a next generation sequencing method. The acquired candidate aptamers were examined by the affinity check with target cardiomyocytes, other cardiomyocytes and fibroblasts. Using the acquired aptamers, we have successfully acquired the target cardiomyocytes from the mixture of the cells. These results proved an advantage compared with antibodies because the cell-surface-attached antibodies can’t be removed from the cell surface without any nuclease treatment nor physical stretching. They also suggest that this method may eventually facilitate cell network assay and transplantation of ES- or iPS-derived cardiomyocytes for regenerative medicine.

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0045

Toxicological evaluation of methanol extract of the seeds of Moringa oleifera
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The seeds of Moringa oleifera are used in rural areas of Africa to clarify turbid water for human and animal consumption, and are also used for medicinal purposes, even though the safety/toxicity of the seeds has not been established. The dried, pulverized seeds were subjected to cold extraction with methanol and the methanol extract was used for acute and sub-acute toxicity studies in rats. Although, signs of acute toxicity were observed at 5000 mg kg$^{-1}$ dose, in the acute toxicity test, and mortality was recorded at 5000 mg kg$^{-1}$, no adverse effect was observed at doses lower than 3000 mg kg$^{-1}$. The median lethal dose of the extract in rat was 3870.4 mg kg$^{-1}$. Sub-acute administration of the extract at 0, 400, 800 and 1600 mg kg$^{-1}$ to rats divided into control, groups 1, 2 and 3 respectively, caused a significant ($p < 0.05$) increase in the levels of alanine and aspartate transferases, and a significant ($p < 0.05$) decrease in the weight of experimental rats, at 1600 mg kg$^{-1}$. However, treatment with the extract did not significantly alter the levels of hemoglobin, red blood cell, packed cell volume, mean corpuscular volume and mean corpuscular hemoglobin concentration. Also, the levels of total protein, albumin and globulin in the treated animals were not significantly different from the control. The study concludes that the extract is safe for use as medicinal and water purifying agent.

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0046

Interpretation of in vitro pharmacological profiling data in preclinical safety assessment
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Secondary pharmacology studies are designed to investigate the mode of action and/or the effects of a compound not related to its intended therapeutic target. One method for this is in vitro pharmacological profiling, which involves testing compounds in panels of in vitro assays covering a diverse range of molecular target classes including receptors, ion channels, enzymes and transporters to identify specific molecular interactions that may cause adverse drug reactions. The outputs from profiling in these panels can be challenging to interpret due to the volume and complexity of the data. We will describe how the data can be visualised and interpreted for applied use in decision-making and prediction of the margins needed to avoid adverse drug reactions driven by the off target profile as part of an integrated risk assessment and in the patient and disease context. We will describe a series of important factors that must be considered when interpreting the data that range from basic pharmacological principles relating to molecular target occupancy, through to the assay protocol and technical factors that should be considered. Case studies from AstraZeneca programmes will be presented to illustrate these principles and highlight the importance of expert interpretation of in vitro pharmacological profiling data to enhance its relevance and impact in drug discovery and development in reducing safety-related attrition.

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0047

Correlation of cardiac biomarker levels with hemodynamic and histopathological changes in rats
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Evaluation of biomarkers related to cardiac injury is an important part of cardiovascular safety assessment of drug candidates. This study was conducted to clarify the relationship among five biomarkers (cTnI, cTnT, FABP3, MLC3, and NT-proBNP), hemodynamic parameters, and cardiac histopathology for 24 h in male CD (SD) rats (n = 4 for each group) treated with beta-agonist, isoproterenol (I: 0.5 mg/kg, i.p.), phosphodiesterase3 inhibitor, milrinone (M: 15 mg/kg, p.o.), or fluoroquinolone antibiotic, sparfloxacin (S: 100 mg/kg, p.o.). Blood samples for assessment of the biomarkers were collected at 1, 2, 6, and 24 h after dosing.

I and M increased the heart rate (max. + 40%) and decreased the systemic blood pressure (max. −20 mm Hg) for about 3 h to the same extent, and showed myocardial degeneration/necrosis and infiltration of inflammatory cells at 24 h after dosing. In the I-treated group, the cTn peak response typically occurred at a similar timing to the hemodynamic changes, and the concentration returned to control levels at 24 h after dosing. MLC3 and NT-proBNP showed the highest levels at 6 h after dosing, and the levels were maintained even at 24 h. In contrast, M did not show such time-course of changes in the biomarkers. S induced neither the hemodynamic, histopathological nor biomarker changes.

Taken together, these results suggest that drugs with similar hemodynamic and histopathological changes do not necessarily show the same time-course of changes in the biomarkers. Combined measurement of the cardiac biomarkers with hemodynamic and histopathological examinations can provide important information for overall cardiovascular safety assessment.

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0048

A new noninvasive biomarker of proarrhythmic risk, index of cardiac electrophysiological balance (iCEB) in myocardial ischemia and reperfusion and drug-induced long QT
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A new noninvasive biomarker of proarrhythmic risk, index of cardiac electrophysiological balance (iCEB) in myocardial ischemia and reperfusion and drug-induced long QT
We evaluated a new biomarker, iCEB, in the isolated rabbit hearts, in anesthetized dogs with myocardial ischemia (I) and reperfusion (R) and conscious dogs with drug-induced long QT, as an indicator of proarrhythmic risk. In the isolated rabbit hearts, subjected to global I (10 min) and R (20 min), iCEB was reduced from 7.9 ± 0.4 at baseline to 6.6 ± 0.5 at the end of R. Flecainide (10 μM) markedly reduced iCEB (3.9 ± 0.4) at the end of R. The marked reduction in iCEB was associated with high incidence of ventricular tachycardia (VT) (10/11 during I and 17/17 during R). In conscious dogs, dovetilide and JNJ-303 (I Ks blocker) significantly and dose-dependently increased iCEB and JNJ-303 (I Ks blocker) massively increased iCEB from 6.9 to a maximum of 14.3, which was associated with TdPs (5/6), whereas moxifloxacin only slightly increased iCEB from 4.1 to a maximum of 5.7 and did not induce TdPs.

Our data suggest that iCEB may be useful as a non-invasive biomarker to predict susceptibility to cardiac arrhythmias in pathological condition such as myocardial I/R and in drug-induced long QT.

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0049
Differential effects of a novel ino-dilator in conscious dogs with normal or dilated-cardiomyopathic ventricles: A look through left-ventricular pressure-volume analyses
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Reduction of myocardial loading with enhanced function is a primary target for the pharmacological treatment of heart failure (HF). However, the determination of true inotropic/lusitropic properties for vaso-active compounds remains equivocal. For instance, while nitroxyl (HNO) donors enhance LV function in patients with HF, the demonstration of these effects under common paradigms in normal animals is hindered by their potent vascular effects. This study compared the load-dependent and independent conscious cardiovascular responses to an HNO-donor (HNO+) in both normal and HF dogs. Dogs were chronically instrumented for arterial pressures (AP) and for LV pressure-volume (LVPV) recordings. A subset of animals had HF induced via chronic overdrive pacing (e.g., EF: 36 ± 2% and NT-proBNP > 2500 pM/L). Data were obtained before/during a HNO+ infusion (100 μg/kg/min); load-independent function was examined by LVPV relationships.

HNO+ reduced LV preload (EDV, −16 ± 3 vs. HF: −7 ± 2%), end-systolic (ESP, −18 ± 2 vs. HF: −17 ± 1%) and filling pressures (EDP, −12 ± 2 vs. HF: −21 ± 3%), without any effects in HR and dP/dtmax. In normal dogs, HNO+ decreased stroke volume (SV, −14 ± 3%), while arterial elastance (Ea: −3 ± 5%) and EF (3 ± 3%) were unchanged. In HF, both SV (+9 ± 1%) and EF (+18 ± 2%) increased, while Ea decreased (−24 ± 1%). In all cases, HNO+ increased load-independent inotropic indices (e.g., PRSW, +12 ± 2 vs. HF: +18 ± 2%).⁎ P < 0.05 vs. baseline

In conclusion, only the combined use of both load-independent indices of LV function and a clinically-relevant disease-model allowed the establishment of true pharmacological responses.

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0050
Translating the gap in the evaluation of drug-induced QTc-interval prolongation from in vivo to the clinic
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Background: Assessment of the propensity of non-antiarrhythmic drugs in prolonging QT/QTc interval is critical for the progression of compounds into clinical development. The objective of the current work is to demonstrate the advantages of a model-based approach to create a correlation factor between in vivo findings and QTc-interval prolongation in humans and to predict the clinical outcome using only the preclinical study with a Bayesian hierarchical model.

Methods: Pharmacokinetic and pharmacodynamic (ECG) data from experiments in conscious dogs and clinical trials following administration of nine compounds with QT shortening, borderline and QT prolonging effects were used to describe the relationship between QT prolongation and drug exposure, taking into account inter-individual differences in pharmacokinetics and other physiological factors known to alter QTc interval [1].

Results: Preliminary results reveal that in vivo protocols show clear differences in species sensitivity to QT-prolonging effects and in the estimates of drug potency. Furthermore, our approach enabled the estimation of drug- and system-specific parameters as well as the overall probability associated with QTc-interval prolongation > 10 ms compared between clinical and preclinical species. Based on this correlation dogs appear to be a suitable, but less sensitive species to the drug-induced QTc-prolonging effects, as compared to humans. This approach can be used to predict clinical outcome based on preclinical data.

Conclusions: A model-based and validated approach is critical for the evaluation of cardiovascular risk and provides a framework for translation of drug-induced effects on cardiac conductivity dogs to the clinic.

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0051
Inclusion of neurobehavioural and respiratory safety pharmacology endpoints in the four-week rat general toxicity studies: A CRO perspective
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