Review Article

Novel insights of toxicological evaluation of herbal medicine: Human based toxicological assays

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Abstract

Herbal medicine is still the mainstay of about 80% of the world population for their primary healthcare. Recently, herbal medicine is being accepted as a promising therapeutic modality against many chronic diseases where western medicine perceived to be less successful. Clearly, majority of herbal remedies are considered as safe for consumption, however, there are concerns about their safety. Hence, toxicological evaluation is imperative to reduce the risk associated with herbal products and to confirm their safety and effectiveness. To date, research with experimental animals is considered as a gold standard in toxicology testing, nonetheless, *in vivo* animal tests are constrained by time, ethical considerations, experimental cost and lack of sufficient congruence between the animal and human physiologies. With the advent of science and technology several novel human-based toxicological models have been introduced. The current review briefly describes the human based toxicity assays introduced as alternatives to *in vivo* assays, under the categories of cell-based cytotoxicity assays, organ cultures and bioengineered organs on chips, molecular biological models (toxicogenomics and next generation sequencing) and *in silico* models. Particularly, the promises, limitations and prospects of these assays with respect to herbal drug toxicity are discussed herein.

Keywords: Herbal medicine, toxicity, cytotoxicity, organs on chip, toxicogenomics, next generation sequencing, *In silico*

Introduction

Herbal medicine is still the mainstay of about 80% of the world population for their primary healthcare (Bent, 2008). Plant based medicines are well accepted as therapeutic agents for emerging diseases such as diabetes, arthritis, liver diseases, cardiovascular diseases due to their multi-targeted synergistic mode of action (Jayasinghe et al., 2015). Particularly, herbal medicine provides a safer alternative to synthetic western medicine (Bent, 2008). Thus, the prospect for efficient and less toxic herbal drug combinations is enticing.

Traditional health care systems embrace practices, approaches, knowledge and beliefs conveyed over generations and generally considered as safe remedies (Ifeoma and Oluwakanyinsola, 2013). The botanical wisdom accumulated by the indigenous people led to the development of traditional systems of herbal medicines. The great civilizations such as ancient China, India, and Africa provided the written documents of utilization of herbal products. Subsequently, Ayurveda, Unani, Kampo, and traditional Chinese medicine (TCM) have being flourished as systems of herbal medicines (Petrovska, 2012).

Despite the nascent demand for traditional herbal medicines, there are still concerns about their safety after being subjected to suspicions of toxicity (Datta-Mitra and Ahmed, 2015). Toxicities related to herbal products mainly categorized into intrinsic and extrinsic effects (Ifeoma and Oluwakanyinsola, 2013). The intrinsic toxicity mainly resulted from the innate active compounds in the herbal preparation (Ifeoma and Oluwakanyinsola, 2013). Moreover, improper dosage and interaction of herbal drugs with other orthodox drugs also evident as an intrinsic toxic effect (Ifeoma and Oluwakanyinsola, 2013). Herb related toxicity may also result from foreign substance present in

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the herbal preparation such as metal contaminations, microbial products and misidentification of plant species. These factors may lead to adverse reactions at clinical stage of herbal medicine (Chan, 2003). Hence, there is a strong impetus for toxicological evaluation of herbal preparation.

To date, research with experimental animals is considered as a gold standard in toxicology testing, as the whole animal is usually closely correlated to human toxicity as the system incorporates pharmacokinetic such as absorption, distribution and metabolism (Parasuraman, 2011). Animal toxicological models were introduced in 1920 by J. W. Trevan and proposed to use 50% mortality of animals to determine the lethal dose (LD_{50}) of individual chemical (Parasuraman, 2011). However, these models required large number of animals and the experiments are time consuming. In addition to animal ethics considerations, another drawback of using animal is the lack of sufficient congruence between the animal and human physiologies (Andersen and Krewski, 2009).

To minimize the use of laboratory animals for toxicological evaluation international level polices were also put forwarded. The U.S. National Research Council (NRC) in 2007 released a report titled "Toxicity Testing in the 21st Century: with a vision and a strategy," to limit the animal-based toxicology tests and encouraged adopting human based alternatives for toxicological assessments (Andersen and Krewski, 2009). These alternative strategies broadly categorized into cellular, molecular and computational methods (Ifeoma and Oluwakanyinsola, 2013). Thus far, these methods were restricted to the toxicological evaluation of xenobiotic, however increasingly been applied to the toxicity evaluation of herbal medicine.

Unlike synthetic drugs, herbal medicine has been consumed by humans over centuries and indications on toxicological impact on human have been partly accrued. In this regard, human based toxicological analysis would appropriately extrapolate the toxicological impact of herbal medicine rather than the animal model. Moreover, adopting human based toxicological insights will reduce the number of animals, costs and time of experiment (Ifeoma and Oluwakanyinsola, 2013). Though, these techniques are gaining momentum in the field of toxicology, there are still many challengers.

The current review aims at provide a brief introduction to novel human based toxicological assays could be incorporated to toxicity determination of herbal remedies. These methods are broadly categorized into cellular, molecular biological and *in silico*/computations methods and their advantages and limitations are discussed along with their prospects.

Toxicity of herbal medicines

Toxicity related to herbal products are mainly categorized as intrinsic and extrinsic toxic effects, that are evident in herbal preparation (Ifeoma and Oluwakanyinsola, 2013).

Intrinsic toxicity

Intrinsic toxicity is related to inherent properties of herbal preparation such as toxicity due to active constituents, over dosage and drug interaction (Drew and Myers, 1997). Plants synthesize a plethora of metabolites characterized as 'phytoprotectants" which could be harmful for vertebrates due to the conserved biological nature among the animal kingdom (Ifeoma and Oluwakanyinsola, 2013).

Particularly, phytochemicals like alkaloids, flavonoids, terpenoids and saponins are implicated in the development of some toxic effects in human (Ifeoma and Oluwakanyinsola, 2013). Alkaloids behave as agonistic or antagonistic of neurotransmitter systems and may interfere with mammalian nerve system (Ifeoma and Oluwakanyinsola, 2013). Similarly, some lipid soluble terpenes have shown inhibitory properties against mammalian cholinesterase (Kennedy and Wightman, 2011). Saponins are potent surfactants that can affect lipid-rich cellular membranes of human erythrocytes and lead to hemolytic activity (Ifeoma and Oluwakanyinsola, 2013).

Over dosage is the commonest cause for intrinsic toxicity effects of herbal medicines. Thus, adopting an appropriate dosage may minimize the adverse effects of most phytochemicals present in a preparation. Usually, toxic substance follows a hermetic dose response: a biphasic model characterized by a low-dose stimulation and a high-dose inhibition or cytotoxicity (Calabrese and Baldwin, 2000). Thus, precise calculation of dosage is important in minimizing the toxic effect entail in herbal preparation. Over dosage of certain herbal products such as *Mahashankha Vati* prescribed in Ayurveda is known to interact with other drugs (Panda and Debnath, 2010).

Interactions between herbal medicines and prescribed drugs can occur when they are concurrently present in the body and may lead to serious health consequences (Hu et al., 2005) Both pharmacokinetic and/or pharmacodynamic modifications can alter the drug interaction in the body and may manifest toxicological effects (Hu et al., 2005). Some herbs, notably St. John's Wort (*Hypericum perforatum*), ginkgo (*Ginkgo biloba*), ginseng (*Panax ginseng*), kava (*Piper methysticum*) and garlic (*Allium sativum*) reportedly showed significant interaction with some co-administered drugs by modulation of Cytochrome P450 (Ernst, 2002).

Extrinsic toxicity

Herb related toxic effect also has resulted from foreign substance present in the herbal preparation such as metal contaminations, microbial products, etc. (Ifeoma and

Table 1. Toxicological evaluation of different herbal plant/extracts/formula tested on human cell lines

Cell lines	Name	Outcome	References
human adenocarcinoma cells of the cervix (HeLa), human breast cells (MCF-12A)	Root of *Antidesma venosum* (tassel-berry) Bark of *Bridelia micrantha* (coastal golden leaf)	The IC_{50} was not reached at the concentrations tested (0.1 µg/ml – 1 mg/ml)	Steenkamp et al., 2009)
human proximal tubule HK-2 cells	*Calea zacatechichi* (Dream herb)	potentially nephrotoxic	(Mossoba et al., 2016)
human cervical cancer (HeLa) cell line	*Fruit of Solanum Nigrum* (black night shade)	IC_{50} 847.8 – SRB assay IC_{50} 265.0- MTT assay	(Patel et al., 2009)
HeLa and MCF-7 breast cancer cell lines	Las 01 Herbomineral preparation	at higher concentrations (500 mg/L) there was higher effect in toxicity so that in only 20% and 18% in MCF-7 and HeLa, respectively,	(Sheikh et al., 2012)
HepG2 cell line	*Trigonella foenum-graecum* (fenugreek), *Atriplex halimus* (salt bush), *Olea europaea* (olive), *Urtica dioica* (nettle), *Allium sativum* (garlic), *Allium cepa* (onion), *Nigella sativa* (black seed), and *Cinnamomon cassia* (cinnamon)	*Cinnamomon cassia* is cytotoxic at concentrations higher than 100 µg/mL others are cytotoxic at higher 500 µg/mL	(Kadan et al., 2013)
HepG2 cell line	*Pinus kesiya* *Glochidion daltonii*, *Cladogynos orientalis*, *Acorus tatarinowii* and *Amomum villosum*	The extract of *Pinus kesiya* ; IC_{50} value of 52.0 ± 5.8 µg/ml, Extract of *Catimbium speciosum* IC_{50} 55.7 ± 8.1 µg/ml. *Glochidion daltonii*, *Cladogynos orientalis*, *Acorus tatarinowii* and *Amomum villosum* IC_{50}; ranging 100-500 µg/ml	(Machana et al., 2011)

Oluwakanyinsola, 2013). Heavy metals such as lead, cadmium, arsenic and mercury are frequently found as contamination of herbal preparations (Gair, 2008).

Particularly, contamination of lead and mercury can cause serious neurological impairments Ifeoma and Oluwakanyinsola, 2013). In Ayurveda medicines, certain heavy metals such as lead, arsenic and mercury are incorporated into primary herbal formulations of *Bhasma* as adjuvant (Kumar and Gupta, 2012). In ancient preparations, heavy metals are "purified-out" through multiple neutralizing systems and by addition of specific mineral herbs the toxic effects of the metals are minimized (Gair, 2008). However, recent evidences from various countries imply that most of the current herbal formulations contains higher levels of toxic heavy metals than recommended in traditional pharmacopeias (Ernst, 2002). For example; excessive contamination of traditional formulations with heavy metals in Singapore was reported by Koh and Woo, 2000. Another parallel study established contamination of nine heavy metals including cadmium, cobalt, copper, iron, manganese, nickel, lead, zinc, and mercury were in 42 Chinese herbal medicinal plants (Wong et al., 1993).

Misidentification of medicinal plants may also result adverse reactions. Common Gentian *(Gentiana luteum L.,* Gentianaceae) Skullcap (*Scutellaria lateriflora* L., Lamiaceae) Chinese star anise *(Illicium verum Hook.* f.) are some of the plants which are often being misidentified (Jordan et al., 2009).

Experimental evidence for herbal medicine related toxicities

Although, traditional medicines are largely considered as safe, there have been numerous reports of significant adverse effects associated with herbal remedies. It is assumed that the low incidence of toxicity of herbal medicine is partially due to consumers believes on relative safety of herbal products (Jordan et al., 2009). A survey conducted in United Kingdom revealed 30 % of people consumed both conventional and herbal drugs have shown adverse reaction (Jordan et al., 2009).

In 2002, according to the poison control centres (PCC) total 23,000 cases of toxic exposures to dietary supplements, herbs and homeopathic products have been reported (Watson et al., 2003) and over the years number of cases has been steadily increasing (Woolf et al., 2005).

Many of the herbal medicines have the potential to cause liver injury. Herbal medicine-related hepatotoxicity

Table 2. Application of *in silico* approach for prediction of toxicity of phytochemicals

Model	Herbal plants	Outcome	References
QSAR to study the cytotoxic activity	37 sesquiterpene lactones	several specific structural elements and skeletal types are required for the greatest cytotoxic activity	(Scotti et al., 2007)
Artificial neural network	55 sesquiterpene lactones	The cytotoxic activity was accurately predicted in 89% of the test chemical	(Fernandes et al., 2008)

represents the second most common cause of drug-induced liver injury (DILI) in Western countries. In the United States, between 2004 and 2013, among 839 patients who had suffered DILI, 130 has being reported to be associated with consumption of herbal dietary supplement (Navarro et al., 2014). In Europe, a survey conducted during performed between 1994 and 2004, reported that 9% of 461 cases of DILI were caused by intake of medicinal herbs (Andrade et al., 2005).

Renal toxicity is another common toxicological manifestation of herbal medicine. The mostly renal toxicity is caused by medicinal herbs containing aristolochic acid (AA) nephropathy a plant alkaloid, which is nephrotoxic (Asif, 2012). Especially certain plants like borage (*Borago officinalis*), comfrey (*Symphytum s*pp.), coltsfoot (*Tussilago farfara*) and life root (*Seneci oaureus*), sassafras (*Sassafras albidum*) and germander (*Teucrium chamaedrys*) are advisable to avoid in dialysis patients due to presence of nephrotoxic compounds (Asif, 2012).

In addition, some medicinal plants could possess cyto and genotoxic effects. Plants such as *Chenopodium ambrosioides* (Gadano et al., 2002) *Inula. Viscosa* (Aşkin Celik and Aslantürk 2010), *Azadirachta indica (A. Juss), Morinda lucida (Benth.), Cymbopogon citratus (DC Stapf.), Mangifera indica (Linn.)* and *Carica papaya* (Linn.) hence exhibited mitodepressive effects on cell division and induced mitotic spindle disturbance in *Allium cepa* bioassay (Gadano et al., 2002).

Hence, it is imperative to conduct a proper toxicological evaluation of herbal products prior to their clinical applications.

Introduction of human based toxicological assays for herbal drug toxicity evaluation

Toxicological assessment is paramount in herbal medicine to identify adverse effects and dosage determination to safeguard from possible adverse effects (Ifeoma and Oluwakanyinsola, 2013). Evaluation of toxicological impacts of herbal medicine at pre -clinical and clinical stages facilitates the identification of toxicants which can be discarded or modified into safer alternative (Kennedy and Wightman, 2011).

Generally wide range of toxicity tests are done in non-human experimental models prior to their clinical applications of any drug. Thus far, the animal models were considered as gold standard in toxicology testing as the whole animal is usually closely correlated to human toxicity as the system incorporates pharmacokinetic (absorption, distribution, metabolism) (Andersen and Krewski, 2009). Test organisms used in toxicity testing range from simple systems like brine shrimp to other animals like mice, rats, guinea pigs and rabbits (Andersen and Krewski, 2009). However, animal experiments are time consuming, and more restricted by animal ethics and rights laws (Doke and Dhawale, 2015).

Ethical consideration in animals in research gave rise to the adoption of 3 R's principals and arose the need to reduce the number of animals, refine the tests methods used to minimize pain and suffering of experimental animals, and replace animal tests with validated alternatives employing human cells where possible (Doke and Dhawale, 2015).

There are several limitations foreseen in predicting the human toxicity. In animal studies, usually high doses of compounds are used for toxicological assessment and those levels are higher than human exposure levels Moreover, for *in vivo* studies standard laboratory animals of single strain are used and which cannot accurately predict the variability in responses seen in the human population (Haller et al., 2002).

With the advent of cellular and molecular biology, novel human based toxicological assessment methods have been introduced as alternatives to laboratory animal model. These novel methods are categorized into cellular, molecular and *in silico* methods as represent in figure 1.

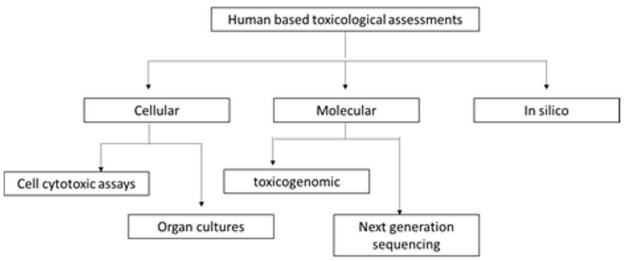

Figure 1. Classification of novel human based toxicological assessment methods

Cell based toxicological assays: cytotoxicity testing

Cell based assays are indispensable tools in toxicological

evaluation and provide insight towards the carcinogenic and genotoxic dispositions of herb products (Ifeoma and Oluwakanyinsola, 2013). Several end points such as inhibition of cell proliferation, decrease of cell viability, damage to membrane integrity, effects on morphologic and intracellular differentiation are assessed for toxicological determination (Ifeoma and Oluwakanyinsola, 2013). Both primary cell cultures and modified cell lines are used for this purpose. Though, primary cells are more similar to those of the original tissue; obtaining reproducible results is challenging. Conversely, cell lines are homogeneous and standardized than primary cultures; however, their metabolism is different from normal cells (Bourdeau et al., 1990). The more commonly used cell lines for toxicological assessment includes diploid human fibroblast lines (e.g. WI-38) and tumor cell lines (e.g. HeLa) (Bourdeau et al., 1990). Table 1 summarizes the toxicological evaluation of herbal preparation tested on human cell lines.

The renewed interest of cell-based assays in toxicology is largely due to the current advances in sensitive detection, automated fluid handling and imaging, which enable simultaneously quantitative and efficient analysis of different mechanisms involved in cytotoxicity (Bourdeau et al., 1990).

Introduction of cell culture techniques has greatly reduced the number of animals being used for toxicity evaluation and enabled to understand the molecular mechanisms underlying the impact. Importantly these assays can be altered for high-throughput screening of herbal preparation (Ifeoma and Oluwakanyinsola, 2013).

Apart of these advantages cell cultures tend to exhibit problems in obtaining large cell populations in primary cell cultures and some cell lines are not very stable during the culture process) (Bourdeau et al., 1990). Moreover, for more precise evaluation, toxicity testing on multiple cell types is encouraged as single cell type poorly resembles the whole organism (Ifeoma and Oluwakanyinsola, 2013). In such instances, stem cells of human origin are proposed as effective candidate owing to their ability to be differentiated into different cell types (Udalamaththa et al., 2016). Particularly, human embryonic stem cells (hESCs) provides valuable insight for the developmental toxicities.

However, cytotoxicity assays provide limited information about toxicokinetics of tested compound (Anon, 2015) while expedition for developing a sophisticated *in vitro* system which mimics *in vivo* condition continues.

Human organ cultures and bioengineered organ on a chip

Establishment of *in vitro* systems capable of mimicking the functionality of specific human organs is currently a pivotal point of toxicological research. Recently, the single cell type culture has been extended to co-culturing of multiple cell types or part of organ or cell aggregates (Anon, 2015) and these are termed as organotypic models (Anon, 2015). Thus far, organotypic models have been developed for the skin, eye, lung, liver, and central nervous system (Anon, 2015). Organotypic models have gained credibility in toxicological research due to their close anatomical resemblance to whole organs and ability of evaluating of metabolism, biodistribution of toxic compounds in an *in vitro* system (Anon, 2015). However, application of these models for high-throughput testing is constrained by difficulties in efficient *in vitro* culturing of tissues/organs (Oleaga et al., 2016).

Simultaneously, a novel approach known as organ-on-a-chip model was developed with the ability to simulate the cellular physiology in an artificial environment (Anon, 2015). These models are micro engineered biomimetic system consist of transparent 3D polymeric micro channels aligned by human cells (Anon, 2015).

This is a sensitive, reproducible and robust technique which has the potential to be developed as high throughput toxicity screening tool in herbal drug industry. However, integrating multiple organ chips in a physiologically relevant way that is more similar to whole human physiology remains as a huge challenge (Oleaga et al., 2016).

Molecular biological methods for toxicity evaluation

Since DNA was first sequenced in 1997 molecular studies have undergone rapid developments and now its applications have been expanded up to toxicity prediction of chemical compounds or herbal drugs. Generally, it is considered DNA profiles are more efficient in prediction of geno-toxicity than phenotypic or metabolic profiles of cell cultures (Ifeoma and Oluwakanyinsola, 2013).

Moreover, in herbal medicine DNA based technique can be used to identify foreign materials in herbal preparation which is potentially difficult to determine by macro and microscopic methods (Ifeoma and Oluwakanyinsola, 2013). Moreover, molecular based tools preferable as high throughput screening tools of herbal drug toxicity. Toxicogenomics and next generation sequencing technology are strong predictive tools of toxicology of various compounds including herbal drugs (Anon, 2015).

Toxicogenomics

Toxicogenomic is a combination of genomics, proteomics, metabolomics, and bioinformatics and used to gain a molecular level understanding of toxicity of compounds including herbal medicine (Hamadeh et al., 2002). This concept was introduced in 1999 and has become a robust area in toxicological field ever since (Hamadeh et al., 2002).

In toxicogenomic model, toxicants induce genome

expression and proteomics are used as screening criteria toxicity screening. Genome-wide analysis of toxicant-induced expression profiles may provide a means for prediction of toxicity prior to classical toxicological endpoints (Pennie et al., 2000).

Toxicological effects of a chemical compound can be predicted by the gene expression changes associated with signal pathway activation (Suter et al., 2004). Access to a relatively large toxicogenomics database containing gene expression data of herbal products helps to classify compounds early in the drug development and consequently save animals, time, and money in pre-clinical toxicity studies (Suter et al., 2004).

Apart from the advantages, toxicogenomic tool cannot address all aspects of toxicology, hence a combinatorial approach is required. In addition, sophisticated equipment and expertise are required to evaluate probable health outcome of compounds including herbal compounds (Suter et al., 2004).

Next generation sequencing technology

Next generation sequencing (NGS) technology is another advance molecular biological tool uses for toxicity prediction of compounds including herbal drugs (Ifeoma and Oluwakanyinsola, 2013). Next Generation sequencing basically refers to non-sanger based high throughput DNA sequencing technology which sequences millions of DNA strands in parallel (Behjati and Tarpey, 2013).

NGS has advantages over sanger methods due to ability of detecting very small numbers of DNA at varying degree of degradation. Hence, NGS is particularly important in detection contamination of herbal products (Byard et al., 2015).

In order to increase efficiency of toxicity prediction using NGS, databases of genetic biomarkers of toxicity of herbal medicines need to be enriched (Byard et al., 2015). This can be done by creating genomic signatures of identified phytochemicals which can serve as data library for herbals (Ivanova et al., 2016).

NGS can be employed as effective and cost-efficient way to authenticate highly processed Traditional Chinese Medicine (TCM) and Ayurveda medicine and to monitor their compliance with legal codes and safety regulations (Ivanova et al., 2016).

Herbal supplements representing three different producers from five medicinal plants: *Echinacea purpurea*, *Valeriana officinalis*, *Ginkgo biloba*, *Hypericum perforatum* and *Trigonella foenum-graecum* has been authenticated using NGS (Ivanova et al., 2016). It has revealed a diverse community of fungi, known to be associated with live plant material and/or the fermentation process used in the production of plant extracts. Hence, NGS is recommended as a promising method for herbal plant authentication (Ivanova et al., 2016).

Computational or *in silico* models

In silico toxicology assessments aim to complement existing toxicity tests with the use of computational methods along with molecular biological techniques to toxicity of compounds (Raunio, 2011).

In silico toxicology incorporates a wide array of computational tools (A) databases for storing compounds and their toxicity, and chemical properties; (B) generating molecular descriptors; (C) simulation tools for systems biology and molecular dynamics; (D) modeling methods for toxicity prediction; (E) expert systems that include pre-built models in web servers or standalone applications for predicting toxicity; and (G) visualization tools (Raies and Bajic, 2016).

These methods can predict properties relevant to physiological properties such as physico-chemical, gastrointestinal permeability, blood–brain barrier permeability, binding to plasma proteins, affinity for transporter proteins, metabolic clearance, potential to inhibit or induce drug metabolizing enzymes and generation of reactive metabolites (Raies and Bajic, 2016).

In silico models are less expensive, rapid, and reproducible thus enables high through put screening of herbal products. Moreover, provide complete alternatives for laboratory animals. However, sometimes these applications are constrained by complicated modelling systems and difficulties in interpreting data (Raies and Bajic, 2016).

Table 2 presents two studies which have used *in silico* approach to predict the toxicity of sesquiterpens of natural origin.

Usefulness of the toxicological analysis in regulation of herbal medicine

With the emergence of various toxic effects, there is an urgent need for the regulation of herbal products. More than 70% of herbal drugs are purchased as over-the-counter (OTC) dietary supplements without proper prescription or guidance from medical practitioner (Panda and Debnath, 2010). Less than 10% of herbal products in the global market are standardized or quality controlled (Tarkan et al., 2016). Hence, regulation of herbal medicines is essential to ensure the safety, efficacy and quality of herbal medicinal products.

Herbal drugs are regulated by the Food and Drug Authority under the Dietary Supplement Health and Education Act (DSHEA) (Abdel-Rahman et al., 2011). The regulatory framework for herbal drugs includes establishing current good manufacturing procedures, mechanisms for pre-market safety notifications for new ingredients, and a mechanism for establishing claims used in product labeling. Most importantly, the FDA is responsible for

overseeing the safety of herbal drugs (Abdel-Rahman et al., 2011).

Hence, establishment of comprehensive toxicological analysis for evaluation of herbal medicine is timely requirement. Specifically, integration of novel human based assays for toxicological evaluation may undoubtedly bring about significant advances in predicting toxicological impacts of herbal medicine. Moreover, most of the assays described herein are rapid and can easily be adapted to high throughput screening with limited cost and labour.

Conclusion

The NRC report foresees a future in which all toxicity testing would be conducted in human based methods which eliminate the use of animals. Integration of novel human based innovative such as cell based, molecular biological and computations models undoubtedly bring about significance advances in toxicity predication of xenobiotic, synthetic and herbal drugs while minimizing the use of animals. Mostly, these techniques have been implemented in toxicity prediction of xenobiotic and apparently limited in herbal drug toxicity testing. However, it is anticipated that in future these novel humans based toxicological tools will play a major role in herbal drug toxicity prediction specially in nutraceutical industries.

Conflicts of Interest

There are no conflicts of interest

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