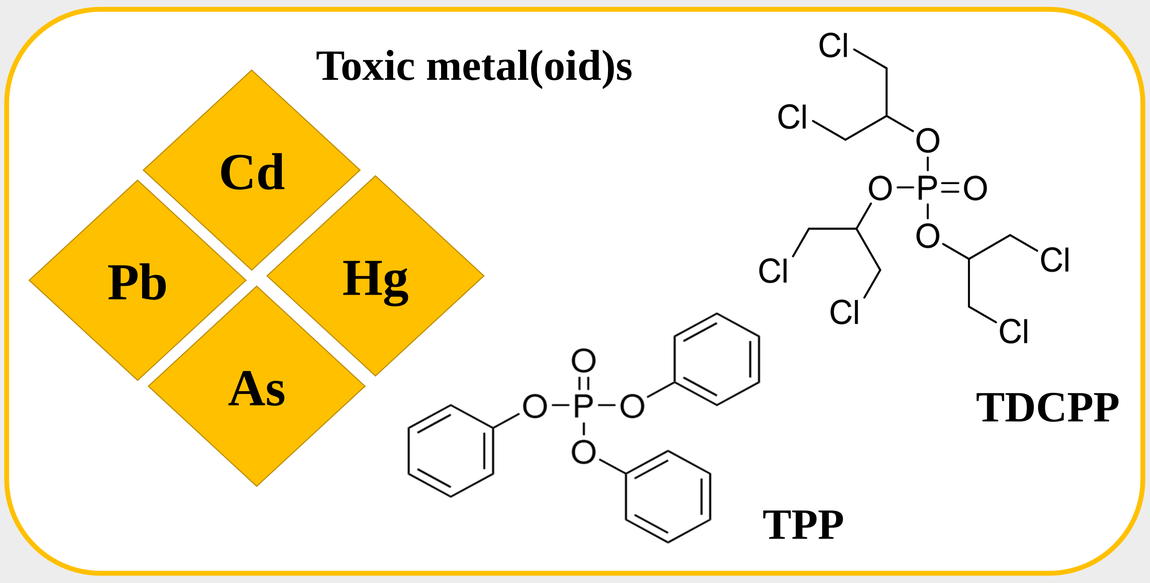
Figure 1. Constructed network between gene set linked to investigated mixture (GeneMANIA prediction server)

Gene	Interaction type common to all six substances	Degree
BAX	expression of BAX protein*	↑
BCL2	expression of BCL2 protein*	↓
CASP3	activity of CASP3 protein*	↑
CAT	activity of CAT protein*	↑
CYP1A1	expression of CYP1A1 mRNA*	↑
IL6	expression of IL6 mRNA*	↑
HMOX1	expression of HMOX1 mRNA*	↑ ↓
NQO1	expression of NQO1 mRNA*	↑

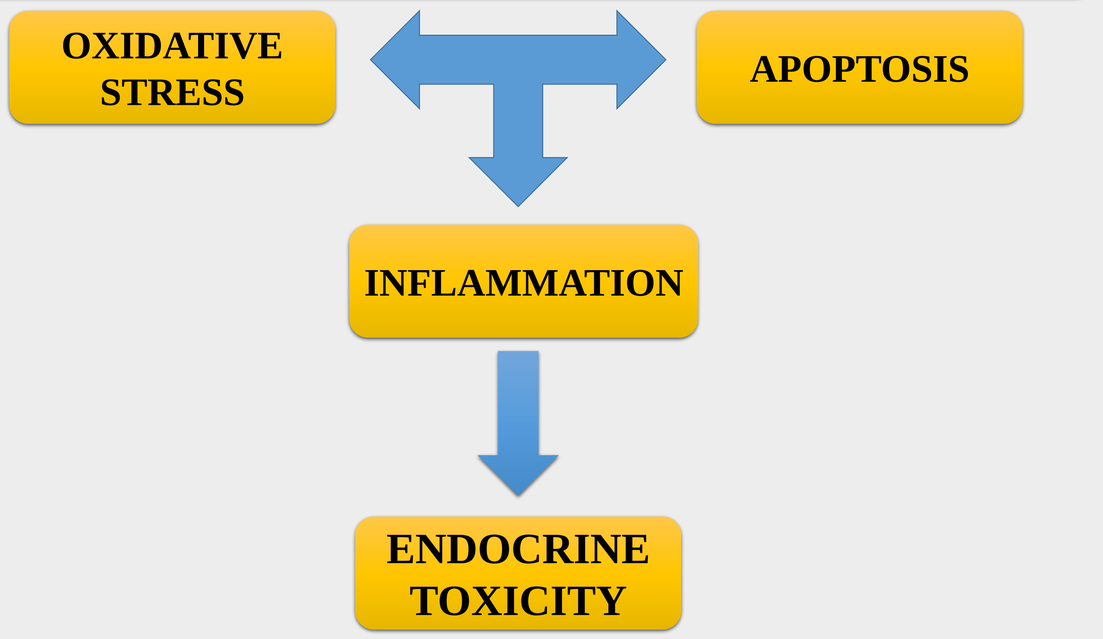
Table 1. 8 genes that interact with all six substances and share common interactions (CTD database)

Conclusions

- **Exposure to a mixture of chemicals during e-waste recycling activities could be linked to endocrine disorders through interconnected mechanisms of oxidative stress, apoptosis and inflammation.**
- **Extracted genes as possible genomic biomarkers of OPFRs and toxic metal(oid)s mixture endocrine toxicity.**
- **Current research as an introduction to further *in vivo* and *in vitro* experiments for enhancing the comprehension of molecular mechanisms associated with exposure to a specific mixture of environmental chemicals.**



Potential molecular mechanisms involved in the mixture toxicity



Results

- **CTD analysis indicated that all six substances present in the mixture act on 24 mutual genes.**
- **Investigated toxic mixture interacted with 15 genes related to endocrine diseases (BAX, BCL2, CASP3, CAT, CYP1A1, FAS, GSR, GSTT1, HMOX1, IGF1, IL6, MT2A, NQO1, RELA, SOD1).**
- **49.48% of the interactions among these genes potentially affected by the investigated mixture, along with 20 related genes, were predicted by the server (Figure 1.)**
- **From the provided GO (molecular functions and biological processes) and pathway terms related to endocrine disorders and the investigated mixture of toxic substances, oxidative stress, apoptosis and inflamation were found to be the most prominent mechanisms.**
- **The analysis has revealed 8 genes with matching interactions for all six substances present in the mixture (Table 1.).**
- **Finally, our phenotype analysis suggested the most pronounced effects on the reproductive tissues in both male and female, as well as on the thyroid.**

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