Clopidogrel Toxicity Measuring by Flow cytometry: A pilot study in HepG2 cell line

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INTRODUCTION: Mortality and morbidity in acute coronary syndromes (ACSs) are mainly caused by plaque erosion or rupture, leading to thrombus formation. The thienopyridines, in particular the prodrug clopidogrel, are currently used to prevent thrombotic events in ACS patients. However, a large number of patients taking clopidogrel have shown resistance to treatment, leading to lower therapeutic response and toxicity. **OBJECTIVE**: The aim is to investigate aspects of clopidogrel cytotoxicity in hepatocellular carcinoma cells (HepG2). **MATERIAL AND METHODS**: The human HepG2 cells were cultured in Roswell Park Memorial Institute medium (RPMI) containing 5% Exosome-Depleted Fetal Bovine Serum and supplemented with penicillin (10.000 UI/mL), streptomycin (10.000 UI/mL), and sodium bicarbonate (44 mmols/L) at 37°C in 5% CO_2 air. Clopidogrel treatments were performed during 24 and 48 h using the concentrations of 6.25, 12.5, 25, 50, and 100 µM. The cytotoxicity was evaluated in triplicates by flow cytometry, using the propidium iodide (PI) staining protocol (50 µg/mL) to analyze DNA fragmentation. **RESULTS AND DISCUSSION:** Our results revealed that in both periods of treatment the concentrations of 6.25 e 12.5 µM had similar profile that observed for the control cells. In relation to other concentrations (25, 50, and 100 µM) it was observed a dose-dependent increase in DNA fragmentation. **CONCLUSIONS:** In conclusion, the results indicate that the concentration of 12.5 µM is close to the sub toxic dose for HepG2 cells. This definition is important to design new experiments involving the study of clopidogrel toxicity. Alternatively, the results indicate the safety range of clopidogrel concentrations that can be used when toxicity is not desired.

Keywords: Clopidogrel toxicity; HepG2 cells; Flow cytometry

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