Elucidation of the mechanism of amiodarone-induced immune related adverse reaction and search for biomarkers to predict its pathogenesis

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Background
Although amiodarone is a benzofuran derivative widely used to treat arrhythmias, its use is limited by adverse reactions. There is evidence that some of the severe adverse reactions such as liver injury and interstitial lung disease are immune-mediated; however, details of the mechanism have not been elucidated. We tested the ability of amiodarone to induce the release of danger-associated molecular patterns (DAMPs) that activate inflammasomes.

Methods
Human hepatocarcinoma functional liver cell-4 (FLC-4) cells were cultured with amiodarone or dronedarone for 7 days, and then the supernatant was added to differentiated THP-1 cells and incubated for 24 hr. The control was incubation without drugs. IL-1β concentration in the THP-1 culture medium was measured using an ELISA kit. Caspase-1 activity was also measured using the Caspase-Glo® 1 Inflammasome Assay. DAMPs were evaluated by western blotting using the hepatocyte supernatant.

Results
Amiodarone is known to be oxidized to reactive quinone metabolites. The supernatant from the incubation of amiodarone with FLC-4 cells for 7 days increased caspase-1 activity and production of IL-1ß by THP-1 cells. In the supernatant of FLC-4 cells with amiodarone, the heat shock protein (HSP) 40 was significantly increased. Addition of a cytochrome P450 inhibitor to the FLC-4 cells prevented the release of HSP40 from the FLC-4 cells and activation of THP-1 inflammasomes by the FLC-4 supernatant.

Conclusions
These results suggested that the reactive quinone metabolites of amiodarone can cause the release of DAMPs from hepatocytes which can activate inflammasomes. Dronedarone, a safer analog of amiodarone, did not activate inflammasomes. Inflammasome activation may be an important step in the activation of the immune system by amiodarone, which in some patients, can cause immune-related adverse events. Detected HSPs may be predictive biomarkers of adverse effects.

Key Words
Amiodarone, Dronedarone, Inflammasome activation, Hepatocytes, Immune-related adverse events