Chapter

Metabolomics: Special Emphasis on Basic Drug Discovery and Development

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Abstract

Metabolomics utilizes analytical profiling technique for measuring and comparing large numbers of metabolites produced in the biological fluids. Traditional process of drug development is not sufficient enough to understand the proper biochemical processes within the targets which may finally lead to the failure. Metabolomics can be very useful to overcome such failure as it involves in the detailed profiling and understanding of the biochemical processes which helps in identification of target engagement (TE) markers as well as predicting mode of action (MOA). Currently pharmaceutical companies are utilizing this approach in the early drug development stage to combat failure. This chapter will mainly highlight the advantages of this concept over traditional concept of drug development along with recent developments of it.

Keywords: metabolomics, drug, disease, drug discovery and development, analytical approaches

1. Introduction

Pharmaceutical company that wants to thrive in this highly competitive field of introducing novel therapeutic agents for a variety of ailments at an extremely high pace, must devote a significant number of resources to the process of drug discovery. To create newer, more potent, and safer medications, they must make strides and look at different possibilities. The drug development process scenario has seen significant change since the turn of the twenty-first century [1].

From the initial step of target validation through clinical trials to clinical practice, the drug development process is a drawn-out, painful, and incredibly expensive one. The net output of such a method has also been subpar and frequently results from small improvements in currently available therapeutic agents. The current paradigm for drug development asks for the discovery of particular molecular targets that can be used to create highly powerful and selective inhibitors with little off-target activity. These substances may be synthetic small compounds that need medicinal chemistry optimization, or they may be natural products and their synthetic derivatives.

A systems biochemical understanding of the disease, the therapeutic agents' pharmacological properties (i.e., absorption, distribution, metabolism, excretion, and toxicity, or ADMET), and their functional impact on the human body both on-target and off-target would logically be necessary for the entire process, from target discovery to target validation to clinical testing and ultimately clinical adoption.

System biochemistry refers to the "global biochemical networks and molecular regulations". This is a challenging task for drug discovery, development, and its application using conventional methods, as is the case with all systems techniques. The effective and prosperous commercialization of promising therapeutic medicines is fundamentally hampered by the absence of systems biochemical methods and, consequently, functional understanding in the past. A systems biochemical understanding of the human body can now be envisioned for the first time due to the advancements in genomes, functional genomics, proteomics, and now metabolomics. Once created, this will hasten the knowledge of disease mechanisms and the creation of therapeutics at a rate never before seen. Comprehension of the biology of disease requires a thorough understanding of metabolism in human disorders. The metabolome is an essential component of molecular homeostasis regulation. Metabolomics, or the investigation of the metabolome, is currently being utilized for treating a wide range of disorders, mostly through metabolite profiling for biomarker discovery [2].

In recent years, the rapidly developing discipline of metabolomics has taken on a significant role due to its numerous applications in the area of drug discovery and development. Metabolomics has recently made significant strides, and it may now be used as a key tool in the process of finding and developing new drugs.

2. Metabolomics and its evolution

Dr. David Wishart's Human Metabolome Project has completed a year of study on the human metabolome, which contains 2500 metabolites, 1200 medicines, and 3500 dietary components. The $7.5 million research recruited 53 scientists and archived findings on the Human Metabolome Database. The study employs cutting-edge techniques such as NMR spectroscopy, mass spectrometry, multidimensional chromatography, and machine learning to profile metabolites without bias and characterize metabolite interactions using multivariate methodologies [3]. Professor Jeremy Nicholson first put up the idea of metabolomics in 1999. Using a similar principle to genomics and proteomics, metabolomics is a method to quantitatively analyze all metabolites produced by organisms to determine their link to pathological and physiological changes [4, 5].

The substrates and end products of metabolism, known as metabolites, fuel vital cellular processes such as energy production and storage, signal transduction, and apoptosis. Metabolites can come from bacteria, xenobiotics, food, and other foreign sources as well as to being produced naturally by the host organism [6]. Metabolomics is the study of the organism's internal store of non-proteinaceous small molecules. A thorough examination of the entire metabolome (the sum of all the low molecular weight compounds that are present in cells during a specific physiological condition. It alludes to the list of molecules found in a particular organism) under a specific set of circumstances is called metabolomics. Metabolomics is the only technique that can quantify interactions between the genome, proteome, and the biological "wild card" known as the outside environment. The emerging field of genomic science is metabolomics. The metabolomic society correlates with other post-genomic sciences; ideally, metabolomic data sets will be merged with its other omic sciences to provide comprehensive views

into the molecular processes of system biology. However, unlike genomics or proteomics, which concentrate on characterizing huge macromolecules (DNA, RNA, and proteins), metabolomics concentrates on characterizing the small molecule, catabolic, and metabolic products resulting from the interactions of these large molecule [7].

3. Metabolomic study design

Metabolomics experiment involves Experimental design, sample collection and preparation, sample analysis, data processing, and interpretation (**Figure 1**).

3.1 Experimental design

A proper experimental design is crucial for accurate interpretation of data, including sufficient subjects, matching covariates, proper sample collection, and appropriate data analysis techniques [9].

3.1.1 Sample collection and preparation

Metabolic profiling analyses metabolites in both vivo and vitro samples. Metabolomics can analyze various biological materials, including biofluids, cells, tissue, and feces [10]. Standardized procedures for sample collection and storage improve

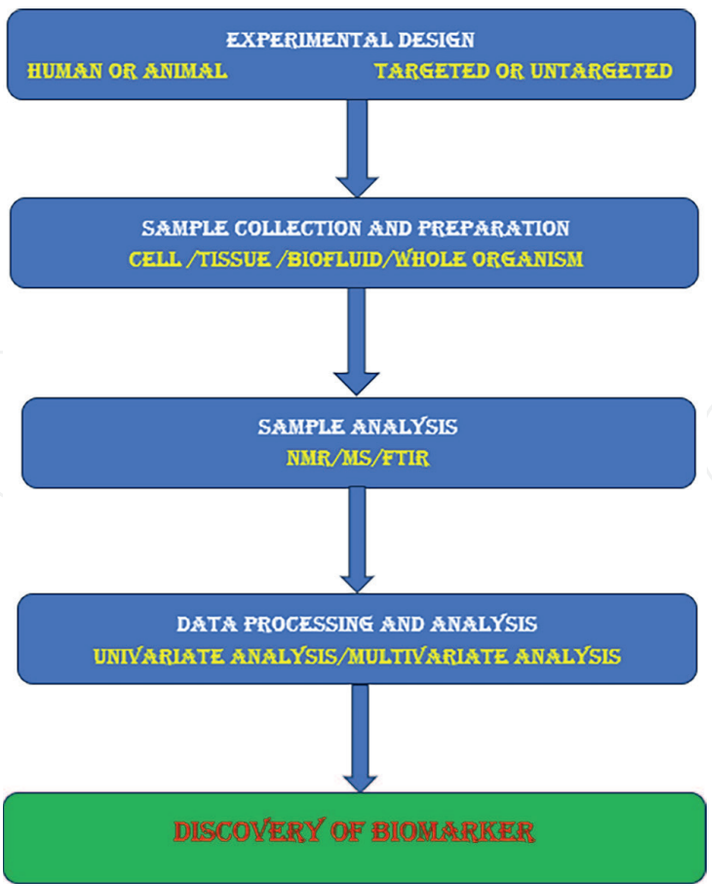

Figure 1.
Metabolomic workflow [8]. **Note:** *NMR (nuclear magnetic resonance), MS (mass spectroscopy), FTIR (Fourier transform infrared spectroscopy).*

quality and reproducibility. Factors like fed vs. fasting state, medications, blood collection time and processing time, addition of additives and potential sample hemolysis considerations can affect metabolites, leading to false-positive or false-negative results. Sample preparation depends on the analytical approaches to be utilized for analysis, such as NMR approaches requiring less preparation as a result it does not affect the sample much whereas mass spectrometry-based approaches requiring sample extraction by using different solvents. Metabolomics studies examine metabolites at specific time points, enabling more dynamic assessments of specific metabolic pathways. Recent development by introducing metabolite with isotope (13C) allows dynamic assessment of metabolic pathways, determining intracellular molecule sources, metabolite fate, flux, and cellular redox balance, providing details information which are not available in case of steady-state metabolomics experiments [10–14].

Sample pretreatment plays an important role. Pretreatment separation modalities include gas chromatography for gas phase separation of molecules useful in the analysis of traces of volatile compounds in samples, high performance liquid chromatography allows high pressure elution that leads to increase in the chromatographic separation of the samples makes it more versatile separation technique, capillary electrophoresis works on the principle of electrokinetic separation useful in the separation of small inorganic ions to larger proteins. These techniques offer advantages in characterizing specific aspects of a metabolome [15].

3.2 Detection methods

Targeted and untargeted metabolomic analyses are the two main categories that can be used, in theory. Targeted analysis would concentrate on a certain number of identified compounds. Untargeted metabolomics, also known as discovery metabolomics, tries to gather all the metabolomic data in a sample, while In the latter, features of relevance are identified after being filtered using various uni and multi-variate statistical techniques following data capture [16].

For the isolation and quantification of metabolome components, a wide range of targeted and untargeted approaches have already been documented in the literature. It was discovered, however, that no one analytical platform is able to collect complete metabolomics data in a single run (**Figure 2**) [17].

Metabolic profiling manly based upon the two specialized analytical techniques viz. NMR spectroscopy and MS. These techniques are efficient enough to identify and quantify wide range of molecules requiring small amount of sample. NMR spectroscopy based on the frequency pattern resulting from the interaction of nuclei of the molecule with the electromagnetic field. This pattern can give the information such as structure of the molecule, its motion, and chemical environment [18]. The identification and measurement of metabolites utilizing NMR techniques, such as proton NMR, 13C NMR, 19F NMR, and 31P NMR spectroscopy, have improved recently. The development of cryoprobes and microprobes, which have decreased the detection limit by a factor of around 3 to 5, is noteworthy. This approach is further enhanced by using two-dimensional total correlation spectroscopy (2D TOCSY) for the confirmation of assigned peaks. Other examples of two-dimensional NMR that have been used to enhance NMR-based data acquisition and metabolite structure analysis include Nuclear Overhauser effect spectroscopy (NOESY), heteronuclear single quantum coherence (HSQC), exchange spectroscopy (ES), and J spectroscopy (JS). These techniques provide better information than one-dimensional NMR, particularly for small-molecule metabolites. Furthermore, better outcome in the metabolome

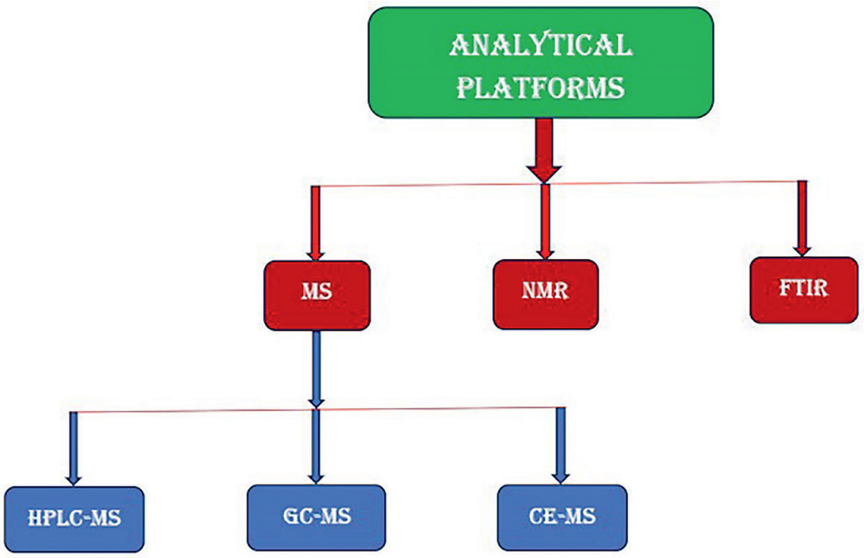

Figure 2.
Major analytical platforms for metabolomic studies in human and animal samples. **Note:** *HPLC (high performance liquid chromatography), GC (gas chromatography), CE (capillary electrophoresis).*

analysis can be achieved by the combination of 2D NMRs such as NOESY and HSQC and TOQCY and HSQC, by combining with MS with NMR or by 3DNMR [8]. NMR spectroscopy is rapid, non-destructive and gives reproducible results which makes its most reliable. Furthermore, samples that have undergone NMR analysis may undergo subsequent MS analysis. This technology's shortcomings are its lack of sensitivity and high user obtaining the necessary tools comes at a significant initial start-up cost and requires special training [19]. In contrast, the destructive analytical method known as mass spectrometry relies on the production of gas phase ions that are then distinguished by their charge/mass ratio. The number of ions for each mass/charge ratio is then calculated once the ions have reached the detector. In order to determine the molecular identities of the constituents, this is processed and compared against accessible mass spectral databases. MS is a very sensitive sample analysis technique that may be applied to both targeted and non-targeted analyses. However, the experimental setup and the instrument parameters have a significant impact on the detection's sensitivity and accuracy [18, 19].

The MS methodology is typically combined with chromatographic methods that have variable degrees of sensitivity, such as liquid chromatography (LC-MS), particularly high-performance liquid chromatography (HPLC- MS), and gas chromatography (GC-MS). Capillary electrophoresis and the MS method can also be combined for better outcome (CE-MS) [20].

A part from NMR and MS, FTIR is another tool which can be utilized successfully for metabolomic study (**Table 1**) [21].

3.3 Data analysis

This phase entails identifying metabolites and figuring out their relative abundance. The platform and method (targeted vs. untargeted) affects how metabolite identification occurs [23].

Standards are typically run in targeted studies and metabolite identification is less uncertain [24]. Whereas metabolite identification in the untargeted technique

Properties	Techniques			
	NMR	HPLC-MS	CE-MS	GC-MS
Sensitivity	–	++	+	+++
Reproducibility	High	Low	Low	Low
Resolution	Low	High	High	High
Quantity of sample requirement	Low	Medium	Medium	Extensive
Cost	Costly	Cost friendly	Cost friendly	Cost friendly
Range of metabolites	Polar and Non-Polar	RPLC: Non-Polar; HILIC: polar	Polar	Volatile polar and non-polar
Metabolite identification	Easy	Difficult (Database need to be improved)	Difficult (Database need to be improved)	Easy (Spectral libraries)

Note: *RPLC: Reverse Phase Liquid Chromatography; HILIC: Hydrophilic Interaction Liquid Chromatography [22].*

Table 1.
Comparison of commonly employed analytical techniques.

is uncertain comparatively, the spectra's are analyzed using either proprietary or publicly accessible software [25].

Additionally, the data should be checked for anomalies and samples or metabolites with a large number of missing values. Samples or metabolites that do not adhere to quality control standards should be eliminated at this stage. These actions result in the acquisition of a set of robustly measured metabolite [26, 27]. Rapid and precise statistical tools are required to handle the complexity and volume of the enormous amount of created data. For data analysis, several metabolomic features may be employed as the input. Spectral bin areas, metabolite concentrations, and spectral peak areas are some of these [28].

Numerous univariate and multivariate statistical methods can be used, focusing on data pre- and post-processing tasks such peak fitting, noise reduction, run order drift correction, and signal extraction/peak recognition. They are collectively referred to as chemometric approaches. The metabolomic characteristics are independently analyzed using univariate techniques [29]. Due to the fact that they use more widely accepted and understood statistical techniques, they are frequently simpler to interpret. However, the presence of interactions between various metabolic features is not taken into account in this approach. Confounding factors like gender, nutrition, or BMI are not taken into account. This raises the likelihood of receiving inaccurate results. Unlike univariate analysis, multivariate analysis takes into account all imputed metabolomic variables and attempts to uncover connections between them. These methods are divided into two categories: supervised methods and unsupervised methods. The most prevalent unsupervised method is principal component analysis, which is capable of detecting data patterns with biological variables. Supervised approaches find patterns within variables of interest while ignoring other sources of variation. Partial least squares regression analysis is the most commonly used supervised statistical procedure [30].

There are a number of software tools available for doing metabolite set enrichment analysis and visualizing the results. Metaboanalyst (www.metaboanalyst.ca)

is a website for metabolomics data analysis, which includes many tools for pathway enrichment analysis [31]. Metlin (https://metlin.scripps.edu), a massive database of metabolites with their MS-derived ions that serve for creative pathway analysis, is an online platform that allows extensive analyses and interpretation of omics data [9]. Metabox (free at http://kwanjeeraw.github.io/metabox/) (retrieved on January 10, 2020) under the GPL-3 license) [32] and MetaboAnalyst (http://www.metaboanalyst.ca/MetaboAnalyst [accessed: January 10, 2020]) [22]. Others such as SECIMTools, Meta XCMS, XCMS, XCMS2, MetAlign, MZmine for MS data processing, and MetDAT for statistical analysis and pathway visualization are among the tools available [8].

4. Metabolomics in the drug discovery and development

Drug research has become increasingly expensive, challenging, and risky over the previous two decades. The costs of research and development have risen, as has the cost of bringing a novel medicine through all stages of testing, which has a direct impact on the frequency with which a new drug is launched to the market. This is because pharmaceutical companies are unable to identify the therapeutic target or because of the multiple targets in case of complicated diseases such as cancer, heart disease, obesity, Alzheimer's disease etc. Another key issue is that the rate of converting a promising lead to a drug is decreasing; just one out of every thirty lead compounds reaches phase-I clinical trials, and only one out of every six drugs crosses clinical trials and enters the market. Still, there is a 5% chance that the drug may be withdrawn from the market due to adverse occurrences. Such failures can be costly for pharmaceutical corporations, affecting drug pricing directly. These failures can be addressed or reduced by strengthening drug target screening, tracking drug toxicity in the preclinical or developmental stage, and monitoring and reporting ADRs during the prescription or physician stage. Metabolomics, an emerging discipline of Omics science, can help to mitigate such failures and accelerate the drug discovery process [33–35]. In recent development ADME studies are commonly employed in drug discovery to optimize the balance of attributes required to turn leads into safe drugs. Metabolite characterization has recently emerged as a key driver in the drug discovery process, assisting in the optimisation of ADME characteristics and increasing drug success rates. For decades, it has been a valuable and important aspect of the drug development process. Over the last decade, there has been an increased effort to solve metabolic concerns using high throughput technologies for screening compounds, which has resulted in a strong need for more quick approaches for metabolite identification [7]. It has been discovered that metabolomics can provide drug researchers and regulators with an efficient and cost-effective method for discovering and developing viable medicinal products (**Figure 3**).

Metabolomics could aid in the following areas of drug discovery and development process:

4.1 Lead compound identification

Metabolomics, natural product chemistry and synthetic medicinal chemistry mainly focusses on the identification and characterization of small molecules as maximum of the FDA approved drugs are small molecules with a molecular weight (≤1500 Da) [37]. It is estimated that majority of these approved drugs are of pre-existing metabolites or preexisting natural product in nature (few examples such as

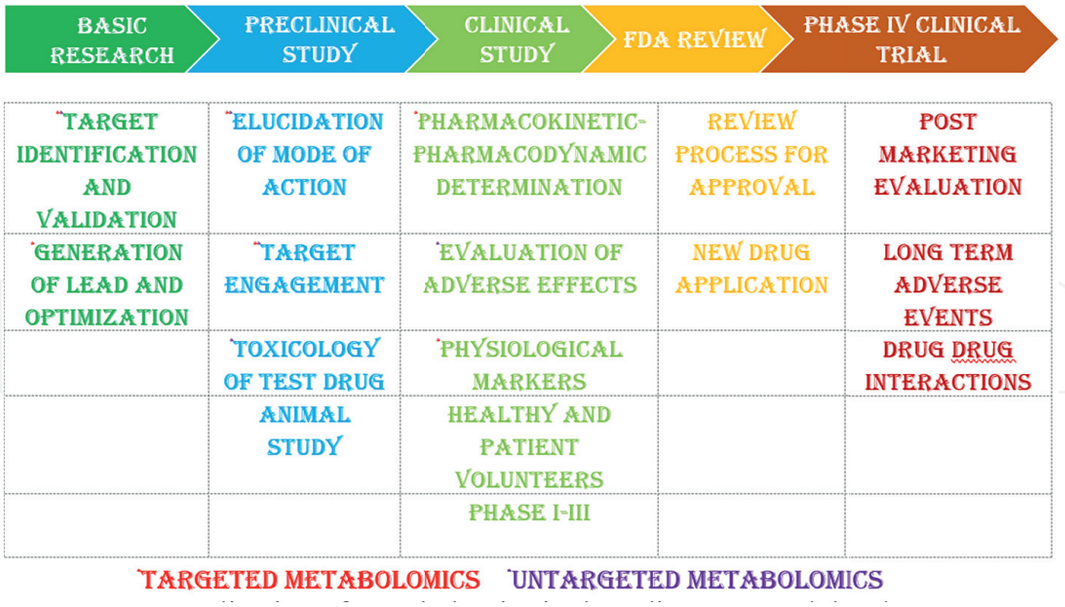

Figure 3.
Application of metabolomics in drug discovery and development [36].

corticosteroids and their derivatives to treat inflammation, ascorbic acid from citrus fruit to treat scurvy, quinine from cinchona tree to treat malaria, paclitaxel from yew bark to treat breast cancer etc.).It has been observed that small molecules are cofactors or substrates for at least one third of human proteins, hence identifying the unexplored ones from plant and human could become an excellent source for new drug candidates or drug scaffolds. Combination of experimental, computational and bibliographic surveys are underway to identify and characterize new or little-known metabolites that plays important roles in cell physiology, enzymatic activity, stress response and disease [38, 39]. Prior metabolite knowledge about known or hypothetical compounds is used in conjunction with NMR or MS data to identify novel compounds which are similar to currently available molecules. More than 20 million NMR and MS libraries of compounds are now available online can have impact on identification of new metabolite [40].

4.2 Lead prioritization and optimization

A number of leads will be generated as a result of any screening programme. One of the most important considerations in the drug development process is deciding which lead to prioritize. Compounds can be prioritized via metabolic profiling based on their capacity to generate the desired biochemical changes. Prioritization is now based on response strength and theoretical considerations of metabolism and toxicity. An inaccurate guess at this time could lead to the failure of the entire programme. A metabolomic analysis can help to distinguish the leads based on their primary and secondary reactions.

In order to progress from a lead to a therapeutic candidate, the lead is employed as a basis structure for the synthesis of hundreds of derivatives in a process known as "lead optimisation." In this step, chemists make several changes to the initial lead and test the impact of the alterations on activity. Each lead is assigned a metabolic profile, and the lead is optimized depending on the profile. This procedure is continued until the final lead candidate with the fewest secondary effects is chosen [41–44].

4.3 Target identification and validation

Identifying bioactive molecule targets is a major difficulty in chemical biological research. Metabolons, a proprietary technology platform, allows researchers to precisely gauge the multitude of metabolic alterations inherent in a specific disease and then map these changes to known pathways, helping them to better understand the disease.to establish a biochemical hypothesis for an ailment quickly. Based on this concept, illness-causing enzymes and proteins can be identified, and druggable disease targets can be located [7]. There are two basic methods for identifying chemical inhibitor targets: direct and indirect. The target proteins bound to the inhibitor are isolated and immediately identified by mass spectrometry in the direct technique. The indirect technique to identifying chemical inhibitor targets is searching for candidates by profiling biological data. If the drug was discovered to disrupt some cellular event for which the regulatory signaling pathway is known, targets can be identified by studying the compound's influence on each stage of the pathway. In some circumstances, omics research (for example, proteomics, transcriptomics, and metabolomics) can aid in the thorough analysis of a compound's effect on a potentially huge number of biological stages. Metabolomic profiling approaches gives the information about the biochemical fingerprint for a specific target. The target can be biochemically validated by identifying any unanticipated adverse effects and by comparing the target to the disease itself. It is also feasible to discover unanticipated secondary effects inherent in a target, so as to discard a target that may pose prohibitive risk [45, 46].

4.4 Biomarker identification

Being able to quantitatively detect, measure, or monitor the disease for which the medicine is being created is crucial to drug development. A medicine to treat a disease cannot be produced until the disease is measured. Many of today's disease assays and diagnostic tests utilize small molecules (i.e., Metabolite) biomarkers as indicators of disease or condition (absence/presence/severity). Metabolomics is primarily concerned with the detection and quantification of small molecule biomarkers (i.e., biomarkers for disease and therapeutic efficacy). Metabolomics has already been used to identify small molecule biomarkers or multi-metabolite signatures for a wide range of diseases such as hypertension, heart failure, Parkinson's disease, prostate cancer, breast cancer, ovarian cancer, schizophrenia, Alzheimer's disease, and coronary heart disease, etc. [47–56]. The most successful application of metabolomics is to discover inborn errors of metabolism (IEMs), with NMR-based metabolomics capable of identifying and monitoring more than 85 different IEMs and MS-based metabolomics capable of detecting and monitoring 130 different IEMs. (For example, phenylketonuria can be diagnosed by low tyrosine levels, Tay Sachs disease by high GM2 ganglioside levels, and cystinuria by high lysine and cystine levels.) [36, 57].

4.5 Mode of action

The justification of MoA using metabolomics usually demands prior information on the impacted metabolic pathways. In other words, drug-induced metabolic changes are statistically analyzed using route maps, and the most significantly affected nodes (proteins) are then selected for additional investigation/validation. This type of analysis can be performed by combining untargeted metabolomics with

in silico or chemoinformatic techniques [58]. It can be utilized to predict not just the mode of action of the drug, but also the harmful mechanism of action [7].

4.6 Measuring drug metabolism

ADME testing is one of the most important aspects of drug development process. The process is very time consuming, expensive and error-prone [59]. The emergence high throughput metabolomic methods has opened up a new avenue for experimentally monitoring ADME and identifying metabolites and metabolic pathways associated with drug metabolism [60]. Methods for identifying drugs or drug metabolites rely more directly on experimental data analysis and spectral comparisons of dosed and un-dosed bio samples. These include mass defect filtering techniques [61] and multivariate data analysis of LC-MS chromatograms for MS and STOCSY for NMR spectroscopy, which can be used in conjunction with high resolution MS instruments such as FTICR (Fourier Transform Ion Cyclotron Resonance) or OrbitrapTM [60] spectrometers to generate exact molecular formulas. Compound identification is clearly only a subset of what is usually necessary for full ADME experiments. It is crucial to measure the amount, distribution, and location of a substance once it has been identified. It is also crucial to compare these results to those of other metabolites over time and space, and to conduct these measurements using additional biological or technical replicas. Fortunately, the high-throughput characteristic of most metabolomics technologies enables these multicomponent, multi-sample analyses to be carried out with great repeatability and at a reasonable cost [61].

4.7 Preclinical study

Metabolomics approaches can be applied rapidly and noninvasively in a number of toxicological assays such as Identification of target organ or region of toxicity, identification of biochemical mechanism of toxicity, identification and quantification of biomarkers profile which measure toxic effects, measurement of time course of toxic effects [62]. This can be employed as a safety screening mechanism by many pharmaceutical companies as an alternative to expensive and time-consuming toxicological and histopathological screening. Furthermore, it can be utilized to find toxicity biomarkers as a result of the identification of various biomarkers utilizing this platform [63].

4.8 Clinical study

Clinical trial monitoring and screening is very important in the drug discovery and development process. Clinical trial monitoring not only involves the effect of drug but also involves the influence of diet, drug use/abuse and behavioral factors on the outcome. Metabolomics can be can be useful in-patient monitoring by detecting the presence of over-the-counter drugs, herbal supplements, drugs of abuse and food by products in clinical trials settings to assure patients compliance [64–67]. This method be useful in the patient screening and drug patient matching. This approach can be utilized to examine drug metabolism characteristics in individual patients. There is significant diversity in patient responses to specific classes of medicines (such as antidepressants), which is largely owing to differences in drug metabolism profiles [68]. Certainly, detecting susceptible/refractory patients prior to recruitment (in clinical trials) or prescription (in clinical practice) would improve outcomes. The use of metabolomic-based phenotyping in clinical trials or drug dosage in clinical practise

has the potential to be a quick and low-cost screening method for patient selection or drug dosing. These metabolomic assays would almost certainly be far less expensive in terms of both time and money than traditional approaches. Metabolomics is extremely effective in monitoring drug doses of relatively toxic pharmaceuticals (anticancer drugs such as methotrexate, 6-mercaptopurine, 5-flurouracil, etc., blood thinning agents such as warfarin, immunosuppressants such as mycophenolic acid and ciclosporin). Furthermore, it plays a critical role in detecting and monitoring ADRs, which are a huge burden for the pharmaceutical business and healthcare system (due to patient death and hospitalization). Endogenous metabolite levels in the blood and urine can be used to detect many undesirable medication effects [61].

5. Challenges for metabolomics

Data analysis is one of the most difficult aspects of metabolomics research. Metabolomics provides enormous volumes of data, often with complicated structures and patterns that necessitate the use of advanced computer techniques to analyze.

A second difficulty is sample deterioration and standardization. Metabolomics is a young discipline with few widely agreed standards for sample collection, processing, and analysis. Importantly, after a sample is extracted, its metabolic signature may differ from what it was in the biological system. This emphasizes the importance of sample preparation and makes comparing results between research and replicating trials challenging.

Another issue is the existing metabolomics techniques' lack of sensitivity and specificity. Despite substantial technological breakthroughs, contemporary metabolomics techniques are still incapable of detecting all metabolites in a given sample. This can result in false negatives or incorrect results [69].

6. Conclusion

Metabolomic principles have the potential to revolutionize the drug discovery and development process. Metabolomics focuses on the small molecules that are essential for life to exist and act as the molecular foundation for cells by supplying fuel for cellular processes. Small molecules also help to preserve cellular integrity, fights environmental stress, and acts as a cellular messenger for a variety of cellular activities. Drug discovery is a time-consuming, high-risk, and tremendously expensive procedure. Metabolic profiling is a sensitive indicator of phenotypic alterations as well as pharmacological on and off target of drug candidates. High-resolution metabolic profiling is possible using minimal sample preparation, and as part of various drug testing processes. Metabolomics can also offer drug selection markers that can be employed *in vivo* which are phenotypic specific.

Metabolomics allows scientists to investigate the known roles of small molecule metabolites in greater detail and with better precision than ever before. The increasing breadth of available coverage, increased sensitivity, greatly improved analytical software, and trends towards more accurate quantification enable certain types of novel metabolomics experiments that were only a dream a few years ago. Continuing development in the field metabolomics along the growing investment by the pharmaceutical industry players and FDA will foster a rapid and more cost-effective drug discovery and development process.

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Conflict of interest

The authors declare no conflict of interest.

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