The application of multi-omics and systems biology to identify therapeutic targets in chronic kidney disease

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ABSTRACT

The quest for the ideal therapeutic target in chronic kidney disease (CKD) has been riddled with many obstacles stemming from the molecular complexity of the disease and its co-morbidities. Recent advances in omics technologies and the resulting amount of available data encompassing genomics, proteomics, peptidomics, transcriptomics and metabolomics has created an opportunity for integrating omics datasets to build a comprehensive and dynamic model of the molecular changes in CKD for the purpose of biomarker and drug discovery. This article reviews relevant concepts in omics data integration using systems biology, a mathematical modelling method that globally describes a biological system on the basis of its modules and the functional connections that govern their behaviour. The review describes key databases and bioinformatics tools, as well as the challenges and limitations of the current state of the art, along with practical application to CKD therapeutic target discovery. Moreover, it describes how systems biology and visualization tools can be used to generate clinically relevant molecular models with the capability to identify specific disease pathways, recognize key events in disease development and track disease progression.

Keywords: chronic kidney disease, data integration, omics, systems biology, therapeutic target

INTRODUCTION

Chronic kidney disease (CKD) constitutes a class of renal disorders diagnosed on the basis of progressive renal failure, which leads to characteristic pathological hallmarks of interstitial fibrosis and inflammation [1, 2]. CKD can also be non-progressive; moreover, diabetes, hypertension and ageing contribute as the primary causes, increasing cardiovascular disease risk [3]. The complex molecular nature of this disease with its co-morbidities has led to many failed major clinical trials, underscoring the limitation of approaches targeting signs and symptoms (e.g. inhibitors of the renin–angiotensin system targeting blood pressure cardiovascular mortality in CKD) [4] as opposed to targeting the underlying molecular causes of the disease (directly targeting affected cellular pathways). Therefore, there exists a great need for effective treatments for the progression of disease, and accurate, safe and non-invasive biomarkers capable of diagnosing and staging CKD are also needed as invaluable tools for tracking the success and predictive capability of potential treatments across diverse CKD-affected populations [5].

The multifaceted pathology as well as varied epidemiology of CKD creates challenges for drug and biomarker discovery approaches [6]. Clinically relevant translational models, such as mouse or rat animal models, do not fully recapitulate human disease pathways, and choices of disease models are restricted to one specific aspect of a disease rather than the full spectrum of pathophysiology [7]. Although insufficient for autonomous use in biomarker and drug discovery, these animal models nevertheless have tremendous utility in preclinical studies for suitable candidates. Imaging techniques lack the temporal resolution to capture functional kidney impairment at the earliest stages of disease; instead, they delineate structural changes such as fibrosis or inflammation that develop in the late stages of CKD, when therapeutic interventions are limited due to irreversible pathological damage and inevitable disease progression.
An ideal translatable methodology for drug and biomarker discovery should not only target clinically evident stages of disease, but focus on very early stages of disease when therapeutic interventions can still modify or stop disease progression.

The molecular map of diverse interactions, signalling and regulation within a cell can be elucidated using high-resolution analytical ‘omics’ platforms, encompassing genomics, proteomics, peptidomics, transcriptomics and metabolomics [9]. Aspects of this omics molecular interactome can be efficiently and non-invasively generated from patient samples (e.g. urine), at various stages of disease, providing the spatio-temporal resolution to track clinically relevant molecular changes with the progression of illness [10]. The availability of vast omics data in recent years has resulted in various applications and bioinformatics analysis tools, ranging from sophisticated databases [e.g. the Chronic Kidney Disease database (CKDdb)] to more advanced platforms with visualization features that aim to integrate omics together with information on biological pathways and interactions in a stepwise manner [e.g. Nephromine, Kidney and Urinary Pathway Knowledge Base (KUPKB)] [11]. ‘Stepwise’ generation implies the collection of each omics individually (and from multiple tissues, etc.) and then step-by-step analysis (these steps may be protein–protein interaction networks for the proteomics, then correlation to transcriptomics, and reconstruction of a metabolic network, etc.) and integration of all these data into a single and comprehensive interactome. While a stepwise generation of the entire molecular interactome is intractable, systems biology offers a practical solution—the integration of specific omics information within the scope of disease-associated biochemical pathways, thus ensuring clinical relevance of the data [12, 13].

This manuscript reviews relevant concepts in omics data integration using systems biology with practical application to CKD therapeutic target discovery.

**NEPHROLOGY OMICS RESOURCES AND LIMITATIONS**

Recent years have seen an increase of omics studies in a variety of disciplines, including cancer, cardiovascular disease and, of course, CKD [14–16]. The research paradigm shift to omics from other classical translatable approaches such as enzyme-linked immunosorbent assay (ELISA) and quantitative polymerase chain reaction (qPCR) was mostly spurred by the innovation in omics techniques, including various types of mass spectrometry (MS) [e.g. capillary electrophoresis–mass spectrometry (CEMS)] [17], microarrays and sequencing methods with the advent of more sensitive and precise instrumentation and the development of bioinformatics tools and algorithms, as well as computational platforms to handle such big datasets [18]. In nephrology, several databases have been developed to tackle these challenges, including CKDdb, KUPKB, GeneKid and Nephromine [11, 19]. While these data sources provide public access to various omics datasets and bioinformatics mining tools, none fully integrates the whole landscape of omics disciplines into a comprehensive, dynamic and visual model of cellular biochemistry and disease alterations [20]. In the last 5 years genomics, peptidomics and proteomics in kidney disease alone have generated >1000 scientific articles and transcriptomics and metabolomics in renal disorders about 500 publications each. In contrast, nephrology research utilizing ‘integrated or systems biology omics’ approaches resulted in just over 20 studies, highlighting the gap between analyses conducted in one area of omics as opposed to a unified contribution from several omics platforms, and such comprehensive examples are discussed in this review (Table 1).

Although advances in methodology and instrumentation have almost eliminated some of the technological obstacles in omics data integration (e.g. data quality and reproducibility due to laboratory protocol differences on sample preparation), there is still technical variability due to instrument detection thresholds and a lack of high-throughput methods for parallel sample measurements [9]. Additional remaining problems include data heterogeneity due to biological variability and inadequate cohort numbers, non-unique data annotation and poor curation and lack of standardization of various omics data types to allow for maximum compatibility [9]. The biggest hurdle appears to be the lack of ample algorithms for multidimensional data modelling to develop clinically relevant predictive models and proper elucidation of the disease signature [13, 28]. Longitudinal omics data are crucial for accurate modelling of disease progression, and various approaches for sample collection have been implemented for the most accurate data collection, even in the case of unexpected events, such as human error. The experimental capability of collecting samples at various disease stages is a strength of the omics methodology, however, it is still a challenge for omics computational analysis, and current published multi-omics studies do not address this extra level of complexity.

An example of a computational limitation can be specifically pinpointed in various transcriptomics studies that aim to recapitulate the full spectrum of disease phenotypes but only identify a general CKD signature; indeed, most omics studies are not based on personalized pathologies and therefore cannot model a specific CKD phenotype. Although there exist large-scale transcriptomics studies that look for composite phenotypic signatures for diseases [29], the algorithms utilized for such studies analyse the transcriptional differences between the phenotypes and are incapable of dividing the composite into individual phenotypes [30]. In large part this stems from the physiological heterogeneity of disease that lacks consistent correlation to molecular-level information. For example, in their transcriptome analysis of human diabetic kidney disease (DKD), Woroniecka et al. [26] found differential regulation of overlapping transcripts between the glomerular and tubular kidney compartments. The study supported previous findings of key pathways implicated in DKD [e.g. vascular endothelial growth factor (VEGF)] and discovered novel pathways (e.g. integrins), however, this differential regulation of inflammation and immune-related pathways varied upon repeat analysis and specific molecular pathways could not be assigned to a clinical phenotype. Such limitation in capturing the full spectrum of disease phenotypes is not a question of omics type or the specific analytical method used, but merely a question of study design. Although an experimental design may produce data that is too complex for phenotype modelling, and identifies only a general disease signature, as in the case of this particular
transcriptomics study [26], it is highly possible that by adding a drug molecule, consistent data could be collected with respect to the effect of the drug. Therefore, such studies appear better suited to drug repurposing or discovering generic phenotype signatures rather than examining the phenotype-specific biological mechanisms that are perturbed in a disease state [30].

**CONCEPTS IN OMICS INTEGRATION**

The systems biology approach to reconstructing biological processes aims to combine experimental data with computational modelling to elucidate the system as a whole (Figure 1). The stepwise integration of the dynamic network and the functions that govern its interactions can proceed in two ways: top down or bottom up. Bottom-up integration links known interactions of individual ‘modules’ to infer their pathway functions in order to fill in the gap between molecules and physiology; in contrast, top-down systems biology integrates the correlated molecular interactions from global omics studies to construct molecular networks and elucidate function [31]. An example for the bottom-up approach in CKD is the study by Song et al. [25], who investigated dysregulated pathways modulating renal cyst growth in autosomal dominant polycystic kidney disease.

**Table 1. Selected multi-omics studies implementing systems biology approaches**

<table>
<thead>
<tr>
<th>Study authors</th>
<th>Year</th>
<th>Approach</th>
<th>Systems biology relevance</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hamon et al.</td>
<td>2014</td>
<td>Bottom-up</td>
<td>Parameterization of the time course omics data from the Wilmes et al. study to elucidate the time-dependence of cyclosporine nephrotoxicity</td>
<td>[21]</td>
</tr>
<tr>
<td>He et al.</td>
<td>2008</td>
<td>Top-down</td>
<td>Integration of a meta-analysis of the glomerular transcriptome into a protein–protein interaction network in the glomerulus (GlomNET)</td>
<td>[22]</td>
</tr>
<tr>
<td>Husi et al.</td>
<td>2014</td>
<td>Combined</td>
<td>Comprehensive integration of human and animal proteomics and reconstruction of a metabolic network to elucidate molecular mechanisms in diabetic arteriopathy</td>
<td>[23]</td>
</tr>
<tr>
<td>Perco et al.</td>
<td>2010</td>
<td>Bottom-up</td>
<td>Integration of literature transcriptomics and proteomics datasets into a comprehensive molecular map of CKD (omicsNET)</td>
<td>[24]</td>
</tr>
<tr>
<td>Song et al.</td>
<td>2009</td>
<td>Bottom-up</td>
<td>Clinically relevant genomics-trancriptomics study identifying PKD1-dependent modulation of renal cyst development in ADPKD</td>
<td>[25]</td>
</tr>
<tr>
<td>Wilmes et al.</td>
<td>2013</td>
<td>Top-down</td>
<td>Extensive transcriptomics-proteomics-metabolomics study implementing pharmacokinetics to monitor nephrotoxicity in a renal cell model</td>
<td>[14]</td>
</tr>
<tr>
<td>Woroniecka et al.</td>
<td>2011</td>
<td>Top-down</td>
<td>Broad genomics-trancriptomics study identifying systemic differential expression and novel enriched pathways in human diseased glomeruli and tubuli biopsy samples</td>
<td>[26]</td>
</tr>
<tr>
<td>Yagil et al.</td>
<td>2005</td>
<td>Bottom-up</td>
<td>Clinically relevant genomics-trancriptomics study identifying differential expression and novel enriched pathways in hypertension</td>
<td>[27]</td>
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**FIGURE 1:** Systems biology approaches in omics integration. Top-down approaches incorporate omics clinical datasets with complex phenotypic disease signatures and reconstruct protein–protein interactions between transcriptomics and proteomics data in a stepwise manner, as well as combine proteomics and metabolic networks at the pathway integration level. Such ‘whole-system’ analysis overlaps with detailed molecular pathway information from bottom-up approaches, e.g. the Polycystic Kidney Disease study by Song et al. [25], in order to bridge gaps between findings from different kidney-relevant studies for a comprehensive picture of the renal molecular interactome and a robust disease model.
(ADPKD) caused by \textit{PKDI} mutation, and identified several dysregulated genes, including \textit{CDKN1A} and \textit{Fah} [25]. In a large-scale top-down study, He et al. [22] performed a meta-analysis of the glomerular transcriptome and constructed a comprehensive protein–protein interaction network in the glomerulus (GlomNet). This extensive network resembling molecular characteristics in the glomerulus supported the identification of novel molecular pathways and processes crucial to the function, physiology and disease pathology of the kidney filtration system. Through GlomNet, novel dysregulated genes, such as \textit{CDKN1A}, from the Song et al. study (Figure 2; blue arrows) and other proteins have been linked to provide a general ‘whole system’ picture of how these molecules fit together in the renal molecular interactome. Although bottom-up approaches have been widely used to consolidate in-depth literature information, benefiting from highly in-depth and supported published results, these approaches are time consuming and especially challenging to implement in computational models due to variability in experimental conditions in published works [33]. However, bottom-up approaches highly complement top-down omics strategies, so that when combined, they are faster, global and dynamic and capable of tracking perturbations and kinetics in the regulatory network; indeed, the merging of bottom-up and top-down starts with overlapping common findings, such as the overlap of the Song et al. and He et al. studies [33].

Alternatively, as demonstrated in a recent study, a combined top-down and bottom-up systems biology approach can be used in the same study (in the example to elucidate the molecular processes in the pathogenesis of diabetic arteriopathy) [23]. The computational workflow included a proteomics analysis to identify statistically significant proteins (disease case versus healthy control), followed by integration of the statistically relevant information using gene ontology (GO) keywords for the proteins, interactome cluster analysis for grouping and physical linkage and protein mapping to pre-assembled and metabolic and signalling pathways. Furthermore, relevance to human disease pathways encompassing diabetic conditions was established using the Online Mendelian Inheritance in Man (OMIM) disease clustering and literature mining, implicating decreased glycolysis and fatty acid metabolism as key players in the pathogenesis of diabetic arteriopathy. Although this was predominantly a proteomics study, nevertheless the workflow also incorporated literature data and bioinformatics analysis tools to reconstruct a metabonomics network and, as such, is a comprehensive example of top-down and bottom-up omics integration [13, 28].

**TOOLS IN OMICS INTEGRATION**

 Usually omics studies are not integrated across various omics domains, as it is difficult to merge omics platforms and standardize the workflows [34]. To this effect, Mayer et al. [13] have developed a comprehensive conceptual workflow of full omics integration and potential pitfalls, such as the aforementioned limitations. However, despite the difficulties, there is much added value from integrating various omics platforms, and there are numerous bioinformatics resources for this purpose. For example, integrating transcriptomics with proteomics utilizing the Gene Expression Omnibus (GEO) and Proteomics Identifications Database (PRIDE) can provide information at the phenotypic level, which is crucial for multifactorial and otherwise complex disorders [24]. At a more fundamental level, integration of transcriptomics and proteomics enabled elucidating the relationship between mRNA abundance and protein level, which is affected by gene expression, post-translational modifications and protein degradation, among others [35]. Because a simple correlation is inadequate to clarify the relationship between protein expression and protein abundance, protein interaction networks (PINs) have been used to combine transcriptomic and proteomic profiles [24]. There are several bioinformatics tools that help guide the data integration, including the directed graphs (ordered pairs) in the Kyoto Encyclopedia of Genes and Genomes (KEGG) and undirected graphs (simple linkage, no orientation/directionality) in the Online Predicted Human Interaction Database (OP HID), as well as a more comprehensive platform called omicsNET. OmicsNET integrates information on gene and protein correlations from omics experiments using a protein dependency network with pairwise dependencies for currently curated human genes [36].

In a recent study by Perco et al. [24], the authors used all of the above-mentioned tools including omicsNET to examine deregulated features at the mRNA level for three CKD-relevant transcriptomics and proteomics datasets. First, statistically significant protein profiles were established using the Protein ANalysis TThrough Evolutionary Relationships (PANTHER) package that groups molecules into families and subfamilies based on evolutionary functional relationships [37], resulting in non-overlapping functional profiles of 153 transcripts in the ‘protein metabolism and modification’ category and five proteins in the ‘blood circulation and gas exchange’ category. Overlapping functional profiles were found to be in the ‘cell structure’, ‘cell structure and motility’, ‘cell adhesion’ and ‘immunity and defense’ categories. Second, transcriptomic and proteomic profiles were examined with KEGG joint pathway analysis, resulting in the ‘extracellular matrix (ECM)-receptor interaction pathway’, ‘focal adhesion’ and ‘complement and coagulation cascade’ pathways being significantly enriched. Lastly, due to limited coverage of PANTHER and KEGG pathways [13], analysis was also performed with omicsNET [36, 38], which identified ‘blood clotting’, ‘cell structure’, ‘cell adhesion’ and ‘immunity and defense’ as the main functional categories. This integrative method not only elucidated significantly enriched pathways in CKD, but also indicated that increased levels of certain proteins were the effect of kidney damage (e.g. increased permeability of the kidney filtration barrier or reduced re-absorption from tubules) rather than elevated mRNA expression levels. Protein–protein interaction networks may also be capable of deciphering the overlapping functional profiles of amino acid metabolism that was identified in the study by Husi et al. [23] as a molecular feature of diabetic arteriopathy, and in a hypertension genomics-transcriptomics study by Yagil et al. [27]. In both studies, different genes (\textit{Sic2Sa} in Husi et al. [23] and \textit{Fah} in Yagil et al. [27]) were part of the amino
FIGURE 2: KUPNetViz viewer representation of TGF beta transcriptomics datasets in polycystic kidney disease. Gene expression networks for human patients and mouse data were created by querying KUPNetViz for TGFB1, TGFB2, TGFBR1 TGFBR2 and TGFBR3 and using their first-level interaction neighbours. (A) The gene expression associated with TGF beta is mostly up-regulated (red), such as the gene CDKN1A (blue arrow) in patients with PKD according to the study by Song et al. [25] (B and C) KUPNetViz organization of time-dependent data shows the progressive up-regulation, such as the gene CDKN1A that mimics patient data up-regulation over time (blue arrows) of the TGF beta network in a PKD mouse model from the Chen et al. study [32]. Each network depicts a distinct stage of the disease, specifically (B) week 1 and (C) week 3, after mapping the three experimental conditions from the dataset [32]. KUPNetViz visualization can aid in correlating mouse model and human omics data and identifying significant molecules or interactions for further investigation. Red: up-regulation; green: down-regulation.
acid metabolism profile, and the question remains about their common link at the protein level. It is possible that a comprehensive protein–protein interaction network of amino acid metabolism could link the Sic25a1 and Fah gene products. Without omics integration, such causal and functional relationships between the genome, transcriptome and proteome would not be discovered, posing the question, ‘what other key molecular events are not elucidated without full omics integration?’

**PREDICTIVE DISEASE MODELS**

A major goal of the integrative omics strategy is to identify disease biomarkers and suitable therapeutic targets for high-throughput drug discovery [28, 39]. Although omics approaches have proven successful in the efforts to identify CKD-relevant biomarkers [5, 40], the quest for therapeutic targets is still under way [41] and has not yet resulted in major achievements. One reason for this lack of success is the yet nascent status of systems biology–driven predictive and clinically relevant disease modelling pipelines. Integrating all omics that describe specific disease phenotypes and allow for hypothesis testing appears to enable defining the most appropriate therapeutic targets. These efforts are currently progressing but are hindered by a lack of comprehensive data and appropriate bioinformatic solutions [42]. Despite this, several individual omics studies enabled linking biomarker profiles to potential therapeutic targets. Linked disease-specific genes, proteins and other cellular components frequently appeared in co-localized clusters instead of showing random distribution in separate pathways or functional modules [42]. Investigating these ‘local hypotheses’ simplifies the search for therapeutic targets to the affected functional modules and pathways rather than isolated molecules, and also simplifies the *in silico* validation of such targets (expanded in omics in therapeutic target discovery). Therefore, in addition to powerful algorithms capable of integrating and organizing omics data into comprehensive interactome networks, information mining and visualization platforms are crucial for successful drug discovery.

Recently such a visualization platform, called KUPNetViz was implemented in the KUPKB database [43], allowing for simple interactive visualization and exploration of biological network data (Figure 2). The functionalities of this platform include the data mining of protein–protein interactions and association networks across species, the mapping of general association networks to kidney-specific pathways via gene/protein/miRNA expression profiles, the exploration of interactome relationships via kidney anatomy or disease models and finally hypothesis testing and experiment design using data extrapolation across network interactions, anatomies and disease models. KUPNetViz integrates multilayered data across species, interactomes, disease profiles, biological functions and biochemical pathways into a meta-network of so-called super nodes and super edges of agglomerated information. While data agglomeration and extrapolation of multilayered data across species may introduce strong assumptions and bias, weighted against evolutionary conservation and the added value of a comprehensive profile, these risks appear justifiable. Multilayer data organization and visualization is the key, and visualization platforms capable of integrating data into a simple yet comprehensive network representation for hypothesis testing and predictive model generation would be invaluable for biomarkers and therapeutic targets in CKD and many other disorders [11].

**OMICS IN THERAPEUTIC TARGET DISCOVERY**

The strength of an integrated omics approach versus individual omics or classical techniques is the feasibility of building a dynamic multidimensional network of biochemical interactions via the ‘local hypothesis’ that can track specific molecular interactions over time, rather than just a static map of molecular connectivity [39]. For example, while genomics attempts to map phenotypic features to genetic background with genome-wide association studies (GWASs), these only identify single nucleotide polymorphisms (SNPs), but not risk genes [44]. And while GWASs prompted transcriptomics studies to assess the correlation between SNPs and the expression of proximal genes, transcriptomic data on gene regulation does not include information on protein expression level, isoform or post-translational modifications [45]. Proteomics is the next link in the chain, and can fill the gap from transcriptomics and even provide information about protein interactions and degradation (peptidomics). However, it cannot capture the function of a protein in its biochemical pathway [46]. Metabolomics brings the chain of omics full circle, providing spatio-temporal, functional and even phenotypic profiles for proteins influenced by the cellular environment (and all other omics) [16, 47]. Moreover, each omics benefits from longitudinal data collection, and those omics datasets that can be collected using easily accessible samples such as urine produce great amounts of clinically relevant data. Unfortunately, efficient and rapid algorithms capable of handling such huge datasets are still under development.

Current algorithms are capable of combining omics data at the pathway integration level using protein–protein interaction networks, cluster analysis, metabolic networks and other analyses focussed on linking molecular features between different omics traits. The resulting molecular interactome organized into functional pathways could substantially support drug discovery (Figure 3) [48]; it allows for a logical selection of the most likely candidates for drug targets by eliminating general pathways, pathways and processes that have the potential for too many side effects and pathways that failed drug targeting. A good example for such an approach is seen in a study published by Song et al. [25]. The authors identified several potential targets for intervention in ADPKD and consequently investigated their druggability based on a literature search. Among their candidates, they identified CDKN1A, a cyclin-dependent kinase inhibitor protein. This protein was identified in multiple cancer studies and became the focus of many drug discovery projects; however, all clinical trials failed [25]. Hence, this protein was dismissed as a potential drug target in ADPKD. Another candidate, Fah, has been investigated as a drug target in the context of type 1 hereditary tyrosinemia. The Fah gene
product, fumarylacetoacetase, has been successfully targeted, and nitisinone therapy is already in the clinic for the treatment of tyrosinemia \[49\]. Therefore, nitisinone could be repurposed from tyrosinemia to ADPKD or used as a drug lead for further ADPKD drug discovery efforts. When omics integration leads to a truly novel molecular pathway, where not much is known about the molecular mechanisms, an \textit{in silico} drug discovery pipeline can help develop a biophysical profile of the molecular target. If the structure of the protein is unknown, homology models can map sequence information onto available structural templates in order to identify binding pockets or surfaces where drugs can inhibit binding interactions. Subsequently, \textit{in silico} high-throughput screening of small molecule libraries can be performed to find suitable drug-like ligands for testing in cell-free and cellular assays before proceeding to animal studies.

There is a lack of omics-based drug discovery studies in nephrology; however, in a unique effort by Wilmes \textit{et al.} \[14\], proteomics, transcriptomics and metabolomics data from human renal cells were integrated along with a pharmacokinetic model in order to assess the safety of cyclosporine for predictive toxicology. Each omics dataset was assessed individually for statistical significance using software tools including Ingenuity Pathway Analysis (IPA), R statistical package, OpenMS and BRB-ArrayTools. The concentration measurements (endpoints) of cyclosporine were fit into a pharmacokinetic model to simulate the drug’s distribution kinetics in renal cells, as well as integrated into pathway analysis for functional evaluation of toxicity. This approach was not only capable of profiling the key signalling pathways in drug-induced cell stress, but also tracking relevant events independent of gene transcription with an additional dynamic spatio-temporal component from the pharmacokinetic modelling. This study has been further exploited by fitting a time course of the omics markers to elucidate the Nrf2 oxidative response pathway and nephrotoxicity caused by cyclosporine in a time-dependent manner, making it an ideal computational model to evaluate the effects of drugs on relevant pathways \[21\]. In principle, such a model could be adapted to examine nephroprotective, rather than nephrotoxic, drugs in disease-relevant pathways in the quest for ideal therapeutic targets. These two studies \[14, 21\] clearly demonstrate a combined and integrated omics approach produced substantial added value and displays a more complete picture of cellular complexity in biochemical regulatory mechanisms than each omics technology would contribute individually.

**CONCLUSIONS**

With the increasing prevalence and incidence of CKD worldwide, novel biomarkers and drug targets are essential to ease the burden of this deadly disease \[40\]. Drug discovery with omics-driven approaches has the advantage of non-invasive (e.g. urine) data collection from human samples throughout disease progression for directly translatable and clinically relevant disease phenotype modelling and identification of key disease-related molecular signatures. In addition, omics data integration using systems biology approaches, bottom-up integration of published basic science information and top-down clinical omics integration and model generation substantially increases the confidence in the identified biomarkers and drug targets, as statistical significance from comprehensive data overcomes biological variability. This results in robust predictive models capable of mapping disease phenotypes, tracking disease progression, elucidating molecular mechanisms perturbed in disease and a systematic identification of potential therapeutic targets \[50\]. Although systems biology approaches,
specifically computationally tractable algorithms capable of generating truly dynamic spatio-temporal models, are still under development, hypothesis-driven ‘data mining’ of available molecular disease models is crucial for focussing on the specific cellular mechanisms that map back to disease phenotypes and potential therapeutic targets that can be exploited to modify these phenotypes (Figure 3). To this effect, molecular models of disease can be extended to include biophysical profiling of potential therapeutic targets, especially for those molecules with available structural information, whereby in silico screening can be applied to search chemical libraries for small molecule inhibitors, agonists or protein–protein binding disruptors. Such an integrative strategy has the capability to find biomarkers for conclusive, non-invasive diagnosis and pathological staging with CKD prognosis that will aid in testing new therapies. Moreover, it can also be adapted for patient segmentation, identifying at-risk populations on the basis of disease epidemiology and eventually tailoring treatments to patients via personalized medicine [41, 51, 52].

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AUTHORS’ CONTRIBUTIONS

K.C. conceptualized and drafted the manuscript, M.K. and J.K. prepared figures and H.M. provided expert intellectual content; all authors revised and approved the manuscript.

CONFLICT OF INTEREST STATEMENT

H.M. is the founder and co-owner of Mosaiques Diagnostics, Germany, and K.C. is employed by Mosaiques Diagnostics. The content presented in this paper has not been published previously in whole or part, except in abstract form and the authors have no conflict of interest.

REFERENCES

Sustained remission in lupus nephritis: still a hard road ahead

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ABSTRACT

End-stage renal disease caused by lupus nephritis (LN) is an avoidable outcome yet there is considerable uncertainty and variability among nephrologists in their approaches to this disorder. This review discusses recent evidence relevant to the management of LN including recent consensus statements. Long-term results are encouraging compared with 30 years ago, but despite the use of the best available current therapies and achieving high levels of early clinical responses, the kidney often sustains long-term damage and nephritis relapses affect over 50%. Major hurdles to management include the complexity of the clinical presentation, histological features and serological tests, and the absence of reliable outcome predictors or markers of treatment.

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