ToxComp - In Vitro-In Vivo Extrapolation System for Drug Proarrhythmic Potency Assessment

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Abstract

The project aims to develop the systems biology driven, modeling and simulation based, easy to use for non-modelers platform for the drugs proarrhythmic potency assessment at the population level. Platform realizes the in vitro – in vivo extrapolation approach thus the input data comes from the patch clamp in vitro ionic currents inhibition studies.

Covariates describing variability in population include cardiomyocyte, heart and plasma physiological parameters which are correlated with basic demographic information (age, gender).

Simulated output data includes action potential and ECG derivatives where APD50, APD90 and QTc are analyzed as the endpoints. System was implemented with use of the Java/JavaFX technology and is available both on line and off line and distributed under the GPLv3 license. Its main role include clinical trials optimization and waiving, helping with the go-no go decision making.

1. Introduction

In vitro – in vivo extrapolation is a relatively well known concept more and more widely utilized in the drug development area. Drugs pharmacokinetics (PK) prediction is a field where IVIVE concept combined together with physiologically-based pharmacokinetic (PBPK) modelling under the umbrella of systems biology is used with success, where the success is defined both as a the drugs fate in the human body assessment as well as ability for the mechanistic insight into the pharmacokinetics processes [1]. Mature methodology, flexible and constantly curated models together with available software makes the IVIVE techniques natural continuation of the in vitro wet lab studies allowing i.e. for the laboratory animals use optimization. What is even more important predicted endpoints describe human situation. Use of the population approach allows for the inter-individual variability assessment. To apply for the latter one detailed description of the human physiology is needed. What is worth noted more and more sources with freely available databases for the physiologically based pharmacokinetic model become available [2].

Although well established in the pharmacokinetics, in vitro-in vivo extrapolation approach is still fighting for its place in the pharmacodynamics field. There are some examples which prove that such approach can be fruitfully used as for example the DILI-sim initiative which aims in the development of a liver metabolism model which could be used by the pharmaceutical industry to evaluate the plausibility of drug-induced liver injury across multiple metabolic pathways.

As the drug cardiotoxicity is one of the main reasons for the promiscuous compounds removal at various levels of development as well as marketed drugs withdrawal, there is a need for development a reliable system for early toxicity prediction.

Our project aims in a development of a systems biology driven, modelling and simulation based, easy to use for non-modellers platform for the drugs proarrhythmic potency assessment at the population level.

2. Main assumptions

There are some major assumptions taken as a background for the platform development. The system realizes the in vitro – in vivo extrapolation approach thus by default the input data comes from the in vitro ionic currents inhibition studies. The above mentioned extrapolation process aims at the prediction of the human situation thus the models utilized describe human physiology. System allows for the inter-individual variability assessment of the simulated effect.

3. Elements of the system

System consists of a few main elements as presented below in Figure 1. All of them are further described is the subsequent paragraphs.

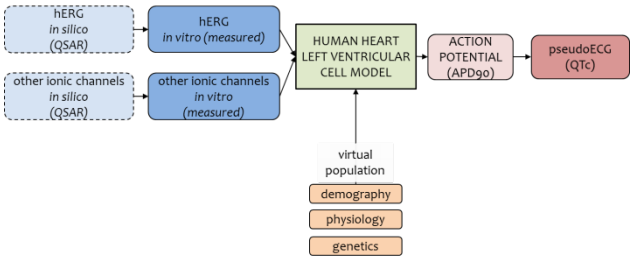

Fig.1. ToxComp system scheme – current status.

3.1. Human cardiomyocyte model

Model of the physiology of human heart left ventricular cell is a core of a system. Among all available models we have chosen one proposed by ten Tusscher (TNNP model) [4]. The main reason lying behind such decision was listed above as one of the assumptions, namely extrapolation to the human situation. According to the results of the cardiomyocyte models metaanalysis published by Niederer, TNNP model was built to most degree based on the human data [5].

Apart from the single cell simulations system offers 1D fibre model paced at the endocardial side as the transmural heterogeneities in ionic currents between endocardial, midmyocardial and epicardial cells are incorporated. By default the distribution is 50:30:20 respectively but can be easily modified by user. The string length by default is 1.3 cm width what mimics the human heart wall. Such default average value proposed by various authors is probably not physiological what was proved by the results of an in house analysis of the available studies where heart wall thickness was measured in various locations and with various methods utilized (unpublished data). Based on that it was decided to implement a model describing the relation between age and gender and heart wall thickness [6]. It is an element of the virtual population generator. The diffusion coefficient is by default set to 0.0016 cm^2/ms as proposed by various authors. Such value can be modified by user according to his/her data.

Forward Euler method is used to integrate model equations. The output for the single cell simulation is an action potential describing the membrane dynamics. For a one-dimensional string of cells the results are used to calculate a pseudo-ECG. A space step and a time step are by default set to $\Delta x=0.01$ mm and $\Delta t=0.01$ ms.

3.2. Ionic currents

All main ionic currents are mathematically described as proposed by ten Tusscher without modifications. Five of them namely IKr, IKs, Ito, INa and ICaL can be modified by the drugs accordingly to the user-provided data which describes the concentration dependent ionic current inhibition (0-100%) by simple multiplying by the inhibition factor (0-1). System allows for up to five drugs simulation at the same time. All inhibition effects are summed up.

The results of the previous study proved that the in vitro research settings have significant impact on the final channel inhibition potency [8]. The default combination of the cell line and measurement temperature which was assumed as closest to the human conditions was defined as HEK (Human Kidney Cell) in a physiological temperature. Inter-system extrapolation factors were prepared for those of users who perform their in vitro studies in other conditions (various cell lines and room temperature).

3.3. Extended QSAR models

ToxComp system can be used at the early stage of the drug development as a screening tool even in the situation when no in vitro data are available. In such situation three independent models are available for the in vitro channels inhibition prediction. All of the models for the IKr, IKs and ICaL currents were developed with use of the artificial neural networks as the leading algorithm and extended QSAR methodology was applied. The latter one includes the in vitro research settings embedded into the single model together with the molecular descriptors of the chemical entity of the interest. Based on the results of sensitivity analysis, each model for the particular ionic channel was built on the individual set of the input parameters.

3.4. Virtual population generator

Apart from the previously mentioned heart wall thickness there is a list of other parameters describing human physiology which are elements of the model. Their use allows for the population variability assessment. Some of them are currently correlated with the demographic parameters including age and gender as presented in table below (Table 1).

It is noteworthy that parameters which are currently not correlated with the demographic parameters still offer the capability of the variability mimicking. Final values are randomly picked up from the distribution which parameters were set up based on the in house available data.

Heart rate model was developed based on the data derived from the publicly available sources and validated with use of the in house data derived from the cardiology clinic database [11]. Apart from the age and gender correlation it also considers the circadian variation.

Genetic variability is simulated by combination of the population data describing the polymorphism/mutation frequency and channel activity change defined by the modification of the gating parameters.

Table 1. List of physiological parameters.

Physiological parameter	Independent parameters	Source
Cardiomyocyte volume	Age	[9]
Cardiomyocyte electric capacitance	Age, Cardiomyocyte area	[9,10]
Sarcoplasmic reticulum volume	-	-
Left ventricle free wall thickness	Age, gender	[6]
Heart rate	Age, gender	[in-house data]
K^+ plasma concentration	-	[in-house data]
Na^+ plasma concentration	-	[in-house data]
Ca^{2+} plasma concentration	-	[in-house data]

3.5. Output

The output has a form of a text file with data separated with comma (.csv file). It allows both easy handling and further analysis in various available systems of users choice including spreadsheets. The results file is structured in the way to allow for easy simulation replication and offers detailed information about every virtual individual involved in the simulation study.

4. Programming language

Application architecture was composed of two languages, Java and JavaFX, based on the common runtime environment. The choice of Java language seems to be a reasonable compromise between capacity and performance. The standard edition was used to create the entire application logic in the form of shared logic modules, distributed as JAR files. Java also helped to create a clear and logical model of the application which, in accordance with the fundamental architectural principles of language defined as loose coupling and tight cohesion. Individual libraries works with the model structure, while keeping the autonomy and the ability to replace them. The choice of Java was supported by the fact of advanced possibilities of multithreaded, available from 1.5 version. Support for advanced calculation has been optimized using the thread pool architecture, and non-blocking collections available in standard language library like ConcurrentLinkedQueue and others. These solutions allow for optimal use, more and more popular multi-core machines, while minimizing delays resulting from the use of explicit synchronized block, which are typical for well-known solutions based on the mutual exclusions.

The core of the application, prepared in the Java SE, is available by the user interface based on the JavaFX language. The use of scripting dialect JavaFX language in version 1.31 helped to avoid complex architectural solution known from Swing library, mainly due to binding capabilities previously used in simple scripting languages. The major problem is the "publish - subscribe" model used in GUI event handling, usually handled by Observer design pattern, it can cause a lot of problems in the construction of complex applications and lead to memory leaks.

Direct and robust syntax, based on a declarative model, allows for the rapid development of user interface which, thanks to provided runtime environment, can be run in variety of client types, from desktop system operation, to web browsers and tv interfaces. However, despite promising technology, JavaFX as language in version 1.31 was abandoned which is associated with limited availability of runtime environments.

5. Availability and licensing

Stable version 1.2 is currently available at the www.tox-comp.net webpage for download and live run [7]. Platform is freely available and distributed under the GNH GPLv3 license. ToxComp system has been downloaded and used by the scientists and researchers from large pharmaceutical companies and leading academic groups around the world.

6. Future development plans

ToxComp system is constantly curated and new functional elements are being developed. The enhancement pathways include two directions namely human left ventricular cardiomycocyte model and physiological parameters. The first one includes more detailed description of the ionic currents with use of Markov models what will allow for the simulation of the drugs affinity to the channels. The latter one comprise of further development of the circadian rhythms for crucial ions.

It is also planned to couple excitation and contraction models to allow for the electro-mechanical window inspection after the drug dose intake.

7. Conclusions

Cardiotoxic effect is one of the main reasons for the drug market withdrawals thus has to be closely investigated at various levels. We hereby propose an easy-to-use system, based on the well-known and accepted models with easy to use graphic user interface allowing its effective use by non-modellers (Figure 2).

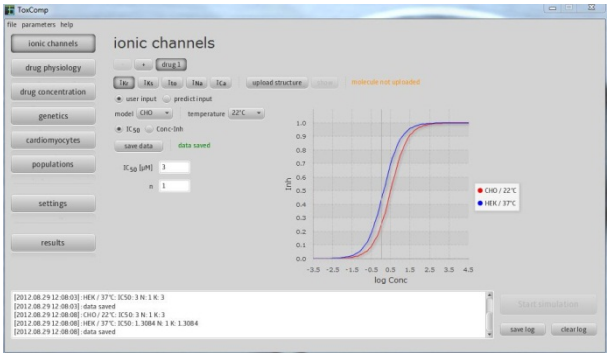

Fig.2. ToxComp graphic user interface.

Modelling and simulation approach utilized in the IVIVE platform ToxComp enables early screening of the chemicals proarrhythmic potency. Its main role include clinical trials optimization and waiving, helping with the go-no go decision making.

Acknowledgements

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References

[1] Rostami-Hodjegan A. Physiologically-Based Pharmacokinetics Joined with In Vitro-In Vivo Extrapolation of ADME: A Marriage under the Arch of Systems Pharmacology. Clin Pharmacol Ther 2012;92(1):50-61.

[2] Thompson CD, Johns DO, Sonawane B, Barton HA, Hattis D, Tardif R, Krishnan K. Database for physiologically based pharmacokinetic (PBPK) modeling: physiological data for healthy and health-impaired elderly. J Toxicol Environ Health B 2009;12:1-24.

[3] http://www.thehamner.org/institutes-centers/institute-for-drug-safety-sciences/dili-sim-initiative/

[4] ten Tusscher KH, Noble D, Noble PJ, Panfilov AV. A model for human ventricular tissue. Am J Physiol Heart Circ Physiol 2004;286(4):H1573-H1589.

[5] Niederer SA, Fink M, Noble D, Smith NP. A meta-analysis of cardiac electrophysiology computational models. Exp Physiol 2009;94(5):486-495.

[6] Sjögren AL. Left ventricular wall thickness determined by ultrasound in 100 subjects without heart disease. Chest 1971;60(4):341-346.

[7] www.tox-comp.net

[8] Wisniowska B, Polak S. hERG in vitro interchange factors - development and verification. Toxicol Mech Methods 2009;19(4):278-284.

[9] Polak S, Fijorek K, Glinka A, Wisniowska B, Mendyk A. Virtual population generator for human cardiomyocytes parameters. In silico drug cardiotoxicity assessment. Toxicol Mech Methods 2012;22(1):31-40.

[10] Polak S, Fijorek K. Inter-individual variability in the pre-clinical drug cardiotoxic safety assessment - analysis of the age - cardiomyocytes electric capacitance dependence. J Cardiovasc Transl Res 2012;5(3):321-332.

[11] Goldberger AL, Amaral LAN, Glass L, Hausdorff JM, Ivanov PCh, Mark RG, Mietus JE, Moody GB, Peng C-K, Stanley HE. PhysioBank, PhysioToolkit, and PhysioNet: Components of a New Research Resource for Complex Physiologic Signals. Circulation 2000;101(23):e215-e220.

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