Mitochondrial cardiotoxicity of a prototype HCV NS3-protease inhibitor is characterized by a specific electrocardiographic signature in mice.

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Introduction: Preclinical cardiotoxicity has been observed for at least 3 different HCV NS3 protease inhibitors. We previously described the ultrarapid clinical and ultrastructural mitochondrial cardiotoxicity after 4 doses of BILN2061 in mice. As proteases are important in the biogenesis of mitochondria, a class toxicity of NS3 protease inhibitors can be feared.

Methods: Single and multiple dose BILN2061 toxicity studies were performed at 30 and 150 mg/kg BID intraperitoneally for 7 days in several mouse strains. Vehicle-dosed animals were used as controls. Peak and trough plasma levels were determined with a HPLC-UV method. The ultrastructural and functional integrity of cardiomyocytes was analysed with electron microscopy and surface electrocardiography. The activity of cardiac mitochondrial oxidative phosphorylation complexes was determined with Blue Native polyacrylamide gel electrophoresis and confirmed with spectrophotometry.

Results: BILN2061 at 150 mg/kg BID is 100% lethal within 2 days in all mouse strains tested, while 30 mg/kg BID induced precocious deaths in 1/6 Balb/c and 2/6 uPA-transgenic mice. Apart from a non-persistent clinical inactivity, the remaining animals tolerated BILN2061-dosing well. Compared to previous oral dosing studies, plasma BILN2061 trough levels were similar after 30 mg/kg BID, but almost 20-fold higher after 150 mg/kg BID. Profound electrocardiographic changes were documented in the J-waves corresponding to the ventricular repolarisation in all

mice treated with 150mg/kg. In Balb/c (n=3) and SCID (n=4) mice an increase in the height and surface area of the J wave was observed, while in uPA-transgenic SCID mice (n=4) BILN2061 dosing induced an elongation of the J-wave without increasing its height. ECG-changes were apparent within 30 minutes after a single dose and recuperated after 6 hours. The 30mg/kg dose induced similar but less pronounced ECG-changes. In selected animals, heart samples showed similar ultrastructural mitochondrial defects as previously described. No apparent defects in the oxidative phosporylation activity could be documented. We are currently analyzing the ECG-kinetic changes with sequentially obtained plasma samples to determine a PK-PD threshold.
Conclusion: The HCV NS3-protease inhibitor BILN2061 induces mitochondrial cardiotoxicity in mice and is associated with specific, rapid but transient electrocardiograohic changes in the ventricular repolarisation wave. As other HCV NS3 inhibitors have been associated with preclinical cardiotoxicity, more extensive studies on the pathophysiological mechanisms are warranted to be able to early detect any class toxicity.