Systems biology to battle vascular disease

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Vascular disease (VD) whether manifested as atherosclerosis or arteriosclerosis in cerebrovascular, coronary or peripheral artery disease, represents the major burden of the health systems due to the high prevalence and is the main cause of mortality in the male and female populations of the European Union. In addition, VD accounts for high costs both on a per capita basis and on a total basis and financial consequences are estimated to just under 110 billion € per year (http://www.heartstats.org). The correlation of the different final stages of VD with genetic variants has been noted in several studies (e.g. [1;2]). VD with its micro- and macrovascular manifestations is especially prominent and especially accelerated in chronic kidney disease (CKD) [3] and diabetes mellitus [4]. Both CKD and diabetes are characterized by the highest morbidity and mortality rates due to VD of any cohort studied and the role of VD has only recently been appreciated in early stages of CKD and diabetes. VD and pre-disposing factors for VD have further been associated with human aging and accelerated telomere shortening [5]. There is increasing evidence that telomere dysfunction and DNA damage accumulation contributes to the development of VD [6;7]. Unfortunately, current understanding of the pathophysiology of the micro- and macrovascular complications, especially of the early events responsible for disease onset and progression, is still limited mainly because of its enormous complexity, its polygenic and multifactorial traits. As a consequence, the therapeutic options are still limited. Detailed knowledge of the underlying biology and initial molecular pathophysiological events will enable early detection, definition of new and more appropriate therapeutic targets, and subsequently provide specific targeted therapies.

Patients with CKD are of special interest, as their pronounced and accelerated VD may serve as a model to elucidate its general pathogenesis. VD has been attributed to increases of oxidative and glycemic stress, microinflammation, endothelial dysfunction, sympathetic and renin-angiotensin overactivity. Furthermore, CKD would present an interesting model to study VD as decreased renal function combines (*i*) accumulation or substances which physiologically undergo renal excretion, (*ii*) reduced synthesis of active substances and (iii) reduced renal metabolism of substances. Studies on the pathogenesis of VD in CKD have predominately focused on individual factors and mechanisms. However, it is most unlikely that single factors of mechanisms account for all there is to VD in CKD and in general. A huge amount of data on VD is available, but still mostly exist as specific observation in particular cohorts or as genomics, proteomics/peptidomics, or metabonomics analyses on selected samples (e.g. [8;9]). These data are by no means complete yet, and need to be combined. For example, longitudinal and cross-sectional cohorts of patients with VD are available (e.g. [10;11]) but are currently only explored as single cohorts. Other, though disparate, data support the hypothesis that disturbance in the homeostasis of the extracellular matrix (ECM) are among the main and early significant changes in both micro- and macrovascular complications but systematic analysis has never been performed to clearly pinpoint the modifications of the ECM at an early stage in VD [12]. Therefore, we are certain that these existing data on VD, based both on human studies and animal- and cellular models, and the additional targeted gathering of “omics” data in clearly identified VD in conjunction to the recent advancements in Systems Biology analysis now provide the unprecedented opportunity to model the onset and progression of vascular events in diabetes and CKD.

The Systems Biology based modelling of sequence of events in VD will rely on the iterative cycle of “system model building, hypothesis generation, experiment, model refinement, hypothesis refinement“, using both top-down and bottom-up systems models. Mechanistic mathematical models (top-down) are based on quantitative biochemical data at the transcript, protein and metabolite level, where known or posited system structures are explicitly employed. On the other hand, probabilistic models (bottom-up), by statistically representing whole-genome data, enable inferences to be made regarding plausible structures for mechanistic models and constrain the feasible range of parameter space, thus extending the applicability of top-down models to larger realistic systems. The interplay between top-down knowledge-driven modeling and bottom-up data-driven modeling to feed the iterative cycle relies upon the supply of high quality and multi-source "omics" and classical (pathophysiological, phenotypical etc) data (Figure 1). Several recent reports clearly demonstrate the high quality of such datasets (e.g. [11;13;14]), hence the technical requirements for the application of Systems Biology towards the definition of the molecular chain of events involved in VD are apparently met.

To characterize the causal chain of events and identify its mediators that may be useful as surrogate biomarkers, an integrative approach combining heterogeneous experimental data, existing prior knowledge and complex systems modelling presents itself as a valid route to deciphering pathophysiology on a molecular level (Figure 1). A tight cross-talk between experiment and modelling is needed to allow identification of relevant molecular pathophysiological mechanisms, also with respect to timing of events and unearthing potential biomarkers and therapeutics targets in vascular lesions in diabetes and CKD. A possible way forward is graphically depicted in Figure 1.

The results of these efforts will be numerous and of expected major impact on the VD clinical management:

a) Computational models of the VD-associated (patho)physiological changes will be established. These models, along with data driven statistical models, will allow displaying the timely correlation of relevant pathophysiological events. As a consequence, valid disease biomarkers and biomarker panels will be identified for early disease detection when no clinically relevant events have yet been registered. Further, these models and biomarkers will enable prognosis of disease development, and facilitate initiation of appropriate (pre-emptive) therapeutic measures, that prevent or delay development of clinically relevant disease. This will also enable stratification of patients based on prognostic biomarkers and establishing of valid mediators as surrogate endpoints, resulting in shorter clinical trials that reach statistical significance based on a lower number of participants.
b) The establishment of an algorithm (similar to the Framingham score) is further expected. This will be based on molecular and classical clinical and laboratory data alike, and will enable the non-invasive assessment and prognosis of development of vascular disease with much higher accuracy than currently possible.
c) Another major result based on the knowledge on molecular changes and networks of these in development and progression of micro- and macrovascular diseases, will be the discovery of new, more appropriate potential targets for individual targeted therapeutic interventions. Upon validation of this newly gathered information in animal and cell culture models, interference studies will be performed aiming at observation if interference/intervention does influence disease and test whether the identified molecular structures represent therapeutic targets. The new validated biomarkers and animal models will result in improvements in clinical intervention trials by enabling preclinical testing in relevant animal models based on biomarkers relevant in human disease. In

parallel analyzing the changes in proteome/metabolome (and transcriptome, if applicable) that are induced by the interfering agents, will enable the development of useful surrogate markers for clinical endpoints.

Based on these thoughts, it appears almost mandatory that a Systems Biology driven approach, as a concerted effort of clinicians, biologists, and systems biologists is being initiated to battle vascular disease. Such a holistic approach integrating massive molecular, clinical, pathological data in combination with novel biostatistical, informatics/computational and bioinformatics approaches will have a major impact on the management of VD and will pave the way for individualized targeted therapy.

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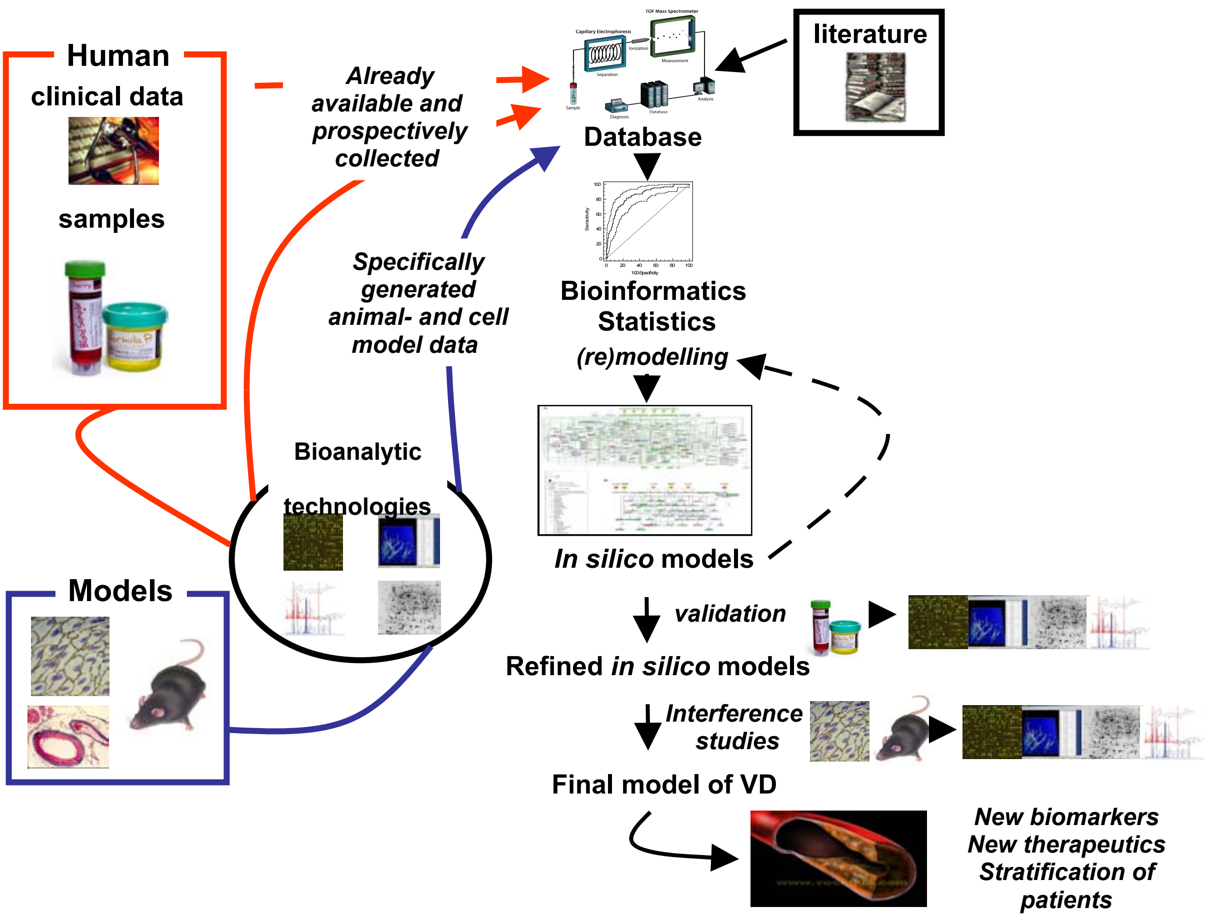
Figure 1 legend: Existing disparate data (clinical, –omics and literature) of human VD are being identified and combined, allowing the construction of a VD database. This will lead to the identification of missing data that subsequently can be prospectively collected. For example, it is anticipated that the existing datasets contain little data on the early events on VD. In parallel, –omics data on animal- and cellular models of VD will be collected to determine to which extent the processes are similar in humans and in these models. This in turn will allow us to progress with appropriate interference studies for validation of pharmacological interventions. All data will be analysed with appropriate bioinformatics and statistics, fed into the Systems Biology modelling process to produce the initial *in silico* model of VD. This initial model should be subsequently refined by the mandatory validation in human VD, followed by validation in animal and cellular models of VD disease. Multiple iterative steps will most be necessary to reach the appropriate final VD model generating new biomarkers, therapeutics and assessment of VD disease.


Human
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Bioinformatics
Statistics
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Specifically generated animal- and cell model data
Bioanalytic technologies
In silico models
Models
validation
Refined in silico models
Interference studies
Final model of VD
New biomarkers
New therapeutics
Stratification of patients