Abstract #668 | Oral or Poster presentation

A systematic review of control compounds tested for validation of Drug-Induced Liver Injury (DILI) *in vitro* models

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Background: Idiosyncratic drug-induced liver injury (DILI) is a complex and unpredictable event caused by different drugs, herbs, and dietary supplements[1]. The early identification of human hepatotoxicity at the preclinical stages stands as a challenge, in which the selection of validated *in vitro* systems has a big impact[2]. Ideally, new DILI predictive models should be tested with a broad battery of drugs, including those with high hepatotoxic risk and those with no hepatotoxic potential. This systematic review aims to establish a unified list of DILI positive and negative control drugs, supported by bibliographic and clinical evidence and endorsed by a committee of experts from the ProEuroDILI Network (CA 17112).

Methods: This systematic review was performed in accordance with the 2020 PRISMA guidelines[3]. Included articles were original research focused on investigating DILI occurrence using *in vitro* human models and conducting at least one hepatotoxicity assay with positive and negative control compounds. A modified version of the "Toxicological data Reliability Assessment Tool" (ToxRTool) was used to assess the bias of the included studies. A deeper analysis of the mostly used DILI-positive and negative drugs found in the literature was performed.

Results: A total of 2936 studies were retrieved from the different databases. Of these, 2885 studies were excluded after screening (duplicates or did not meet the inclusion criteria). Ultimately, 51 articles were included. Among them, 30 were categorized as reliable without restrictions. Diclofenac and buspirone were the most commonly used DILI-positive and negative control drugs. In fact, although there is a broad consensus on the positive compounds, the selection of negative control compounds remains less clear, as up to 19% of these have reported clinical hepatotoxicity cases in various DILI registries. This is primarily due to the subjective nature of determining the cut-off point for positive and negative drugs. Regarding the models used, there is a wide range of cell types and spatial configurations, with the 2D monoculture of human primary hepatocytes being the favored choice. As for the concentrations, a clear *consensus* was not found, since although many articles use the C_{max} values as a basis, these also vary between articles. Short exposure times (≤ 72h) were mostly utilized, and cytotoxicity was the preferred endpoint.

Conclusion: This systematic review has highlighted the lack of agreement in *in vitro* modeling of DILI. As a universal test for the complex DILI process remains not possible, it is necessary to establish a range of well-characterized DILI predictive platforms. Therefore, a list of 10 positive and negative drugs has been

developed for validating new in vitro models. Moreover, it is essential to come to an agreement on the reference drugs to be used for DILI assay validations, as well as the concentration range to be tested and the criteria for data interpretation.

Affix

References

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